# Package 'respirometry'

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<b>Description</b> Provides tools to enable the researcher to more precisely conduct respirometry experiments. Strong emphasis is on aquatic respirometry. Tool focus on helping the researcher setup and conduct experiments. Functions for analysis of resulting respirometry data are also provided. This package provides tools for intermittent, flow-through, and closed respirometry techniques.	
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Contents	
adj_by_temp         calc_alpha         calc_b         calc_E         calc_MO2         calc_pcrit	

2 adj\_by\_temp

	closed	13
	co2_add	14
	co2_flush	16
	co2_rate	17
	conv_nh4	19
	conv_o2	20
	conv_resp_unit	21
	correct_bubble	23
	flush_carb	24
	flush_o2	25
	flush_water	26
	goal_flush_pH	27
	guess_TA	28
	guess_when	30
	import_firesting	31
	import_presens	33
	import_pyroscience_workbench	
	import_witrox	38
	make_bins	39
	max_MO2	41
	mean_pH	42
	min_flow	43
	peri_pump	45
	plot_pcrit	46
	predict_nh3	49
	predict_pH	50
	Q10	52
	respirometry	
	RQ	
	scale_MO2	55
Index		58

 $adj_by_temp$ 

Predict biological parameters at a new temperature

# Description

Predicts the values of any inputted biological parameter (e.g. MO2, Pcrit) at a new temperature based on empirical measurements at a range of temperatures. Data can be fit to a temperature-dependence curve using either Q10 or calc\_E. By default, the predicted values are also plotted alongside the inputted data to allow the user to assess the quality of the fit.

adj\_by\_temp 3

# Usage

```
adj_by_temp(
  meas_temp,
  meas_x,
  temp_new,
  method = "Q10",
  Q10,
  E,
  show_coef = FALSE,
  plot_fit = TRUE
)
```

# Arguments

meas_temp	a vector of temperature values ( $^{\circ}$ C) corresponding to meas_x.
meas_x	a vector of biological values (e.g. MO2, Pcrit) corresponding to meas_temp. These are typically empirically derived.
temp_new	a vector of temperature values (°C) at which new values of "x" should be predicted.
method	which method for calculcating temperature-dependency should be used? Options are "Q10" (default) and "E". If either Q10 or E parameters have values, then this parameter is ignored.
Q10	(optional). A Q10 value to be used for predicting new values of "x". If method = "Q10" and Q10 is not defined here, then an appropriate Q10 value will be calculated internally using Q10.
Е	(optional). An E value to be used for predicting new values of "x". If method = "E" and E is not defined here, then an appropriate E value will be calculated internally using calc_E.
show_coef	logical. Should the temperature-dependency coefficient (i.e. the numeric value of either Q10 or E) be returned alongside the new values of "x"? Default is FALSE.
plot_fit	logical. Should a plot be displayed showing how well the new "x" values fit with the inputted data? Default is TRUE.

# Value

If  $show\_coef = FALSE$  (default), then a numeric vector of new values of "x" are returned. If  $show\_coef = TRUE$ , then a list of new values and the temperature-dependency coefficient are returned.

# Author(s)

Matthew A. Birk, <matthewabirk@gmail.com>

# See Also

```
Q10, calc_E
```

4 calc\_alpha

## **Examples**

```
# I measured Pcrit at four different temperatures. What is the Pcrit at an
# intermediate temperature?
adj_by_temp(meas_temp = c(5, 10, 15, 20), meas_x = c(3.1, 6.3, 7, 8.4), temp_new = 18)
# If requested values exceed the inputted temperature range, a message is reported.
# Biology cannot go on forever like the math can.
adj_by_temp(meas_temp = c(10, 15, 20, 25), meas_x = c(4.8, 6, 12.3, 13.6), temp_new = 0:30)
```

calc\_alpha

Calculate the oxygen supply capacity (alpha)

# **Description**

The oxygen supply capacity  $(\alpha)$  is a species- and temperature-specific value quantifying an animal's ability to extract oxygen from the ambient medium to support its metabolism (e.g. umol O2 / g / hr / kPa). This function calculates  $\alpha$  based on the single highest  $\alpha 0$  (MO2/PO2) value in the dataset. If there are outliers that make this prohibitive, consider setting a threshold MO2 value with mo2\_threshold.

# Usage

```
calc_alpha(po2, mo2, avg_top_n = 1, MR = NULL, mo2_threshold = Inf)
```

#### **Arguments**

po2 a vector of PO2 values.

mo2 a vector of metabolic rate values. Must be the same length and corresponding to

po2.

avg\_top\_n a numeric value representing the number of top  $\alpha 0$  (MO2/PO2) values to aver-

age together to estimate  $\alpha$ . Default is 1. When analyzing a trial where the animal was not at MMR the whole time, we recommend no more than 3 to avoid diminishing the  $\alpha$  value with sub-maximal observations. If all observations are believed to be at maximal O2 supply capacity, Inf can be used to average all

observations.

MR a vector of values for the metabolic rate at which pcrit\_alpha should be re-

turned. Default is NULL. If not specified, then pcrit\_alpha is not returned and

a message is added to the top of the return.

mo2\_threshold a single numeric value above which mo2 values are ignored. Useful to removing

obviously erroneous values. Default is Inf.

#### Value

Returns a list of 1) alpha, 2) a list of the PO2, MO2, and alpha0 value(s) where alpha was reached (the number of observations averaged is set by avg\_top\_n), and 3) the Pcrit at a metabolic rate of MR.

calc\_b 5

#### Author(s)

Matthew A. Birk, <matthewabirk@gmail.com>

# References

Seibel, B. A., A. Andres, M. A. Birk, A. L. Burns, C. T. Shaw, A. W. Timpe, C. J. Welsh. 2021. "Oxygen supply capacity breathes new life into the critical oxygen partial pressure (Pcrit)." Journal of Experimental Biology.

## See Also

```
calc_pcrit, plot_pcrit
```

# **Examples**

```
mo2_data <- read.csv(system.file('extdata', 'mo2_v_po2.csv', package = 'respirometry'))
calc_alpha(po2 = mo2_data$po2, mo2 = mo2_data$mo2, MR = 1.5) # MR set to 1.5 to capture the
# Pcrit corresponding to some of the lowest MO2 values recorded (something close to SMR).</pre>
```

```
# extract the alpha0 values that were averaged together
sapply(calc_alpha(po2 = mo2_data$po2, mo2 = mo2_data$mo2,
    MR = 1.5, avg_top_n = 3)$alpha_obs, function(i) i[3])
```

calc\_b

Calculate the metabolic scaling coefficient, b

## Description

For most organisms, metabolic rate does not scale linearly, but rather according to a power function:  $MO2 = b0 * M^b$ . This function estimates the scaling coefficient, b, and normalization constant, b0, given MO2s from different sized individuals.

# Usage

```
calc_b(mass, MO2, method = "nls", plot = "linear", b0_start = 1)
```

# **Arguments**

mass a vector of animal masses.

MO2 a vector of metabolic rates.

method a string defining which method of calculating scaling coefficients to use. Default

is "nls", which utilizes a nonlinear least squares regression. If this does not fit your data well, "lm" may also be used, which calculates a linear regression of  $log10(MO2) \sim log10(mass)$  with slope and intercept equivalent to b and  $10^b0$ ,

respectively.

6 calc\_E

plot a string defining what kind of plot to display. "linear" for linear axes, "log" for

log10-scale axes, and "none" for no plot. Default is "linear".

b0\_start a single numeric value as the starting point for b0 value determination using

the "nls" method. The default is 1 and should work for most situations, but if the "nls" method is not working and you don't want to use the "lm" method, changing the starting b0 value may help. Ignored when method = "lm".

## **Details**

$$MO2 = b0 * M^b$$

where b0 is species-specific normalization constant, M is mass and b is the scaling coefficient.

#### Value

Returns a list of 1) the b value, 2) a vector of b0 values corresponding to the input M02 values, and 3) an average b0 that can be used for summarizing the relationship with an equation.

#### Author(s)

Matthew A. Birk, <matthewabirk@gmail.com>

#### See Also

```
scale_MO2, calc_MO2
```

# **Examples**

```
# Simple example
mass <- c(1, 10, 100, 1000, 40, 4, 400, 60, 2, 742, 266, 983) # made up values
MO2 <- mass ^ 0.65 + rnorm(n = length(mass)) # make up some data
calc_b(mass = mass, MO2 = MO2)

# How about some mass-specific MO2s?
msMO2 <- mass ^ -0.25 + rnorm(n = length(mass), sd = 0.05)
calc_b(mass = mass, MO2 = msMO2)
calc_b(mass = mass, MO2 = msMO2, plot = "log")</pre>
```

calc\_E

Calculate E temperature coefficient

# Description

An E value is a relatively recent metric to parameterize the temperature-sensitivity of a biological rate (MO2). It is similar conceptually (but not numerically) to Q10.

calc\_MO2

# Usage

```
calc_E(x, temp)
```

# Arguments

x a numeric vector of rate values (e.g. MO2) or any other values (e.g. Pcrit).

temp a numeric vector of temperature values (in Celsius).

## **Details**

E is the slope of the relationship between  $-\ln(x)$  and 1/(kB T), where kB is the Boltzmann constant expressed in eV/K.

## Author(s)

Matthew A. Birk, <matthewabirk@gmail.com>

#### References

Deutsch, Curtis et al. 2015. "Climate Change Tightens a Metabolic Constraint on Marine Habitats." Science 348(6239): 1132–35.

# See Also

```
Q10, adj_by_temp
```

# **Examples**

```
calc_E(x = c(1, 2, 3), temp = c(10, 20, 30))
```

calc\_MO2

Calculate metabolic rate

# Description

Calculates metabolic rate (MO2) given O2 measurements over time. Oxygen measurements are split into bins and MO2s are calculated from each bin (unless bin\_width is set to 0). The bin\_width parameter defines the width of the bins in timed intervals (e.g. 15 minutes). Linear regressions are fit through each bin and the calculated MO2 is returned as the slope of the change in O2 over time.

8 calc\_MO2

## Usage

```
calc_MO2(
  duration,
  ο2,
  o2_unit = "percent_a.s.",
  bin_width,
  vol,
  temp = 25,
  sal = 35,
  atm_pres = 1013.25,
  time,
  pΗ,
  good_data = TRUE
)
calc_mo2(
  duration,
  02,
  o2_unit = "percent_a.s.",
  bin_width,
  vol,
  temp = 25,
  sal = 35,
  atm_pres = 1013.25,
  time,
  pH,
  good_data = TRUE
)
```

# **Arguments**

duration numeric vector of the timepoint for each observation (minutes).

o2 numeric vector of O2 observations.

o2\_unit a string describing the unit used to measure o2. Default is "percent\_a.s." Options

are from conv\_o2.

bin\_width numeric or data frame.

**OPTION 1:** A single number defining how long of a period should be binned for each MO2 determination (minutes). If MO2 is to be calculated from one observation to the next (rather than binned observations), set bin\_width to 0. To calculate a single MO2 value from all observations, set bin\_width to Inf.

**OPTION 2:** A data frame with two numeric columns: "o2" and "width". Useful for Pcrit calculations or another application where bins of different widths are desired at different PO2s. The data frame can be generated automatically by make\_bins or manually by the user. For each row, set the "width" value to the bin duration (minutes) desired for observations >= the value in the "o2" column but < the next greater O2 value in another row.

calc\_MO2

vol volume of the respirometer (liter).

temp temperature (°C). Default is 25 °C.

sal salinity (psu). Default is 35 psu.

atm\_pres atmospheric pressure (mbar). Default is 1013.25 mbar. time (optional). Numeric vector of timestamp observations.

pH (optional). Numeric vector of pH observations.

good\_data logical vector of whether O2 observations are "good" measurements and should

be included in analysis. For intermittent respirometry, flush periods should be labelled as FALSE. Linear regressions will not be fit over bins that include "bad" data. Bins will be split at bad data points. Default is that all observations are

TRUE.

#### Value

A data frame is returned:

**DUR MEAN** Mean duration of the bin (minutes).

**DUR\_RANGE** Range of duration timepoints in the bin.

TIME\_MEAN Exists only if the parameter time has values. Mean timestamp of the bin.

