

# General principle of calibration and validation. Application to optical sensors.

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Thank you for comments from Giorgio Dall'Olmo, Meg Estapa and Collin Roesler.

Questions:

- 1. What do YOU think is meant by calibration?*
- 2. What do YOU think is meant by validation?*

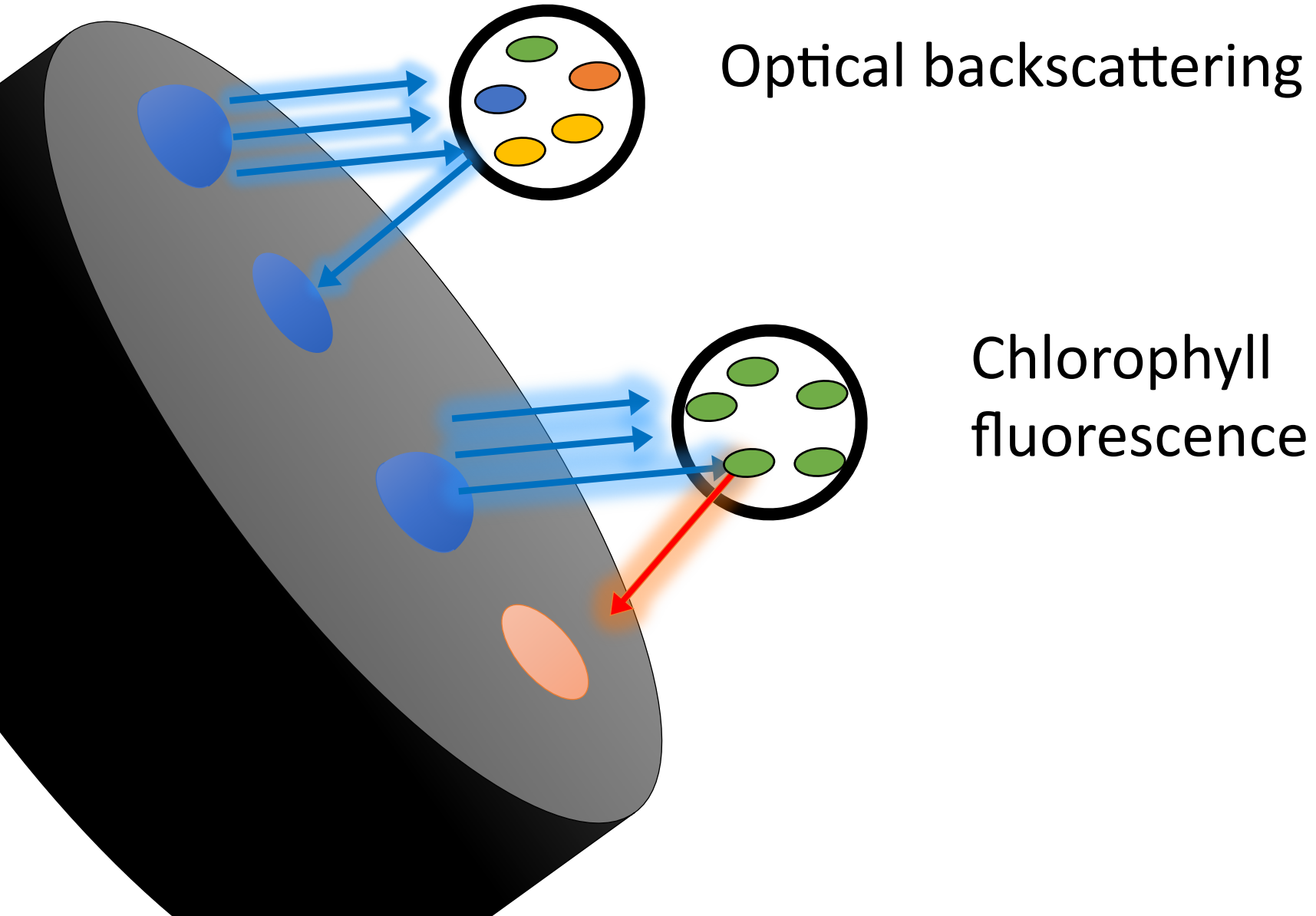
# CALIBRATION

- Calibration Is a process that ensures that accuracy is maintained in the measurements produced by your equipment.
- Calibration performance of any equipment is compared against a reference standard. – e.g. the process that convert raw data (e.g., volts/counts) to data in physical units (e.g., 1/m) using NIST traceable beads.
- Calibration assures accuracy of measurements  
You must periodically calibrate your instruments, identify if there is a drift in the measurements and eliminate it through calibration.
- It should be performed as per calibration SOP