TIME\_RANGE Exists only if the parameter time has values. Range of timestamps in the bin.

**TEMP\_MEAN** Mean temperature of the bin.

**PH\_MEAN** Exists only if the parameter pH has values. Mean pH of the bin. Averaged using mean\_pH().

**O2 MEAN** Mean O2 value of the bin in the unit chosen by o2\_unit).

O2\_RANGE Range of O2 values in the bin.

MO2 Metabolic rate (umol O2 / hour).

**R2** Coefficient of determination for the linear regression fit to calculate MO2.

N Number of observations in the bin.

# Note

Whole-animal MO2 is returned. If mass-specific MO2 is desired, the output from calc\_MO2 can be divided by the animal's mass. No matter what unit of oxygen partial pressure or concentration measurement you put into the function as o2\_unit, the output in the MO2 column is always expressed in umol O2 / hour. This is because there is a vast variety of units for which people prefer to report dissolved oxygen levels, but most physiologists are at least unified in reporting metabolic rate as umol O2 per hour. If you prefer to report MO2 as mg O2 per hour, for example, you can always do something like: conv\_resp\_unit(df\$MO2, from = 'umol\_O2 / hr', 'mg\_O2 / hr') If only beginning and ending O2 observations are known, consider using closed. Both functions will work fine, but closed is simpler.

# Author(s)

Matthew A. Birk, <matthewabirk@gmail.com>

10 calc\_pcrit

#### See Also

```
make_bins, calc_b, closed, scale_MO2, conv_resp_unit
```

# **Examples**

```
# get 02 data
file <- system.file('extdata', 'witrox_file.txt', package = 'respirometry')</pre>
o2_data <- na.omit(import_witrox(file, split_channels = TRUE)$CH_4)
# calculate MO2
(mo2_5_min <- calc_MO2(duration = o2_data$DURATION, o2 = o2_data$O2,</pre>
bin_width = 5, vol = 10, temp = o2_data$TEMP, sal = o2_data$SAL))
# what if measurements from the 10 to 12 minute marks can't be trusted?
bad_data = o2_data$DURATION >= 10 & o2_data$DURATION <= 12</pre>
(mo2_5_min <- calc_MO2(duration = o2_data$DURATION, o2 = o2_data$O2,</pre>
bin_width = 5, vol = 10, temp = o2_data$TEMP, sal = o2_data$SAL, good_data = !bad_data))
# easily make a Pcrit plot
plot(mo2_5_min$02_MEAN, mo2_5_min$MO2)
# I want to express MO2 in mg per min instead.
(mo2\_5\_min\$MO2 <- conv\_resp\_unit(value = mo2\_5\_min\$MO2, from = 'umol\_O2 / hr', to = 'mg\_O2 / min'))
# just endpoint measurement:
calc_MO2(duration = o2_data$DURATION, o2 = o2_data$O2,
bin_width = Inf, vol = 10, temp = o2_data$TEMP, sal = o2_data$SAL)
# In my trial, observations above 77% air saturation were really noisy, but much less noisy at
# lower O2 values. I want to adjust my bin width based on the PO2 to obtain the best balance of
# resolution and precision throughout the whole trial. Below 77% a.s., use 4 minute bins. Above
# 77% a.s. use 10 minute bins.
bins = data.frame(o2 = c(77, 100), width = c(4, 10))
calc_MO2(duration = o2_data$DURATION, o2 = o2_data$O2,
bin_width = bins, vol = 10, temp = o2_data$TEMP, sal = o2_data$SAL)
```

calc\_pcrit

Calculate Pcrit

## **Description**

Calculates Pcrit (commonly understood as the threshold below which oxygen consumption rate can no longer be sustained) based on paired PO2 and MO2 values. Five Pcrit metrics are returned using many of the popular techniques for Pcrit calculation: the traditional breakpoint metric (broken stick regression), the nonlinear regression metric (Marshall et al. 2013), the sub-prediction interval metric (Birk et al. 2019), the alpha-based Pcrit method (Seibel et al. 2021), and the linear low O2 (LLO) method (Reemeyer & Rees 2019). To see the Pcrit values plotted, see plot\_pcrit.

calc\_pcrit 11

## Usage

```
calc_pcrit(
  po2,
  mo2,
  mo2_data,
  method = "Breakpoint",
  avg_top_n = 1,
  level = 0.95,
  iqr = 1.5,
  NLR_m = 0.065,
  MR = NULL,
  mo2_threshold = Inf,
  return_models = FALSE
)
```

# **Arguments**

po2 a vector of PO2 values. Any unit of measurement should work, but the NLR

calculation was optimized using kPa. If the NLR metric is giving you trouble,

try converting to kPa using conv\_o2.

mo2 a vector of metabolic rate values. Must be the same length and corresponding to

po2.

mo2\_data for convenience, the output of calc\_MO2 can be entered here, as an alternative

to specifying the po2 and mo2 parameters (optional).

method Over the years, many different methods of analysis have been proposed to quan-

tify Pcrit. You must choose one of the following: Alpha, Breakpoint (default),

LLO, NLR, Sub\_PI, All. If in doubt, try "All".

avg\_top\_n applies to the alpha metric only (only when method == "Alpha" or "All"). A

numeric value representing the number of top  $\alpha 0$  (MO2/PO2) values to average together to estimate  $\alpha$ . Default is 1. We recommend no more than 3 to avoid

diminishing the  $\alpha$  value with sub-maximal observations.

level applies to the Sub\_PI metric only (only when method == "Sub\_PI" or "All").

Percentage at which the prediction interval should be constructed. Default is

0.95.

igr applies to the Sub\_PI metric only (only when method == "Sub\_PI" or "All").

Removes mo2 observations that are this many interquartile ranges away from the mean value for the oxyregulating portion of the trial. If this filtering is not desired, set to infinity. To visualize which observations will be removed by this

parameter, use plot\_pcrit. Default is 1.5.

NLR\_m applies to the NLR metric only (only when method == "NLR" or "All"). Pcrit is

defined as the PO2 at which the slope of the best fitting function equals NLR\_m (after the MO2 data are normalized to the 90% quantile). Default is 0.065.

MR applies to the alpha and LLO metrics only (only when method == "Alpha",

"LLO" or "All"). A numeric value for the metabolic rate at which pcrit\_alpha and pcrit\_LLO should be returned. If not supplied by the user, then the mean MO2 of the "oxyregulating" portion of the curve is applied for pcrit\_alpha

and NA is returned for pcrit\_LLO.

12 calc\_pcrit

mo2\_threshold applies to the alpha metric only (only when method == "Alpha" or "All").

A single numeric value above which mo2 values are ignored for alpha Pcrit estimation. Useful to removing obviously erroneous values. Default is Inf.

return\_models logical. Should a list of model parameters be returned along with the converged

Pcrit values? Default is FALSE.

#### **Details**

**Alpha Pcrit** Alpha is calculated from calc\_alpha and the Pcrit corresponding to MR is returned. This determine's the animal's oxygen supply capacity and calculates the Pcrit at any given metabolic rate of interest. If no MR is provided, then it defaults to the mean MO2 value from the oxyregulating portion of the curve (as defined by the broken-stick regression).

Breakpoint Pcrit Data are fit to a broken-stick regression using segmented.

- **LLO Pcrit** A subset of observations are chosen only from those with an MO2 < MR. Then, a linear model is fit through the observations and Pcrit is calculated as the PO2 at which the line reaches MR.
- **NLR Pcrit** Data are fit to the following functions: Michaelis-Menten, Power, Hyperbola, Pareto, and Weibull with intercept. Following the method developed by Marshall et al. 2013, the function that best fits the data (smallest AIC) is chosen and the Pcrit is determined as the PO2 at which the slope of the function is NLR\_m (by default = 0.065 following the authors' suggestion).
- Sub\_PI Pcrit This metric builds off the Breakpoint metric and results in a systematically lower Pcrit value. This is useful for applications where it is important to ensure that Pcrit is not being overestimated. It represents a reasonable lower bounded estimate of the Pcrit value for a given trial. Once the Breakpoint Pcrit is calculated, a 95% prediction interval (can be changed with the level argument) is calculated around the oxyregulating region (i.e. using PO2 values > breakpoint Pcrit). By default, iqr provides some filtering of abberant observations to prevent their influence on the calculated prediction interval. Finally, the Sub\_PI Pcrit value is returned at the intersection of the oxyconforming line and the lower limit of the oxyregulating prediction interval.

#### Value

If return\_models is FALSE (default), a numeric Pcrit value is returned based on method. If method == "All", a named numeric vector of Pcrit values calculated using the Alpha, Breakpoint, LLO, NLR, and Sub\_PI metrics is returned. If return\_models is TRUE, then a list of converged Pcrit values, along with breakpoint function parameters, the MR value used for calculating Pcrit-alpha, a data frame of the "oxyregulating" portion of the curve, and NLR parameters are returned.

## Author(s)

Matthew A. Birk, <matthewabirk@gmail.com>

#### References

Birk, Matthew A., K.A.S. Mislan, Karen F. Wishner, and Brad A. Seibel. 2019. "Metabolic Adaptations of the Pelagic Octopod Japetella Diaphana to Oxygen Minimum Zones." Deep-Sea Research Part I 148: 123–31.

closed 13

Marshall, Dustin J., Michael Bode, and Craig R. White. 2013. "Estimating Physiological Tolerances - a Comparison of Traditional Approaches to Nonlinear Regression Techniques." Journal of Experimental Biology 216(12): 2176–82.

Reemeyer, Jessica E., and Bernard B. Rees. 2019. "Standardizing the Determination and Interpretation of Pcrit in Fishes." Journal of Experimental Biology 222(18): jeb210633.

Seibel, B. A., A. Andres, M. A. Birk, A. L. Burns, C. T. Shaw, A. W. Timpe, C. J. Welsh. 2021. "Oxygen supply capacity breathes new life into the critical oxygen partial pressure (Pcrit)." Journal of Experimental Biology.

#### See Also

```
plot_pcrit, calc_MO2, conv_o2, calc_alpha
```

# **Examples**

```
raw_data <- system.file('extdata/pcrit_run/', package = 'respirometry')
o2_data <- import_pyroscience_workbench(folder = raw_data)
mo2_data <- calc_MO2(duration = o2_data$DURATION, o2 = o2_data$CH_1_02, bin_width = 10, vol = 3)
calc_pcrit(mo2_data = mo2_data)
calc_pcrit(po2 = mo2_data$02_MEAN, mo2 = mo2_data$MO2, method = 'All', MR = 100)</pre>
```

closed

Closed respirometry

# Description

Returns the unknown parameter given 3 of 4 parameters to calculate respiration rate in a closed respirometer. This is useful both for basic closed respirometry setups, and also for the closed measurement phase of intermittent respirometry. Note that this is mainly useful for designing respirometry experiments. If you already have a timeseries dataset of oxygen measurements you want to analyze, use calc\_MO2 instead.

# Usage

```
closed(MO2, delta_pO2, duration, vol, temp = 25, sal = 35, atm_pres = 1013.25)
```

# **Arguments**

whole-animal oxygen consumption rate (umol O2 / hour).

delta\_p02 desired change in pO2 (% air saturation).

desired change in pO2 (% air saturation).

duration desired duration to reach delta\_p02 (minutes).

vol volume of the respirometer (liter).

temp temperature (°C). Default is 25 °C.

sal salinity (psu). Default is 35 psu.

atm\_pres atmospheric pressure (mbar). Default is 1013.25 mbar.

14 co2\_add

#### Note

If there are more than two O2 observations, consider using calc\_MO2.

#### Author(s)

Matthew A. Birk, <matthewabirk@gmail.com>

#### See Also

```
flush_water, calc_MO2
```

# **Examples**

```
# I've read in the literature that my animal has an SMR of 200 umol/h. How large of a
# respirometer do I want if I want it to breathe down to 80% air saturation in 30 minutes?
closed(MO2 = 200, delta_pO2 = 100 - 80, duration = 30) # returns respirometer volume

# I've read in the literature that my animal has an SMR of 1000 umol/h. How long will it take to
# breathe down a 50 L respirometer by 10% air saturation?
closed(MO2 = 1000, delta_pO2 = 10, vol = 50) # returns the duration to breathe down the O2

# How does animal size affect how long my measurement periods last?
mass_range <- seq(100, 400, 50)
dur_range <- (closed(MO2 = scale_MO2(mass_1 = 100, MO2_1 = 400, mass_2 = mass_range),
    delta_pO2 = 20, vol = 10))
plot(mass_range, dur_range, type = 'b')

# What is the MO2 if O2 drops 0.44 mg/l in 33 minutes when the respirometer volume is 30 L?
closed(delta_pO2 = conv_o2(o2 = 0.44, from = 'mg_per_l', to = 'percent_a.s.'), duration = 33,
    vol = 30)</pre>
```

co2\_add

Calculate CO2 to add to water

## **Description**

Calculates the moles of CO2 gas to be added to a volume of seawater to achieve the desired pCO2. Useful for ocean acidification experiments where CO2 treatments are desired.

# Usage

```
co2_add(
  goal_pco2,
  start_pH,
  vol,
  temp = 25,
  sal = 35,
  TA = NULL,
```

co2\_add

```
atm_pres = 1013.25
)
```

# **Arguments**

goal\_pco2 the desired pCO2 in the water (uatm).

start\_pH pH of the water before CO2 is added (total scale).

vol volume of the water (liter).

temperature (°C). Default is 25 °C.

sal salinity (psu). Default is 35 psu. If sal < 26 psu, then TA must be provided.

TA (optional) total alkalinity (umol / kg). If undefined TA is estimated from salinity

using guess\_TA.

atm\_pres atmospheric pressure (mbar). Default is 1013.25 mbar.

#### Value

moles of CO2 gas to be added to the seawater.

## Note

It is assumed that all of the CO2 added dissolves and remains in solution. This can be achieved (almost completely) by bubbling CO2 according to Jokiel et al. 2014.

#### Author(s)

Matthew A. Birk, <matthewabirk@gmail.com>

#### References

Jokiel PL, Bahr KD, Rodgers KS. 2014. Low-cost, high-flow mesocosm system for simulating ocean acidification with CO2 gas. Limnol Oceanogr Methods. 12:313–322.

#### See Also

```
co2_rate, flush_carb, carb, peri_pump
```

# **Examples**

```
# I want the 50 L reservoir to have a pCO2 = 1000 uatm. It currently has a pH of 7.88. # How many moles of CO2 gas should be added to the water to reach my desired pCO2? co2_add(goal_pco2 = 1000, start_pH = 7.88, vol = 50)
```

16 co2\_flush

co2\_flush

Calculate CO2 to add to flush reservoir

# Description

Calculates the moles of CO2 gas to be added to a seawater reservoir before flushing a respirometer to achieve the desired pCO2 in the respirometer after the flush. Useful for ocean acidification experiments where CO2 treatments are desired.

# Usage

```
co2_flush(
  goal_pco2,
  resp_pH,
  resp_vol,
  flush_pH,
  flush_remain = 0,
  temp = 25,
  sal = 35,
  TA = NULL,
  atm_pres = 1013.25
)
```

# **Arguments**

goal_pco2	the desired pCO2 in the respirometer after the flush (uatm).
resp_pH	pH inside the respirometer before the flush (total scale).
resp_vol	volume of the respirometer (liter).
flush_pH	pH of the reservoir water used for flushing before CO2 is added (total scale).
flush_vol	volume of the flush reservoir (liter).
flush_remain	volume of the flush reservoir that will remain after the flush (liter).
temp	temperature (°C). Default is 25 °C.
sal	salinity (psu). Default is 35 psu. If sal < 26 psu, then TA must be provided.
TA	(optional) total alkalinity (umol / kg). If undefined TA is estimated from salinity using ${\tt guess\_TA}$ .
atm_pres	atmospheric pressure (mbar). Default is 1013.25 mbar.

## Value

moles of CO2 gas to be added to the flush reservoir.

co2\_rate 17

#### Note

It is assummed that the entire reservoir is drained into the respirometer during the flush. It is also assumed that all of the CO2 added dissolves and remains in solution. This can be achieved (almost completely) by bubbling CO2 according to Jokiel et al. 2014.

#### Author(s)

Matthew A. Birk, <matthewabirk@gmail.com>

#### References

Jokiel PL, Bahr KD, Rodgers KS. 2014. Low-cost, high-flow mesocosm system for simulating ocean acidification with CO2 gas. Limnol Oceanogr Methods. 12:313–322.

## See Also

```
co2_add, co2_rate, flush_carb, carb, peri_pump
```

# **Examples**

```
# I want the respirometer to have a pCO2 = 1000 uatm. It currently has a pH of 7.6 and is 90 L.
# If I have a 200 L reservoir with pH = 7.9 seawater, how much CO2 do I need
# to add to the flush reservoir?
co2_flush(goal_pco2 = 1000, resp_pH = 7.6, resp_vol = 90, flush_pH = 7.9, flush_vol = 200)
```

co2\_rate

Calculate CO2 to add to a respirometer intake flow

## **Description**

Calculates the moles of CO2 gas to be added to a respirometer intake seawater flow to achieve the desired pCO2 in the respirometer. Useful for ocean acidification experiments where CO2 treatments are desired. Can be used for acclimation before a trial begins or for use with flow-through respirometry.

# Usage

```
co2_rate(
   goal_pco2,
   init_pH,
   flow_rate,
   temp = 25,
   sal = 35,
   TA = NULL,
   atm_pres = 1013.25,
   MO2 = NULL,
   RQ = 1
)
```

co2\_rate

# **Arguments**

goal_pco2	the desired pCO2 in the respirometer (uatm).
init_pH	ambient pH of the intake flow (total scale).
flow_rate	rate of water flow into the respirometer (liters / minute).
temp	temperature (°C). Default is 25 °C.
sal	salinity (psu). Default is 35 psu. If sal < 26 psu, then TA must be provided.
TA	(optional) total alkalinity (umol / kg). If undefined TA is estimated from salinity using $guess\_TA$ .
atm_pres	atmospheric pressure (mbar). Default is 1013.25 mbar.
M02	(optional) oxygen consumption rate (umol / hr). If defined, the $\rm CO2$ to be added is reduced to compensate for the $\rm CO2$ produced by the organism.
RQ	(optional) respiratory quotient: ratio of CO2 produced / O2 consumed. Only

## Value

moles of CO2 gas to be added to the intake flow per minute.

used if MO2 is defined. Default is 1.

# Note

It is assumed that all of the CO2 added dissolves and remains in solution. This can be achieved (almost completely) by bubbling CO2 according to Jokiel et al. 2014.

# Author(s)

Matthew A. Birk, <matthewabirk@gmail.com>

# References

Jokiel PL, Bahr KD, Rodgers KS. 2014. Low-cost, high-flow mesocosm system for simulating ocean acidification with CO2 gas. Limnol Oceanogr Methods. 12:313–322.

#### See Also

```
co2_add, flush_carb, carb, peri_pump
```

# **Examples**

```
# I want the respirometer to have a pCO2 = 1000 uatm. How much CO2 per minute do I need # to add to the intake flow if the ambient pH is 8.1 and it is flowing at 3 LPM? co2\_rate(goal\_pco2 = 1000, init\_pH = 8.1, flow\_rate = 3)
```

conv\_nh4

conv\_nh4

Convert between units of ammonia (NH3) / ammonium (NH4+)

## **Description**

Ammonia or nitrogen excretion can be measured in a variety of ways. Convert between different measurements.

# Usage

```
conv_nh4(n_waste, from = "umol_NH4", to = "all")
```

# **Arguments**

n\_waste

a numeric vector of the ammonia or nitrogen value(s).

from

a string describing the unit used to measure n\_waste. Default is "umol\_NH4" Options are:

- umol\_NH3
- umol\_NH4
- mg\_NH3
- mg\_NH4
- mg\_N

to

a single string either describing the unit to which the conversion should be conducted (options are the same as in from), or the string "all" to return all units.

# **Details**

The sum of NH4+ and NH3 species are considered (i.e. TAN). Conversions are based on relationships and values from the package marelac.

# Author(s)

```
Matthew A. Birk, <matthewabirk@gmail.com>
```

# See Also

```
predict_nh3, conv_o2
```

# **Examples**

```
conv_nh4(n_waste = 100)
conv_nh4(n_waste = 100, from = 'mg_N')
conv_nh4(n_waste = 100, from = 'mg_N', to = 'umol_NH4')
```

conv\_o2

conv\_o2

Convert between units of oxygen partial pressure and concentration

# **Description**

Unfortunately, a consensus on the best way to express how much oxygen is in water has not been formed to date. Until then, this function converts between all commonly used forms of dissolved O2 measurements.

# Usage

```
conv_o2(
    o2 = 100,
    from = "percent_a.s.",
    to = "all",
    temp = 25,
    sal = 35,
    atm_pres = 1013.25
)
```

# **Arguments**

02

a numeric vector of the O2 value(s). Default is 100.

from

a string describing the unit used to measure o2. Default is "percent\_a.s." Options are:

- percent\_a.s. (percent air saturation)
- percent\_o2
- hPa
- kPa
- torr
- mmHg
- inHg
- mg\_per\_1
- ug\_per\_1
- umol\_per\_l
- mmol\_per\_1
- nmol\_per\_ml
- ml\_per\_l
- mg\_per\_kg
- ug\_per\_kg
- umol\_per\_kg
- mmol\_per\_kg
- ml\_per\_kg

0

21 conv\_resp\_unit

```
    volumes_percent
```

a single string either describing the unit to which the conversion should be conto ducted (options are the same as in from), or the string "all" to return all units. temperature (°C). Default is 25 °C. temp salinity (psu). Default is 35 psu. sal atmospheric pressure (mbar). Default is 1013.25 mbar.

## **Details**

atm\_pres

Conversions are based on relationships and values from the package marelac which utilizes saturation values from Weiss 1970.

#### Author(s)

Matthew A. Birk, <matthewabirk@gmail.com>

#### References

Weiss R. 1970. The solubility of nitrogen, oxygen, and argon in water and seawater. Deep-Sea Research. 17:721-735.

# Examples

```
conv_o2(o2 = 50)
conv_o2(o2 = 1:50, from = "umol_per_l", to = "ml_per_l", temp = 10, sal = 0,
atm_pres = 1100)
conv_o2()[c('mmHg','kPa')]
```

conv\_resp\_unit

Convert units related to respirometry

# **Description**

Converts units of measurement that are joined by " / " or " \* ". This function expands upon conv\_multiunit to incorporate O2 unit conversion and seawater volume-mass conversions.

# Usage

```
conv_resp_unit(
  value,
  from,
  to,
  temp = 25,
  sal = 35,
 atm_pres = 1013.25,
  o2_conc_base = "per_1"
)
```

22 conv\_resp\_unit

# **Arguments**

value a numeric vector giving the measurement value in its original units.

from, to a string defining the unit with subunits separated by "/" or " \* ". See Details for proper notation regarding O2 and seawater mass/volume.

temp temperature (°C). Default is 25 °C.

sal salinity (psu). Default is 35 psu.

atm\_pres atmospheric pressure (mbar). Default is 1013.25 mbar.

o2\_conc\_base (optional) if converting between pO2 and [O2], should concentrations be "per\_1"

or "per\_kg"? Default is "per\_l".

## **Details**

The O2 units supported by conv\_o2 should be appended with "\_O2" (e.g. "kPa\_O2"; even "percent\_o2\_O2") and O2 unit concentrations should drop "per\_l" or "per\_kg" (e.g. "umol\_O2"). To designate seawater mass-volume conversion, append the unit with "\_seawater" (e.g. "kg\_seawater").