# VALIDATION

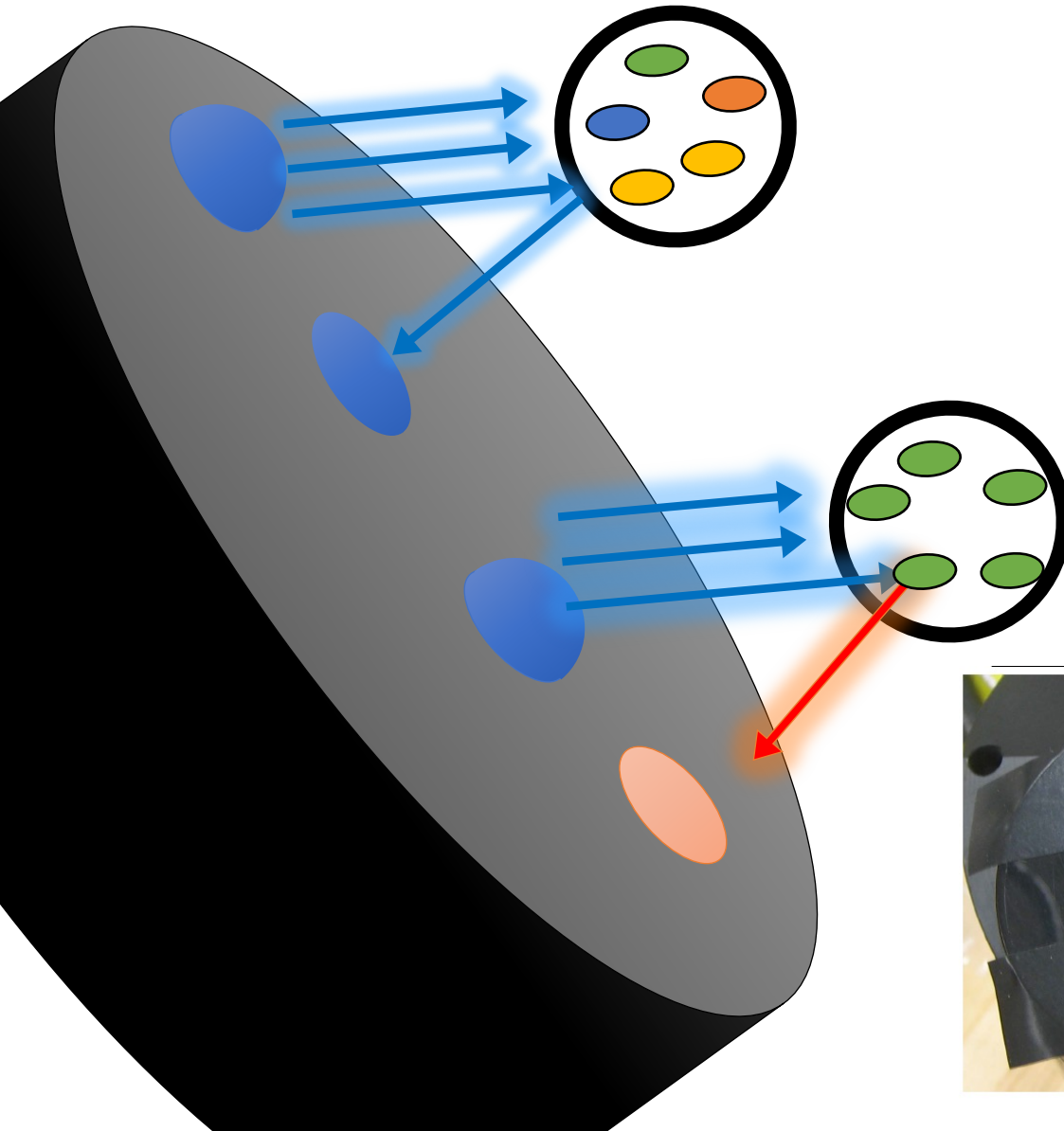
- Validation is a documented process that provides assurance that a product, service or system *consistently* provides results within the acceptable criteria.
- There are no reference standards used in validation.
- Validation provides proof of consistence across all the processes or methods being used.

Example: *how are the Eco-FLBB sensors calibrated?*  
What could be used as a standard?