## Author(s)

Matthew A. Birk, <matthewabirk@gmail.com>

#### See Also

```
conv_multiunit, conv_o2, rho
```

## **Examples**

```
# I read that an animal's MO2 is 1.92 ml O2/kg/min. What is this MO2 in umol O2/g/h?
conv_resp_unit(value = 1.92, from = "ml_O2 / kg / min", to = "umol_O2 / g / hr")

# Krogh's diffusion coefficient for oxygen through gills can be expressed as ml O2 / mm2 (gill
# surface area) / um (gill thickness) / torr (seawater pO2 - blood pO2) / minute at a given
# temperature.
# To convert to another unit:
conv_resp_unit(value = 1e-6, from = "ml_O2 / mm2 / um / torr / min",
to = "umol_O2 / cm2 / um / kPa / hr", temp = 20)

# Now, with a knowledge of gill morphometrics, seawater pO2, and blood pO2, I can compare
# gill diffusion with whole animal MO2.
```

correct\_bubble 23

|--|

# Description

Given the volume of the respirometer and the volume of bubbles or air space, the moles of O2 in the system are calculated, and the volume of a respirometer holding the same quantity of O2 with only water is returned.

# Usage

```
correct_bubble(resp_vol, bubble_vol, temp = 25, sal = 35, atm_pres = 1013.25)
```

#### **Arguments**

resp\_vol volume of the respirometer (liter).

bubble\_vol volume of the gas bubbles or headspace (mL).

temp temperature (°C). Default is 25 °C. sal salinity (psu). Default is 35 psu.

atm\_pres atmospheric pressure (mbar). Default is 1013.25 mbar.

# Details

Depending on temperature and salinity, air holds 20,000x as much O2 as water per unit volume, thus small air bubbles in a respirometer can dramatically increase the amount of O2 an organism has to consume to lower the pO2 or aqueous [O2]. Thus air bubbles lead to underestimations of MO[2]. To correct for this in MO2 calculations after measurement, the volume of the respirometer can be increased. This function calculates the volume needed for MO2 calculations as a function of the volume of air space. Caution: allowing air bubbles into a respirometer is not recommended, even with this post-measurement adjustment. A small error in bubble volume estimation can lead to a large error in calculated metabolic rate.

# Value

The volume of a respirometer holding an equivalent quantity of O2 filled only with water.

#### Note

Due to the high concentration of O2 in air, very small errors in bubble volume estimates can lead to very large differences in the volume returned. Only trust the returned value if you are very confident of the accuracy of your bubble volume estimate.

## Author(s)

Matthew A. Birk, <matthewabirk@gmail.com>

24 flush\_carb

## See Also

molvol

## **Examples**

```
correct_bubble(resp_vol = 50, bubble_vol = 10) # a 10 mL bubble makes a huge difference!
correct_bubble(resp_vol = 50, bubble_vol = 1, temp = 10, sal = 0)
# in calculating MO2, a volume of 63.8 L should be used rather than the true 50 L.
```

flush\_carb

Estimate carbonate chemistry after a flush

# **Description**

Given the seawater pH inside the respirometer and in the flush reservoir, the new carbonate parameters (including pH) in the respirometer after the flush are estimated.

# Usage

```
flush_carb(
  resp_vol,
  flow_rate,
  duration,
  resp_pH,
  flush_pH,
  temp = 25,
  sal = 35,
  TA = NULL,
  atm_pres = 1013.25
)
```

# Arguments

volume of the respirometer (liter). resp\_vol flow\_rate rate of water flow into the respirometer (liters / minute). duration duration of the flush (minutes). resp\_pH pH inside the respirometer before the flush (total scale). flush\_pH pH of the water used for flushing the respirometer (total scale). temperature (°C). Default is 25 °C. temp sal salinity (psu). Default is 35 psu. If sal < 26 psu, then TA must be provided. (optional) total alkalinity (umol / kg). If undefined TA is estimated from salinity TΑ using guess\_TA.

atm\_pres atmospheric pressure (mbar). Default is 1013.25 mbar.

flush\_o2 25

# Value

A data frame returned by carb.

#### Author(s)

Matthew A. Birk, <matthewabirk@gmail.com>

#### See Also

```
carb, flush_water
```

# Examples

```
flush_carb(resp_vol = 90, flow_rate = 10, duration = 3, resp_pH = 7.8, flush_pH = 8.1)
# What will be the pH in the respirometer after this flush?
flush_carb(resp_vol = 90, flow_rate = 10, duration = 3, resp_pH = 7.8, flush_pH = 8.1)$pH
```

flush\_o2

Estimate dissolved O2 after a flush

# **Description**

Calculate the pO2 or [O2] in a respirometer after a flush. Given 5 of the 6 parameters, the 6th parameter is calculated.

# Usage

```
flush_o2(resp_vol, flow_rate, duration, resp_o2, flush_o2, final_o2)
```

# Arguments

resp_vol	volume of the respirometer (liter).
flow_rate	rate of water flow into the respirometer (liters / minute).
duration	duration of the flush (minutes).
resp_o2	O2 inside the respirometer before the flush (units do not matter as long as it is consistant with flush_o2 and final_o2).
flush_o2	O2 of the water used for flushing the respirometer (units do not matter as long as it is consistant with resp_o2 and final_o2).
final_o2	O2 of the water in the respirometer at the end of the flush (units do not matter as long as it is consistant with resp_o2 and flush_o2).

# Author(s)

Matthew A. Birk, <matthewabirk@gmail.com>

26 flush\_water

## See Also

```
flush_water, flush_carb
```

## **Examples**

```
# What will be the p02 in the respirometer after this flush?
flush_o2(resp_vol = 90, flow_rate = 10, duration = 3, resp_o2 = 15, flush_o2 = 21)
# I want to bring the p02 back up to 95% air saturation. How long do I need to flush?
flush_o2(resp_vol = 20, flow_rate = 2, resp_o2 = 75, flush_o2 = 99, final_o2 = 95)
```

flush\_water

Find percent of water exchanged after a flush

## **Description**

Calculate the proportion of water in a respirometer that is new after a flush. Useful for intermittent respirometry. Given 3 of the first 4 parameters, the 4th parameter is calculated.

## Usage

```
flush_water(vol, flow_rate, duration, perc_fresh, plot = FALSE)
```

# Arguments

vol volume of the respirometer (liter).

flow\_rate rate of water flow into the respirometer (liters / minute).

duration duration of the flush (minutes).

perc\_fresh percent of the respirometer volume that is new flushed water.

plot logical. Plot the percent exchanged as a function of flow rate and duration to see

what effect would result if the rate or duration are changed. All parameters must

only have a single value.

#### Author(s)

Matthew A. Birk, <matthewabirk@gmail.com>

# References

Steffensen JF. 1989. Some errors in respirometry of aquatic breathers: How to avoid and correct for them. Fish Physiol Biochem. 6:49–59. Equation 5.

## See Also

```
flush_carb, min_flow
```

goal\_flush\_pH 27

## **Examples**

```
# What proportion of a 90 L respirometer is exchanged after 20 minutes of flow at 2 LPM?
flush_water(vol = 90, flow_rate = 2, duration = 20)

# Would it be worth it to extend the flush another five minutes? How much would that
# improve the exchange?
flush_water(vol = 90, flow_rate = 2, duration = 20, plot = TRUE)
# Another five minutes would increase exchange by nearly 10%.
# Perhaps that's worth the extra time...

# Visualize flushing
vol = 150
flow_rate = seq(0, 10, by = 0.5)
duration = 0:60
perc_fresh = outer(flow_rate, duration, function(flow_rate, duration){
flush_water(vol = vol, flow_rate = flow_rate, duration = duration)
})
persp(flow_rate, duration, perc_fresh, xlab = 'Flow rate (LPM)', ylab = 'Duration (min)',
zlab = '% exchange', theta = 45, phi = 15, expand = 0.5, ticktype = 'detailed', nticks = 10)
```

goal\_flush\_pH

Calculate goal pH of a flush reservoir to achieve the post-flush goal pCO2

## **Description**

Calculates the pH of a flush reservoir that is needed to achieve the goal pCO2 after the flush reservoir has been drained into the respirometer.

# Usage

```
goal_flush_pH(
  goal_pco2,
  resp_pH,
  resp_vol,
  flush_vol,
  flush_remain = 0,
  temp = 25,
  sal = 35,
  TA = NULL,
  atm_pres = 1013.25
)
```

# Arguments

```
goal_pco2 the desired pCO2 in the respirometer after the flush (uatm).

resp_pH pH inside the respirometer before the flush (total scale).
```

28 guess\_TA

resp\_vol volume of the respirometer (liter).
flush\_vol volume of the flush reservoir (liter).

flush\_remain volume of the flush reservoir that will remain after the flush (liter).

temp temperature (°C). Default is 25 °C.

sal salinity (psu). Default is 35 psu. If sal < 26 psu, then TA must be provided.

TA (optional) total alkalinity (umol / kg). If undefined TA is estimated from salinity

using guess\_TA.

atm\_pres atmospheric pressure (mbar). Default is 1013.25 mbar.

#### Value

pH needed in the flush reservoir to achieve the goal pCO2 post-flush (total scale).

## Author(s)

Matthew A. Birk, <matthewabirk@gmail.com>

#### See Also

```
co2_rate, flush_carb, carb, peri_pump
```

#### **Examples**

```
# I want the respirometer to have a pCO2 = 1000 uatm. It currently has a pH of 7.6 and is 90 L.
# If I have a 200 L reservoir which will be drained completely, what do I want
# the pH of the reservoir to be?
goal_flush_pH(goal_pco2 = 1000, resp_pH = 7.6, resp_vol = 90, flush_vol = 200)
```

guess\_TA

Estimate total alkalinity from salinity

## **Description**

Estimate total alkalinity from salinity and temperature of surface seawater according to Lee et al. 2006. Useful when a rough guess of TA is needed because measuring TA is not possible or practical.

## Usage

```
guess_TA(temp = 25, sal = 35, region = NULL, extend = TRUE)
```

guess\_TA 29

# Arguments

temp	temperature (°C). Default is 25 °C.
sal	salinity (psu). Default is 35 psu. $31 \leq \text{sal} \leq 38$ ; may be narrower for some regions.
region	(optional) geographic region. Options are "(Sub)tropics", "Equatorial Upwelling Pacific", "North Atlantic", "North Pacific", and "Southern Ocean". Default is NULL. If undefined, the average from all these regions is used.

(see Details), should a guess be extrapolated? Default is TRUE.

logical. If salinity is  $\leq 5$  psu outside of the bounds defined by Lee et al. 2006

## **Details**

extend

```
(Sub)tropics temp \geq 20 and 31 \leq \text{sal} \leq 38
```

**Equatorial Upwelling Pacific** temp > 18 and 31 < sal < 36.5

**North Atlantic**  $0 \le \mathsf{temp} \le 20$  and  $31 \le \mathsf{sal} \le 37$ 

**North Pacific** temp  $\leq 20$  and  $31 \leq \text{sal} \leq 35$ 

**Southern Ocean** temp  $\leq 20$  and  $33 \leq \text{sal} \leq 36$ 

Estimates total alkalinity using the equations provided by Lee et al. 2006 (Geophysical Research Letters). While these equations are designed for open ocean environments, they can provide a rough estimate even for coastal environments. For improved estimate accuracy, the geographic region can be provided. The North Pacific region is longitude-dependent so a longitude of 150 °W is assumed which provides a typical value within the range. Only applicable for surface waters, not very accurate for the ocean interior.

#### Value

An estimate of the total alkalinity (umol / kg). If NA or NaN are returned, confirm the temp and sal values are within acceptable ranges for the region of interest.

# Author(s)

Matthew A. Birk, <matthewabirk@gmail.com>

#### References

Lee K, Tong LT, Millero FJ, Sabine CL, Dickson AG, Goyet C, Park G-H, Wanninkhof R, Feely RA, Key RM. 2006. Global relationships of total alkalinity with salinity and temperature in surface waters of the world's oceans. Geophys Res Lett. 33:L19605.

#### See Also

```
predict_pH
```

30 guess\_when

## **Examples**

```
guess_TA(temp = 22, sal = 33)
guess_TA(temp = 12, sal = 33, region = "North Atlantic")
guess_TA(temp = 20, sal = 31:35)

guess_TA(sal = 31) # salinity is within bounds
guess_TA(sal = 30) # salinity is outside the bounds and TA is extrapolated
guess_TA(sal = 30, extend = FALSE) # do not extrapolate TA
guess_TA(sal = 25, extend = TRUE) # will not extrapolate with sal > 5 psu out of bounds
```

guess\_when

Estimate when the O2 level will reach a defined level

# **Description**

Estimates the time at which O2 will reach a defined level assuming a linear change in O2 over time.

# Usage

```
guess_when(past_o2, past_time, goal_o2, plot = TRUE)
```

# **Arguments**

past_time a vector of timepoints corresponding to when past_o2 values were recorded. Can be a numeric vector for duration since trial began or a POSIX vector of time values.  goal_o2 a numeric vector or single value describing the O2 level of interest.  plot logical. Do you want to see a plot to visualize this prediction?	past_o2	a numeric vector of at least two oxygen measurements previously during the trial.
	past_time	Can be a numeric vector for duration since trial began or a POSIX vector of
plot logical. Do you want to see a plot to visualize this prediction?	goal_o2	a numeric vector or single value describing the O2 level of interest.
	plot	logical. Do you want to see a plot to visualize this prediction?

## Value

A prediction of the time when O2 will reach goal\_o2. If past\_time is numeric, then a numeric value(s) will be returned. If POSIX, then POSIX will be returned.

#### Note

Viewing the plot can be valuable if the O2 consumption or production is not linear.

#### Author(s)

```
Matthew A. Birk, <matthewabirk@gmail.com>
```

## See Also

```
predict_pH, predict_nh3
```

import\_firesting 31

## **Examples**

```
guess_when(past_o2 = rnorm(n = 10, mean = 100:91), past_time = 1:10, goal_o2 = 75, plot = FALSE) guess_when(past_o2 = rnorm(n = 10, mean = 100:91, sd = 5), past_time = 1:10, goal_o2 = 75) # Viewing the plot can be helpful to see how trustworthy the prediction is # when signal:noise is low.
```

import\_firesting

Import data from Pyro Oxygen Logger

## **Description**

Imports the standard txt file output from Pyroscience's deprecated Pyro Oxygen Logger software and converts the data into one or more data frames. If using the newer Pyroscience Workbench software, use import\_pyroscience\_workbench instead.

## Usage

```
import_firesting(
  file,
  o2_unit = "percent_a.s.",
  date = "%m/%d/%Y %X",
  overwrite_sal = NULL,
  keep_metadata = FALSE,
  drop_channels = TRUE,
  split_channels = FALSE
)
```

# **Arguments**

file a character string. The filepath for the file to be read. o2 unit a character string. The unit of O2 measurement to be output in the data frame. Options are described in conv\_o2. date a character string. The date format to be passed to strptime. overwrite\_sal Default NULL. To overwrite the salinity value(s) from calibration, enter a single numeric value for all channels or a numeric vector with values for each channel. Salinity of water sample (psu). keep\_metadata logical. Should metadata from the file be returned as extra columns in the returned data frame? Default is FALSE. drop\_channels logical. Should channels without any O2 data be dropped? Default is TRUE. split\_channels logical. Should a list of data frames be returned with a separate data frame for each channel? Default is FALSE.

32 import\_firesting

#### **Details**

The following FireSting fiber optic O2 transmitters are supported:

- FireStingO2
- FireStingO2 (1st generation)

If you would like support for the Piccolo2, FireStingO2-Mini, TeX4, or any OEM instruments, email me a data file from the device.

#### Value

A data frame (or list of data frames) is returned.

**TIME** Date and time, POSIXIt format.

**DURATION** Duration of measurement trial (minutes).

CH\_X\_O2 Oxygen measurement in desired unit as determined by o2\_unit.

**CH\_X\_TEMP** Temperature recorded or defined at beginning of measurement trial.

CH\_X\_SAL Salinity (psu).

... Channel columns (CH\_...) are repeated for each channel.

**COMMENT** Comments from FireSting file.

If keep\_metadata = TRUE, then the following columns are appended to the returned data frame:

**ATM\_PRES** Atmospheric pressure (mbar).

**HUMIDITY** Relative humidity (% RH).

**PROBE\_TEMP** Probe temperature.

**INTERNAL\_TEMP** Transmitter internal temperature.

**ANALOG\_IN** Voltage input from the extension port (mV).

CH\_X\_PHASE Phase recorded. Phase is inversely related to O2.

**CH\_X\_INTENSITY** Intensity is an indicator of the quality of the signal. A low intensity warning is produced by the transmitter below 10 mV.

**CH\_X\_AMB\_LIGHT** Ambient light on the sensor. Expressed in mV.

If split\_channels = TRUE, then "CH\_X\_" is removed from the column names and multiple data frames are returned in a named list.

#### Note

Oxygen conversions are estimates based on the marelac package.

#### Author(s)

Matthew A. Birk, <matthewabirk@gmail.com>

## See Also

import\_pyroscience\_workbench, import\_presens, import\_witrox, conv\_o2

import\_presens 33

# **Examples**

```
## Not run:
file <- system.file('extdata', 'pyro_oxygen_logger_file.txt', package = 'respirometry')
import_firesting(file, o2_unit = 'umol_per_1')

# I want each channel as a separate data frame.
data_list <- import_firesting(file, split_channels = TRUE)
data_list$CH_3 # here's the channel 3 data frame.

## End(Not run)</pre>
```

import\_presens

Import data from a PreSens O2 transmitter

# Description

Imports the standard text file output from most single channel PreSens fiber optic O2 transmitters and converts the data into a data frame.

# Usage

```
import_presens(
   file,
   o2_unit = "percent_a.s.",
   date = "%d/%m/%y",
   sal = 35,
   all_cols = FALSE,
   split_channels = FALSE
)
```

# **Arguments**

file	a character string. The filepath for the file to be read.
o2_unit	a character string. The unit of O2 measurement to be output in the data frame. Options are described in conv_o2.
date	a character string. The date format to be passed to strptime.
sal	salinity of water sample (psu). Default is 35 psu. Ignored for Fibox 4 files since salinity is provided by the file.
all_cols	logical. For Fibox 4 files only. Should all columns (including calibration data and serial numbers) be output?
split_channels	logical. For SDR SensorDish only. Should a list of data frames be returned with a separate data frame for each channel? Default is FALSE.

34 import\_presens

#### **Details**

The following PreSens fiber optic O2 transmitters are supported:

- Fibox 4
- Fibox 3
- Fibox 3 trace
- Fibox 3 LCD trace
- Microx TX3
- Microx TX3 trace
- · SDR SensorDish Reader

If you would like support for another PreSens O2 meter, email the package maintainer a data file from the device you would like supported. It is very important to note that the PreSens fiber optics O2 transmitters that are supported with this function (except the Fibox 4) DO NOT account for salinity (i.e. they assume salinity = 0 ppt). If the water sample measured was not fresh water, the oxygen concentrations (e.g. mg per liter or umol per liter) are incorrect in the PreSens txt file. This function corrects these O2 concentrations based on the salinity value defined by the sal argument. Absolute partial pressures (i.e. hPa and torr) will also be slightly different due to the slight influence of salinity on water's vapor pressure. This difference is typically ~0.05% of the recorded value.

#### Value

A data frame is returned.

TIME Date and time, POSIXct format.

**DURATION** Duration of measurement trial (minutes).

**O2** Oxygen measurement in desired unit as determined by o2\_unit.

**PHASE** Phase recorded. Phase is inversely related to O2. Not included in SDR SensorDish Reader files.

**AMPLITUDE** Amplitude recorded. Amplitude is an indicator of the quality of the signal. A low amplitude warning is produced by the transmitter below 2500. Not included in SDR SensorDish Reader files.

**TEMP** Temperature recorded or defined at beginning of measurement trial.

**ATM\_PRES** Atmospheric pressure (mbar).

**SAL** Salinity (psu).

**ERROR\_CODE** Error code from transmitter. See PreSens user manual for translation of error code. Not included in SDR SensorDish Reader files.

#### Note

Oxygen conversions are based on conv\_o2 and therefore differ slightly from the conversions provided by PreSens.

# Author(s)

Matthew A. Birk, <matthewabirk@gmail.com>

## See Also

import\_pyroscience\_workbench, import\_firesting, import\_witrox, conv\_o2

## **Examples**

```
## Not run:

# Import a Fibox 3 file.
file <- system.file('extdata', 'fibox_3_file.txt', package = 'respirometry')
import_presens(file, o2_unit = 'umol_per_1', sal = 25)

# Import a Fibox 4 file.
file <- system.file('extdata', 'fibox_4_file.csv', package = 'respirometry')
import_presens(file = file, date = '%d-%b-%Y')

# Import an SDR SensorDish Reader file.
file <- system.file('extdata', 'sdr_file.txt', package = 'respirometry')
import_presens(file = file, date = '%d.%m.%y%X')

## End(Not run)</pre>
```

import\_pyroscience\_workbench

Import data from Pyroscience Workbench

# **Description**

Imports the raw channel data from Pyroscience Workbench output files. This allows "live" analyses while the trial is still running. This does not utilize the ".pyr" file, nor the text file that is created once the trial is finished. This utilizes the raw channel data found within the "ChannelData" folder that the software makes when the trial starts.

# Usage

```
import_pyroscience_workbench(
  folder,
  o2_unit = "percent_a.s.",
  sal = NULL,
  keep_metadata = FALSE,
  split_channels = FALSE,
  merge_close_measurements = "min"
)
```

#### **Arguments**

folder

a character string. The filepath to the parent folder (directory) which contains ChannelData.

o2\_unit a character string. The unit of O2 measurement to be output in the data frame.

Options are described in conv\_o2.

sal numeric. If o2\_unit is a concentration rather than partial pressure, the salinity of the chamber in which measurements were made must be entered here.

keep\_metadata logical. Should metadata from the file be returned as extra columns in the returned data frame? Default is FALSE.

split\_channels logical. Should a list of data frames be returned with a separate data frame for each channel? Default is FALSE.

merge\_close\_measurements

used only when split\_channels = FALSE (the default). The frequency during which measurements are taken can be set uniquely for each channel in the Pyroscience Workbench software. When this happens, measurements may, at times, be nearly synchronized. When measurements are close together in time (even if not exactly at the same moment), it may be desirable to merge them together in the same row of the output dataframe and consider them to be the same timepoint. This parameter allows you to control whether that happens, and if so, how close is "close enough". Options are:

**0:** Do not merge close measurements no matter how close in time (even if 1 msec apart).

"min" (default): Merge measurements as close as the most frequently sampled channel (e.g. if channel 1 sampled every 5 seconds, channel 2 every 2 seconds, and channel 3 every 10 seconds, then any measurements within 2 seconds of each other will be merged on the same row in the output dataframe.)

"max": Merge measurements as close as the least frequently sampled channel (e.g. if channel 1 sampled every 5 seconds, channel 2 every 2 seconds, and channel 3 every 10 seconds, then any measurements within 10 seconds of each other will be merged on the same row in the output dataframe. Warning: this will duplicate more frequent channels. Do not let your downstream statistics be altered by artificially raising the number of observations.)

**custom-set numeric value:** A numeric value specifying how many seconds apart is "close enough" to merge measurements to the same timepoint. This may result in duplications of the same observations across multiple rows or not merging multiple observations as expected. Examine the output carefully.

## Value

A data frame (or list of data frames) is returned.

**TIME** Date and time, POSIXct format. If split\_channels = FALSE (default), then the timestamp is the average of all the measurements that were merged. For details, see merge\_close\_measurements.

**DURATION** Duration of measurement trial (minutes).

CH X O2 Oxygen measurement in desired unit as determined by o2\_unit.

**CH\_X\_TEMP** Temperature recorded or defined at beginning of measurement trial.

CH\_X\_SAL Salinity (psu). Only displayed if sal != NULL.

CH\_X\_STATUS Warning or error messages from Pyroscience Workbench file.

... Channel columns (CH\_...) are repeated for each channel.

If keep\_metadata = TRUE, then the following columns are appended to the returned data frame:

**CH\_X\_PHASE** Phase recorded. Phase is inversely related to O2.

**CH\_X\_INTENSITY** Intensity is an indicator of the quality of the signal.

CH\_X\_AMB\_LIGHT Ambient light on the sensor. Expressed in mV.

CH\_X\_T\_STATUS Warning or error messages from Pyroscience Workbench file's temperature measurement.

CH\_X\_ATM\_PRES Atmospheric pressure (mbar).

**CH\_X\_P\_STATUS** Warning or error messages from Pyroscience Workbench file's atmospheric pressure measurement.

If split\_channels = TRUE, then "CH\_X\_" is removed from the column names and multiple data frames are returned in a named list.

#### Note

Oxygen conversions are estimates based on the marelac package.

## Author(s)

Matthew A. Birk, <matthewabirk@gmail.com>

## See Also

```
import_presens, import_witrox, conv_o2
```

## **Examples**

```
## Not run:
folder <- system.file('extdata/pyro_wb/', package = 'respirometry')
import_pyroscience_workbench(folder = folder, o2_unit = 'umol_per_l', sal = c(0, 35))
# I want each channel as a separate data frame.
data_list <- import_pyroscience_workbench(folder = folder, split_channels = TRUE)
data_list$CH_2 # here's the channel 2 data frame.
## End(Not run)</pre>
```

38 import\_witrox

import\_witrox

Import data from a Loligo Systems Witrox O2 transmitter

## **Description**

Imports the standard txt file output from Loligo Systems Witrox fiber optic O2 transmitters and converts the data into one or more data frames.

## Usage

```
import_witrox(
   file,
   o2_unit = "percent_a.s.",
   date = "%m/%d/%Y %I:%M:%S %p",
   overwrite_sal = NULL,
   drop_channels = TRUE,
   split_channels = FALSE
)
```

## **Arguments**

file a character string. The filepath for the file to be read.

o2\_unit a character string. The unit of O2 measurement to be output in the data frame. Options are described in conv\_o2.

date a character string. The date format to be passed to strptime.

overwrite\_sal Default NULL. To overwrite the salinity value(s) from calibration, enter a single numeric value for all channels or a numeric vector with values for each channel. Salinity of water sample (psu).

drop\_channels logical. Should channels without any O2 data be dropped? Default is TRUE.

split\_channels logical. Should a list of data frames be returned with a separate data frame for each channel? Default is FALSE.

## **Details**

The following Loligo Systems fiber optic O2 transmitters are supported:

• Witrox 4

If you would like support for the Witrox 1, email me a data file from this device.

## Value

A data frame (or list of data frames) is returned.

TIME Date and time, POSIXIt format.

**DURATION** Duration of measurement trial (minutes).

make\_bins 39

**ATM\_PRES** Atmospheric pressure (mbar).

CH\_X\_PHASE Phase recorded. Phase is inversely related to O2.

**CH\_X\_TEMP** Temperature recorded or defined at beginning of measurement trial.

CH\_X\_SAL Salinity (psu).

**CH\_X\_O2** Oxygen measurement in desired unit as determined by o2\_unit.

... Channel columns (CH\_...) are repeated for each channel.

If split\_channels = TRUE, then "CH\_X\_" is removed from the column names and multiple data frames are returned in a list.

#### Author(s)

Matthew A. Birk, <matthewabirk@gmail.com>

#### See Also

import\_pyroscience\_workbench, import\_firesting, import\_presens, conv\_o2

## **Examples**

```
## Not run:
file <- system.file('extdata', 'witrox_file.txt', package = 'respirometry')
import_witrox(file, o2_unit = 'umol_per_l')

# Oops. I forgot to change the salinity value when I calibrated
# the instrument. Override the values in the file for 35 psu.
import_witrox(file, o2_unit = 'umol_per_kg', overwrite_sal = 35)

# I want each channel as a separate data frame.
data_list <- import_witrox(file, split_channels = TRUE)
data_list$CH_3 # here's the channel 3 data frame.

## End(Not run)</pre>
```

make\_bins

Make time binning thresholds for MO2 calculations

## **Description**

The width of time bins seems to be an under-appreciated consideration when calculating metabolic rates if PO2 or time are interesting covariates. The wider the bins, the higher the precision of your calculated MO2 value (more observations to average over), but at a loss of resolution of an interesting covariate. The narrower the bins, the higher the resolution of the PO2 or time covariate, but at a cost of lower precision. For Pcrit trials, I have found good success using bins of 1/10th the trial duration at the highest PO2s (where good precision is important) and 1/100th the trial duration at the lowest PO2s (where good resolution is important). Thus, these are the defaults, but can be changed as desired.

40 make\_bins

## Usage

```
make_bins(
    o2,
    duration,
    good_data = TRUE,
    min_o2_width = 1/100,
    max_o2_width = 1/10,
    n_thresholds = 10
)
```

## Arguments

02	numeric vector of O2 observations.
duration	numeric vector of the timepoints for each observation (minutes).
good_data	logical vector of whether O2 observations are "good" measurements and should be included in analysis. Default is that all observations are TRUE.
min_o2_width	The duration of the bins at the lowest O2 value, expressed as a proportion of the total "good" trial duration. Default is 1/100th of the total "good" trial duration.
max_o2_width	The duration of the bins at the highest O2 value, expressed as a proportion of the total "good" trial duration. Default is 1/10th of the total "good" trial duration.
n_thresholds	Default is 10.

## Value

A data frame with n\_thresholds rows and two columns is returned. Each row describes the threshold and the duration of observations that will be binned together at or above the corresponding O2 value.

**o2** The various O2 thresholds at which bin widths change.

width The bin width applied to values greater than the corresponding row's O2 value but less than the next greater O2 value.

## Author(s)

```
Matthew A. Birk, <matthewabirk@gmail.com>
```

## See Also

```
calc_MO2
```

## **Examples**

```
# get 02 data
file <- system.file('extdata', 'witrox_file.txt', package = 'respirometry')
o2_data <- na.omit(import_witrox(file, split_channels = TRUE)$CH_4)
# Total trial duration is 21.783 minutes</pre>
```

max\_MO2 41

```
make_bins(o2 = o2_data$02, duration = o2_data$DURATION) # creates the default 10 bins. At the
# highest 02 levels, bin widths are 21.783/10 = 2.1783 mins and at the lowest 02 levels, bin
# widths are 21.783/100 = 0.21783 mins.

bins <- make_bins(o2 = o2_data$02, duration = o2_data$DURATION, min_o2_width = 1/20,
max_o2_width = 1/3, n_thresholds = 5) # creates 5 bins. At the highest 02 levels, bin widths are
# 21.783/3 = 7.261 mins and at the lowest 02 levels, bin widths are 21.783/20 = 1.089 mins.

(mo2 <- calc_MO2(duration = o2_data$DURATION, o2 = o2_data$02,
bin_width = bins, vol = 10, temp = o2_data$TEMP, sal = o2_data$SAL))</pre>
```

max\_MO2

Maximum MO2 supported by flow rate

## **Description**

Calculates the maximum oxygen consumption rate (MO2) supported by a respirometer with a given flow rate. Useful for ensuring an acclimating animal maintains a normoxic environment.

## Usage

```
max_MO2(
  flow_rate,
 min_p02 = 90,
 p02_{in} = 100,
  temp = 25,
  sal = 35,
  atm_pres = 1013.25
)
max_mo2(
  flow_rate,
 min_p02 = 90,
 p02_in = 100,
  temp = 25,
  sal = 35,
  atm_pres = 1013.25
)
```

## Arguments

flow_rate	water flow rate into respirometer (liters / min).
min_pO2	minimum pO2 acceptable in respirometer ( $\%$ air saturation). Default is 90% air saturation.
pO2_in	pO2 of water entering respirometer (% air saturation). Default is 100% air saturation

42 mean\_pH

temp	temperature (°C). Default is 25 °C.
sal	salinity (psu). Default is 35 psu.
atm pres	atmospheric pressure (mbar). Default is 1013.25 mbar.

#### Value

The maximum whole-animal oxygen consumption rate (umol / hr) that can be sustained.

## Note

Keep in mind that most organisms are very stressed upon being placed in a respirometer and their MO2 may be much higher than basal MO2.

#### Author(s)

```
Matthew A. Birk, <matthewabirk@gmail.com>
```

## References

Steffensen JF. 1989. Some errors in respirometry of aquatic breathers: How to avoid and correct for them. Fish Physiol Biochem. 6:49–59. Equation 8.

#### See Also

```
min_flow, flush_water
```

## **Examples**

```
max_MO2(flow_rate = 1)

# What is the maximum MO2 organism I can place in my respirometer and still maintain at
# least 75% air saturation when the intake fresh water is 1.5 LPM, 10 °C and 90% air saturated?
(max_mo2 <- max_MO2(flow_rate = 1.5, min_pO2 = 75, pO2_in = 90, temp = 10, sal = 0))

# If a 300 g individual has an MO2 of 2000 umol/hr, how big of an animal can I use?
scale_MO2(mass_1 = 300, MO2_1 = 2000, MO2_2 = max_mo2) # I can almost support a 1 kg individual!</pre>
```

```
mean_pH
```

Mean pH by [H+]

## **Description**

Calculates mean pH from a vector of pH values by averaging [H+] rather than numerical pH values.

## Usage

```
mean_pH(pH, na.rm = FALSE, ...)
```

min\_flow 43

## Arguments

pH a numeric vector of pH values.

na.rm a logical value indicating whether NA values should be stripped before the computation proceeds.

... further arguments passed to or from other methods.

#### **Details**

Since pH is on a logarithmic scale, averaging pH values directly does not provide the true arithmetic mean of what is likely truly important to the organism, [H+] (however, see Boutilier and Shelton 1980). Thus, the pH values are converted to [H+] then averaged and converted back to a mean pH value.

## Author(s)

Matthew A. Birk, <matthewabirk@gmail.com>

## References

Boutilier RG, Shelton G. 1980. The statistical treatment of hydrogen ion concentration and pH. J Exp Biol. 84:335–339.

## **Examples**

```
mean_pH(c(7, 8)) # 7.26 rather than 7.5!
```

min\_flow

Minimum flow rate to support MO2

## **Description**

Calculates the minimum flow rate into a respirometer required to maintain a high pO2. Useful for ensuring an acclimating animal maintains a normoxic environment. It can also be used to estimate the flow rate needed for a given pO2 decrease desired for flow-through respirometry.

## Usage

```
min_flow(
   MO2,
   min_p02 = 90,
   pO2_in = 100,
   temp = 25,
   sal = 35,
   atm_pres = 1013.25
)
```

44 min\_flow

# **Arguments**

M02	whole-animal oxygen consumption rate (umol / hour).
min_p02	minimum pO2 acceptable in respirometer (% air saturation). Default is 90% air saturation.
p02_in	pO2 of water entering respirometer (% air saturation). Default is 100% air saturation.
temp	temperature (°C). Default is 25 °C.
sal	salinity (psu). Default is 35 psu.
atm_pres	atmospheric pressure (mbar). Default is 1013.25 mbar.

#### Value

The flow rate (liters / min) into the respirometer required for the steady state pO2 to be min\_pO2.

## Note

Keep in mind that most organisms are very stressed upon being placed in a respirometer and their MO2 may be much higher than basal MO2.

## Author(s)

Matthew A. Birk, <matthewabirk@gmail.com>

## References

Steffensen JF. 1989. Some errors in respirometry of aquatic breathers: How to avoid and correct for them. Fish Physiol Biochem. 6:49–59. Equation 8.

## See Also

```
max_MO2, flush_water
```

## **Examples**

```
min_flow(MO2 = 1000)

# What is the minimum flow rate required to maintain at least 75% air saturation in a
# respirometer with an organism(s) with an oxygen consumption rate of 1000 umol/h
# when the intake fresh water is 10 °C and 90% air saturated?
min_flow(MO2 = 1000, min_pO2 = 75, pO2_in = 90, temp = 10, sal = 0)
```

peri\_pump 45

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Calculate peristaltic pump gaseous flow rate

## **Description**

Given the number of moles of a gas, calculates the liters to run through a peristaltic pump.

## Usage

```
peri_pump(
   mol,
   species = "02",
   temp = 25,
   reg_pres,
   reg_unit = "psi",
   atm_pres = 1013.25
)
```

# **Arguments**

number of moles to go through a peristaltic pump.

species character string describing the gas species. Options are available from molvol.

Default is "O2".

temp temperature (°C). Default is 25 °C.

reg\_pres gauge pressure from the gas regulator into the peristaltic pump.

reg\_unit unit used in reg\_pres. Default is "psi".

atm\_pres atmospheric pressure (mbar). Default is 1013.25 mbar.

## **Details**

Most mass flow controllers are programmed with a "standard condition" something like 0 \*C and 1013 mbar for which they account for the pressure and temperature of an incoming gas source. For setups without expensive mass flow controllers, a more affordable alternative is to use a peristaltic pump. These do not account for variations in incoming gas pressure and temperature and thus, it must be calculated to set the peristaltic pump to the correct RPM.

# Author(s)

Matthew A. Birk, <matthewabirk@gmail.com>

#### See Also

```
co2_rate, co2_add
```

46 plot\_pcrit

## **Examples**

```
peri_pump(mol = 0.5, species = '02', temp = 10, reg_pres = 5, reg_unit = "kPa") # To flow 0.5 moles of 02, then flow 11.1 L.
```

plot\_pcrit

Plot Pcrit

# **Description**

Creates a Pcrit plot (the threshold below which oxygen consumption rate can no longer be sustained) based on paired PO2 and MO2 values. Five Pcrit metrics are plotted: the traditional breakpoint metric (broken stick regression, black), the nonlinear regression metric (Marshall et al. 2013, green), the sub-prediction interval metric (Birk et al. 2019, red), the alpha-based Pcrit method (Seibel et al., 2021, blue), and the linear low O2 (LLO) method (Reemeyer & Rees 2019, purple). For details on how the Pcrit values are calculated, see calc\_pcrit.

## Usage

```
plot_pcrit(
  po2,
  mo2,
  mo2_data,
  method = "Breakpoint",
  avg_top_n = 1,
  level = 0.95,
  iqr = 1.5,
  NLR_m = 0.065,
  MR = NULL,
  mo2_threshold = Inf,
  showNLRs = FALSE,
  ...
)
```

## **Arguments**

po2	a vector of PO2 values. Any unit of measurement should work, but the NLR calculation was optimized using kPa. If the NLR metric is giving you trouble, try converting to kPa using conv_o2.
mo2	a vector of metabolic rate values. Must be the same length and corresponding to po2.
mo2_data	for convenience, the output of calc_MO2 can be entered here, as an alternative to specifying the po2 and mo2 parameters (optional).
method	Over the years, many different methods of analysis have been proposed to quantify Pcrit. You must choose one of the following: Alpha, Breakpoint (default), LLO, NLR, Sub_PI, All. If in doubt, try "All".

plot\_pcrit 47

applies to the alpha metric only (only when method == "Alpha" or "All"). A avg\_top\_n numeric value representing the number of top  $\alpha 0$  (MO2/PO2) values to average together to estimate  $\alpha$ . Default is 1. We recommend no more than 3 to avoid diminishing the  $\alpha$  value with sub-maximal observations. level applies to the Sub\_PI metric only (only when method == "Sub\_PI" or "All"). Percentage at which the prediction interval should be constructed. Default is 0.95. iqr applies to the Sub\_PI metric only (only when method == "Sub\_PI" or "All"). Removes mo2 observations that are this many interquartile ranges away from the mean value for the oxyregulating portion of the trial. If this filtering is not desired, set to infinity. To visualize which observations will be removed by this parameter, use plot\_pcrit. Default is 1.5. NLR\_m applies to the NLR metric only (only when method == "NLR" or "All"). Pcrit is defined as the PO2 at which the slope of the best fitting function equals NLR\_m (after the MO2 data are normalized to the 90% quantile). Default is 0.065. MR applies to the alpha and LLO metrics only (only when method == "Alpha", "LLO" or "All"). A numeric value for the metabolic rate at which pcrit\_alpha and pcrit\_LLO should be returned. If not supplied by the user, then the mean MO2 of the "oxyregulating" portion of the curve is applied for pcrit\_alpha and NA is returned for pcrit\_LLO. mo2\_threshold applies to the alpha metric only (only when method == "Alpha" or "All"). A single numeric value above which mo2 values are ignored for alpha Pcrit estimation. Useful to removing obviously erroneous values. Default is Inf. logical. Should all the NLR functions be plotted in a second plot? If FALSE then showNLRs only the best fit NLR function will be plotted. arguments to be passed to plot. segmented.

#### **Details**

**Alpha Pcrit** Alpha is calculated from calc\_alpha and the Pcrit corresponding to MR is returned. This determine's the animal's oxygen supply capacity and calculates the Pcrit at any given metabolic rate of interest. If no MR is provided, then it defaults to the mean MO2 value from the oxyregulating portion of the curve (as defined by the broken-stick regression).

Breakpoint Pcrit Data are fit to a broken-stick regression using segmented.

- **LLO Pcrit** A subset of observations are chosen only from those with an MO2 < MR. Then, a linear model is fit through the observations and Pcrit is calculated as the PO2 at which the line reaches MR.
- **NLR Pcrit** Data are fit to the following functions: Michaelis-Menten, Power, Hyperbola, Pareto, and Weibull with intercept. Following the method developed by Marshall et al. 2013, the function that best fits the data (smallest AIC) is chosen and the Pcrit is determined as the PO2 at which the slope of the function is NLR\_m (by default = 0.065 following the authors' suggestion).
- **Sub\_PI Pcrit** This metric builds off the Breakpoint metric and results in a systematically lower Pcrit value. This is useful for applications where it is important to ensure that Pcrit is not being overestimated. It represents a reasonable lower bounded estimate of the Pcrit value for a given

48 plot\_pcrit

trial. Once the Breakpoint Pcrit is calculated, a 95% prediction interval (can be changed with the level argument) is calculated around the oxyregulating region (i.e. using PO2 values > breakpoint Pcrit). By default, iqr provides some filtering of abberant observations to prevent their influence on the calculated prediction interval. Finally, the Sub\_PI Pcrit value is returned at the intersection of the oxyconforming line and the lower limit of the oxyregulating prediction interval.

#### Value

A plot is created showing the relationship between PO2 and MO2. Based on the method used, the alpha, breakpoint, LLO, NLR, and/or sub-PI Pcrit values are shown in the title and on the plot by inverted triangles.

For breakpoint and sub-PI methods, the broken-stick regression is shown by black lines. The gray bands represent the confidence interval (defaults to 95% but will change with level).

For the sub-PI method, the dashed red curves signify the prediction interval used. Black circles represent oxyregulating observations used in the generation of the prediction interval, while grey circles represent both the oxyconforming observations and those observations outside the IQR threshold (defined by iqr).

For the NLR method, the green curve represents the best fitting NLR function and the green inverted triangle represents the NLR Pcrit (modified by NLR\_m)

For the Alpha method, the blue line represents alpha, which was fit based on the blue circle observation(s). If MR is not defined by the user, then the black points are those that were averaged to choose MR. These are the "oxyregulating" observations based on the breakpoint method.

If showNLRs = TRUE, then a second plot is generated which shows all the NLR functions that converged. Vertical lines represent the Pcrit values corresponding to each curve.

Black = Michaelis-Menten

Red = Power

Green = Hyperbola

Blue = Pareto

Cyan = Weibull with intercept.

#### Author(s)

Matthew A. Birk, <matthewabirk@gmail.com>

## References

Birk, Matthew A., K.A.S. Mislan, Karen F. Wishner, and Brad A. Seibel. 2019. "Metabolic Adaptations of the Pelagic Octopod Japetella Diaphana to Oxygen Minimum Zones." Deep-Sea Research Part I 148: 123–31.

Marshall, Dustin J., Michael Bode, and Craig R. White. 2013. "Estimating Physiological Tolerances - a Comparison of Traditional Approaches to Nonlinear Regression Techniques." Journal of Experimental Biology 216(12): 2176–82.

Reemeyer, Jessica E., and Bernard B. Rees. 2019. "Standardizing the Determination and Interpretation of Pcrit in Fishes." Journal of Experimental Biology 222(18): jeb210633.

predict\_nh3 49

Seibel, B. A., A. Andres, M. A. Birk, A. L. Burns, C. T. Shaw, A. W. Timpe, C. J. Welsh. 2021. "Oxygen supply capacity breathes new life into the critical oxygen partial pressure (Pcrit)." Journal of Experimental Biology.

#### See Also

```
calc_pcrit, calc_alpha
```

## **Examples**

```
raw_data <- system.file('extdata/pcrit_run/', package = 'respirometry')
o2_data <- import_pyroscience_workbench(folder = raw_data)
mo2_data <- calc_MO2(duration = o2_data$DURATION, o2 = o2_data$CH_1_02, bin_width = 10, vol = 3)
plot_pcrit(mo2_data = mo2_data)

par(mfrow = c(2, 1))
plot_pcrit(po2 = mo2_data$02_MEAN, mo2 = mo2_data$MO2, method = 'All', MR = 100, showNLRs = TRUE)</pre>
```

predict\_nh3

Predict NH3 / NH4+ concentration post-respiration

## **Description**

Predicts the [NH3] and [NH4+] of seawater after a defined amount of oxygen consumption. Ammonotelic animals excrete the ionized form NH4+ (ammonium) but some of these ions dissociate into unionized NH3 (ammonia) which is toxic for most fishes and crustaceans around 0.4-2.0 mg/L (Boyd 2012).

## Usage

```
predict_nh3(
    o2_drop = 10,
    o2_unit = "percent_a.s.",
    o2_nh4_ratio,
    temp = 25,
    sal = 35,
    pH = 8.1,
    atm_pres = 1013.25
)
```

## **Arguments**

```
o2_drop a numeric value or vector describing the change in O2. Default is 10.
o2_unit a string describing the unit used to measure o2_drop. Default is "percent_a.s."
Options are from conv_o2.
o2_nh4_ratio molar ratio of O2 consumed to NH4+ produced.
```

50 predict\_pH

temp	temperature (°C). Default is 25 °C.
sal	salinity (psu). Default is 35 psu.

pH seawater pH (total scale). Default is 8.1.

atm\_pres atmospheric pressure (mbar). Default is 1013.25 mbar.

#### **Details**

Given a known amount of oxygen consumed and an estimated O2:N ratio, the amount of NH4 produced can be estimated. Production or consumption of ammonium by "background" microbes or conversion of ammonium to nitrite and nitrate is ignored since bacteria in the respirometer are typically sought to be in low levels. The amount of dissociation to produce ammonia is calculated by Kn.

## Value

A list containing the predicted NH3, NH4+, and TAN produced in mg/l.

#### Author(s)

Matthew A. Birk, <matthewabirk@gmail.com>

## References

Boyd C. 2012. Water Quality. In "Aquaculture: Farming Aquatic Animals and Plants". Blackwell Publishing, Ltd.

## See Also

```
conv_o2, conv_nh4, Kn
```

# **Examples**

```
predict_nh3(o2_drop = 25, o2_nh4_ratio = 10)
```

predict\_pH

Predict pH post-respiration

# Description

Predicts the pH of seawater after a defined amount of oxygen consumption.

predict\_pH 51

## Usage

```
predict_pH(
   start_o2 = 100,
   end_o2,
   start_pH,
   temp = 25,
   sal = 35,
   RQ = 1,
   TA = NULL,
   all_carb = FALSE
)
```

## **Arguments**

start_o2	pO2 at the start of the measurement ( $\%$ air saturation). Default is 100% air saturation.
end_o2	pO2 at the end of the measurment (% air saturation).
start_pH	seawater pH (total scale) at the start of the measurement.
temp	temperature (°C). Default is 25 °C.
sal	salinity (psu). Default is 35 psu. If sal < 26 psu, then TA must be provided.
RQ	respiratory quotient: ratio of CO2 produced / O2 consumed. Default is 1.
TA	(optional) total alkalinity (umol / kg). If undefined TA is estimated from salinity using ${\tt guess\_TA}$ .
all_carb	logical. Should all carbonate chemistry parameters be returned? Default is FALSE.

# Details

Given a known amount of oxygen consumed and an estimated respiratory quotient (see Q10), the amount of CO2 produced can be estimated. From this CO2 production estimate, the carbonate chemistry of the seawater can be estimated. Atmospheric pressure is assumed.

#### Value

If all\_carb is FALSE, then a list of the predicted pH (total scale) at the end of the measurement and the predicted pCO2 (uatm) are returned. If all\_carb is TRUE, then the predicted carbonate chemistry parameters are returned from carb.

## Author(s)

Matthew A. Birk, <matthewabirk@gmail.com>

#### See Also

```
carb, guess_TA
```

52 Q10

## **Examples**

```
predict_pH(end_o2 = 75, start_pH = 8.1) predict_pH(start_o2 = 75, end_o2 = 50, start_pH = 7.96, temp = 15, sal = 33, RQ = 0.88) # I know pH at the end was 7.8, but what was pH at the beginning? predict_pH(start_o2 = 75, end_o2 = 100, start_pH = 8.013536) # reverse the order
```

Q10

Parameters of Q10 Temperature Coefficient

## Description

Calculates parameters from Q10 temperature coefficient for chemical or biological systems. This function can be used in two ways. 1. if four of the first five parameters are given (Q10, R1, R2, T1, T2) then the fifth parameter is returned, or 2. if R\_vec and T\_vec are given, then the best Q10 for those data is returned.

## Usage

```
Q10(Q10, R1, R2, T1, T2, R_vec, T_vec, model = FALSE)
q10(Q10, R1, R2, T1, T2, R_vec, T_vec, model = FALSE)
calc_q10(Q10, R1, R2, T1, T2, R_vec, T_vec, model = FALSE)
```

# Arguments

Q10	factor by which rate changes due to 10 °C increase in temperature.
R1	rate 1. Could also be Pcrit or any other temperature-dependent biological parameters.
R2	rate 2. Could also be Pcrit or any other temperature-dependent biological parameters.
T1	temperature 1 (in °C).
T2	temperature 2 (in °C).
R_vec	a vector of temperature-dependent values, such as rates (e.g. MO2), Pcrit or other biological parameters.
T_vec	a vector of temperature values (in °C).
model	logical. If TRUE, then a list is returned which includes an linear model of log10(R_vec) and T_vec fit by stats::lm(). Default is FALSE.

## **Details**

$$Q_{10} = (R_2/R_1)^{\frac{10}{T_2 - T_1}}$$

respirometry 53

#### Author(s)

Matthew A. Birk, <matthewabirk@gmail.com>

#### See Also

```
scale_MO2, calc_E, adj_by_temp
```

## **Examples**

```
Q10(R1 = 5, R2 = 10, T1 = 10, T2 = 20) # Returns Q10; = 2
Q10(Q10 = 2.66, R1 = 5, T1 = 10, T2 = 20) \# Returns R2; = 13.3
# My species has an MO2 of 9.5 umol/g/h at 10 *C. What MO2 should I expect at 13 *C?
Q10(Q10 = 2, R1 = 9.5, T1 = 10, T2 = 13) # expect ~11.7 umol/g/h at 13 *C.
# I measured MO2 at a spectrum of temperatures. What Q10 value best fits my data?
Q10(R_{\text{vec}} = c(1, 2, 5, NA, 18, 33), T_{\text{vec}} = c(0, 10, 20, 30, 40, 50))
# A 100 g individual at 10 *C has an MO2 of 1270 umol/h. How much
# would a 250 g individual likely consume at 14 *C?
Q10(Q10 = 2, R1 = scale_MO2(mass_1 = 100, MO2_1 = 1270, mass_2 = 250), T1 = 10, T2 = 14)
# Visualize MO2 scaling by mass and temperature:
mass <- seq(10, 200, 10)
temp <- 10:25
base_mass <- 50
base_temp <- 20
base_MO2 <- 750
mo2 <- outer(mass, temp, function(mass, temp){</pre>
scale\_MO2(mass\_1 = base\_mass, mass\_2 = mass, MO2\_1 = Q10(Q10 = 2, R1 = base\_MO2, mass\_2 = mass_2 = m
 T1 = base_temp, T2 = temp))
})
persp(mass, temp, mo2, xlab = 'Mass (g)', ylab = 'Temperature (*C)', zlab = 'MO2 (umol / hr)',
  theta = 35, phi = 15, expand = 0.5, ticktype = 'detailed', nticks = 10)
```

respirometry

Tools for Conducting Respirometry Experiments

## Description

Provides tools to enable the researcher to more precisely conduct respirometry experiments. Strong emphasis is on aquatic respirometry. Tools focus on helping the researcher setup and conduct experiments. Analysis of the resulting data is not a focus since analyses are often specific to a particular setup, and thus are better created by the researcher individually. This package provides tools for intermittent, flow-through, and closed respirometry techniques.

## Author(s)

Matthew A. Birk, <matthewabirk@gmail.com>

54 *RQ* 

RQ

Calculate respiratory quotient

# Description

Calculates the respiratory quotient (RQ), or ratio of CO2 produced to O2 consumed between observations. To calculate CO2 produced, either DIC or both pH and TA must be provided.

# Usage

# Arguments

o2	a numeric vector of O2 values with a length of at least 2.
o2_unit	a string describing the unit used to measure o2. Default is "percent_a.s." Options are from conv_o2.
рН	pH (total scale). Elements must align with o2 vector.
TA	total alkalinity (umol / kg). May be either a vector with length equal to o2 or a single numeric value.
DIC	dissolved inorganic carbon (umol / kg). Elements must align with o2 vector.
temp	temperature (°C). Default is 25 °C.
sal	salinity (psu). Default is 35 psu.
atm_pres	atmospheric pressure (mbar). Default is 1013.25 mbar.

## Value

ratio of CO2 produced to O2 consumed.

## Note

If you want a rough estimate of RQ, but only have pH measurements, TA can be estimated from salinity using guess\_TA.

# Author(s)

Matthew A. Birk, <matthewabirk@gmail.com>

scale\_MO2 55

## See Also

```
conv_o2, guess_TA
```

## **Examples**

```
o2_observations <- c(21, 18, 14.5, 7)
pH_observations <- c(8.05, 7.98, 7.86, 7.65)
TA_observations <- c(2222, 2219, 2208, 2214)

RQ(o2 = o2_observations, o2_unit = 'kPa', pH = pH_observations,
TA = TA_observations, temp = 20, sal = 33)

DIC_observations <- c(2222, 2250, 2284, 2355)
RQ(o2 = o2_observations, o2_unit = 'kPa', DIC = DIC_observations)

RQ(o2 = o2_observations, o2_unit = 'kPa', pH = pH_observations, TA = 2032)
```

scale\_MO2

Mass-correct metabolic rate

## Description

For most organisms, metabolic rate does not scale linearly, but rather according to a power curve. This function estimates MO2 or size of an individual organism given the MO2 and size of another individual of a different size. To mass-correct your MO2 data, plug in your desired mass in mass\_2 and the output from calc\_b to the b parameter.

## Usage

```
scale_MO2(mass_1, MO2_1, mass_2, MO2_2, b = 0.75)
scale_mo2(mass_1, MO2_1, mass_2, MO2_2, b = 0.75)
```

## Arguments

```
mass_1 animal mass for MO2_1.

MO2_1 metabolic rate for mass_1.

mass_2 animal mass for MO2_2.

MO2_2 metabolic rate for mass_2.

b scaling coefficient for MO2. Default is 0.75.
```

56 scale\_MO2

#### **Details**

$$(MO2 = b0 * M^b)$$

where b0 is species-specific normalization constant, M is mass and b is the scaling coefficient which is around 0.75 for many organisms.

For scaling of **mass-specific** metabolic rates, use something closer to b = -0.25 rather than b = 0.75.

#### Author(s)

Matthew A. Birk, <matthewabirk@gmail.com>

#### See Also

```
010. calc b
```

## **Examples**

```
# I know a species has an SMR of 800 umol 02/h at 200 g.
# What would be a likely SMR for a 300 g individual?
scale_MO2(mass_1 = 200, MO2_1 = 800, mass_2 = 300)
# Some squids have a much higher scaling coefficient:
scale_MO2(mass_1 = 200, MO2_1 = 800, mass_2 = 300, b = 0.92)
# A 100 g individual at 10 *C has an MO2 of 1270 umol/h. How much
# would a 250 g individual likely consume at 14 *C?
Q10(Q10 = 2, R1 = scale\_M02(mass\_1 = 100, M02\_1 = 1270, mass\_2 = 250), T1 = 10, T2 = 14)
# Now I have data from real animals and I want to mass-correct them all to a 10 g animal.
mass = 2:20 # obviously not real but you get the point
mo2 = c(44.8, 41, 36, 35, 35, 33.5, 34.5, 40, 30, 23, 27, 30, 25.6, 27.8, 28, 24, 27, 28, 20)
desired_mass = 10
b = calc_b(mass = mass, MO2 = mo2)
scale_MO2(mass_1 = mass, MO2_1 = mo2, mass_2 = desired_mass, b = b$b)
plot(mass, mo2, ylab = 'Raw MO2') # before
plot(mass, scale\_MO2(mass\_1 = mass, MO2\_1 = mo2, mass\_2 = 10, b = b$b),
ylab = 'Mass-corrected MO2') # after
# Visualize MO2 scaling by mass and temperature:
mass \leftarrow seq(10, 200, 10)
temp <- 10:25
base_mass <- 50
base_temp <- 20
base_MO2 <- 750
mo2 <- outer(mass, temp, function(mass, temp){</pre>
scale_M02(mass_1 = base_mass, mass_2 = mass, M02_1 = Q10(Q10 = 2, R1 = base_M02,
T1 = base_temp, T2 = temp))
```

scale\_MO2 57

```
})
persp(mass, temp, mo2, xlab = 'Mass (g)', ylab = 'Temperature (*C)', zlab = 'MO2 (umol / hr)',
theta = 35, phi = 15, expand = 0.5, ticktype = 'detailed', nticks = 10)
```

# **Index**

adj_by_temp, 2, 7, 53	mean_pH, 42
	min_flow, 26, 42, 43
calc_alpha, 4, <i>12</i> , <i>13</i> , <i>47</i> , <i>49</i>	molvol, 24, 45
calc_b, 5, 10, 55, 56	, ,
calc_E, 2, 3, 6, 53	peri_pump, 15, 17, 18, 28, 45
calc_MO2, 6, 7, 11, 13, 14, 40, 46	plot.segmented, 47
calc_mo2(calc_MO2), 7	plot_pcrit, 5, 10, 11, 13, 46, 47
calc_pcrit, 5, 10, 46, 49	predict_nh3, 19, 30, 49
calc_q10 (Q10), 52	predict_pH, 29, 30, 50
carb, 15, 17, 18, 25, 28, 51	
closed, 9, 10, 13	Q10, 2, 3, 7, 51, 52, 56
co2_add, 14, <i>17</i> , <i>18</i> , <i>45</i>	q10 (Q10), 52
co2_flush, 16	
co2_rate, 15, 17, 17, 28, 45	respirometry, 53
conv_multiunit, 21, 22	respirometry-package (respirometry), 53
conv_nh4, 19, 50	rho, 22
conv_o2, 8, 11, 13, 19, 20, 22, 31–39, 46, 49,	RQ, 54
50, 54, 55	
conv_resp_unit, <i>10</i> , 21	scale_MO2, 6, 10, 53, 55
correct_bubble, 23	scale_mo2(scale_MO2), 55
correct_bubble, 25	segmented, <i>12</i> , <i>47</i>
flush_carb, 15, 17, 18, 24, 26, 28	strptime, <i>31</i> , <i>33</i> , <i>38</i>
flush_o2, 25	
flush_water, 14, 25, 26, 26, 42, 44	
goal_flush_pH, 27	
guess_TA, 15, 16, 18, 24, 28, 28, 51, 54, 55	
guess_when, 30	
import_firesting, 31, 35, 39	
import_presens, <i>32</i> , <i>33</i> , <i>37</i> , <i>39</i>	
<pre>import_pyroscience_workbench, 31, 32, 35,</pre>	
35, 39	
import_witrox, 32, 35, 37, 38	
Kn, 50	
make_bins, 8, 10, 39	
marelac, 19, 21, 32, 37	
max_M02, 41, 44	
max_mo2 (max_MO2), 41	