# Example: how are the Eco-FLBB sensors calibrated?

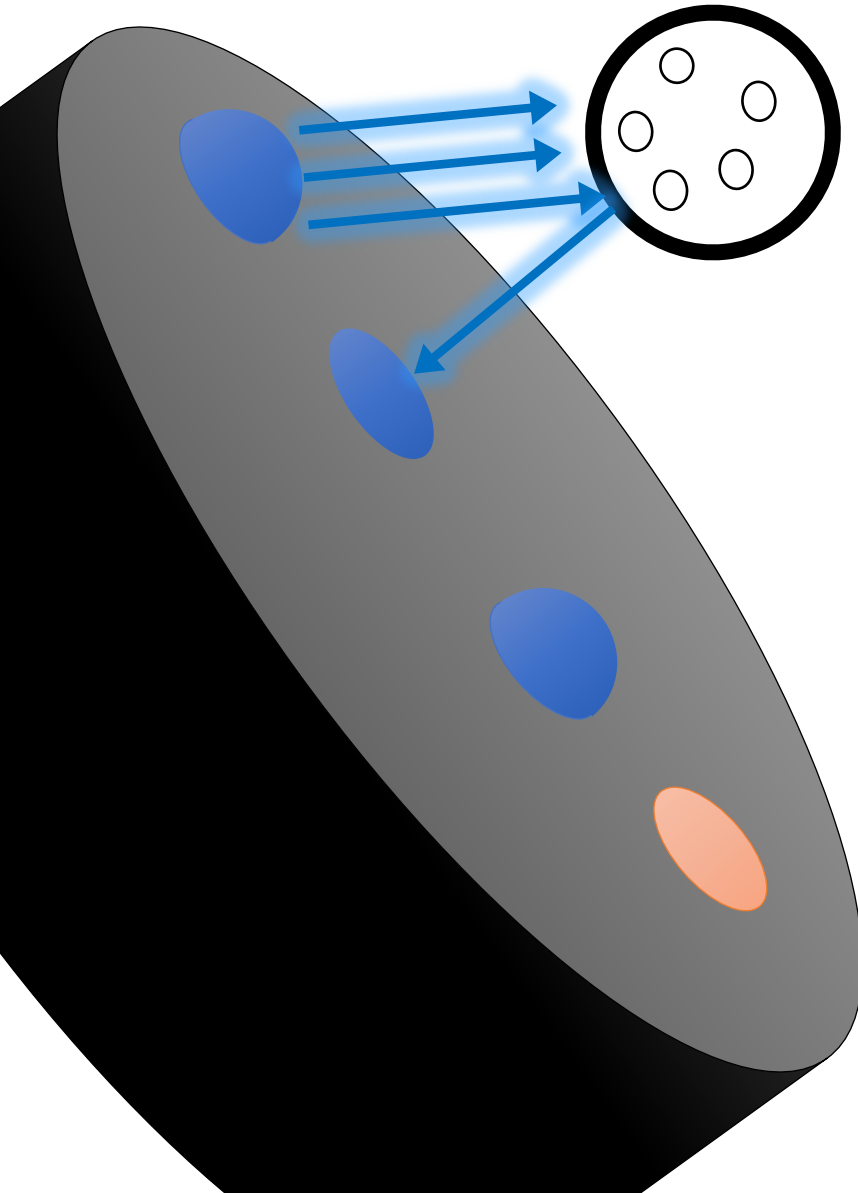
Calibrated value = (raw – dark) x scale factor



- black tape over detector only
- put in water to match refractive index
- average time record (~30 s)

# First way to calibrate optical backscattering sensors:

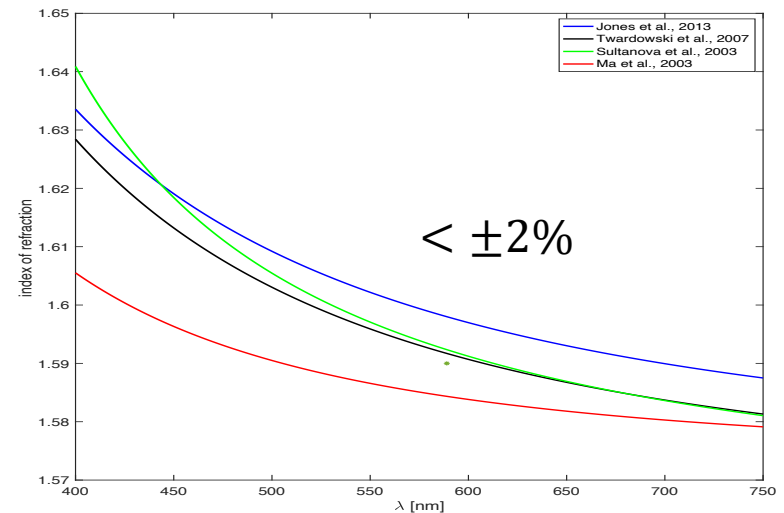
## Mie theory



Use NIST-traceable calibration beads:



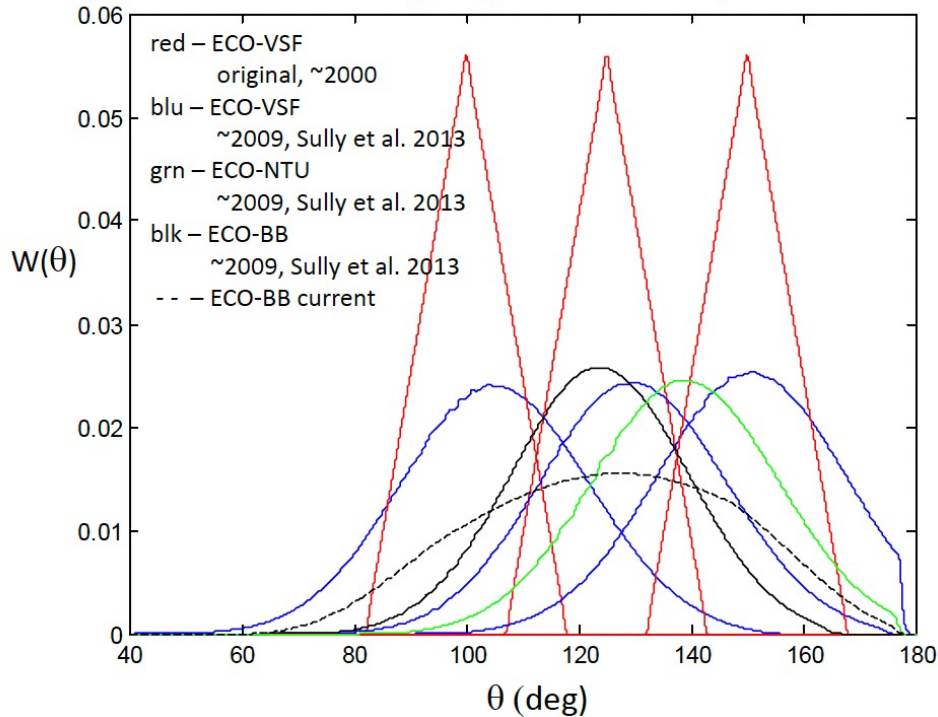
With known optical properties:



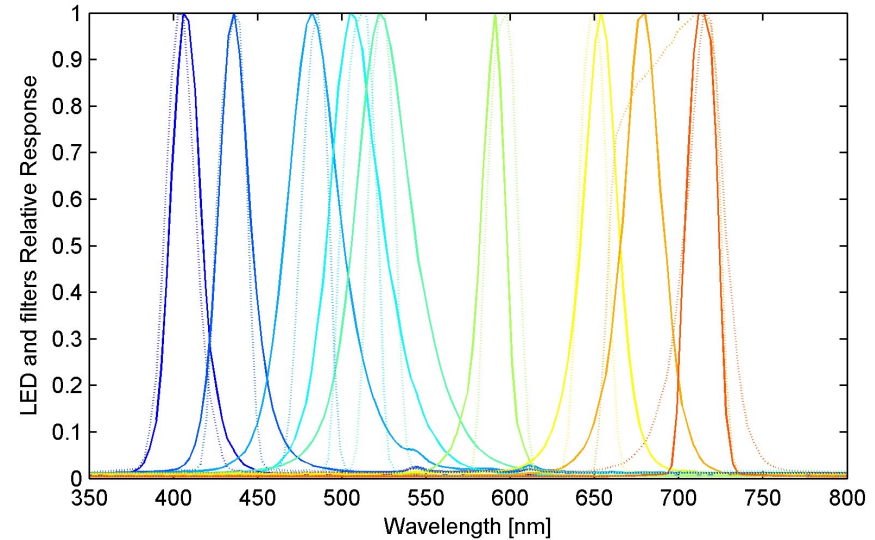
# What else do we need to know to calibrate?

Angular response (theoretical):

ECO weighting function history



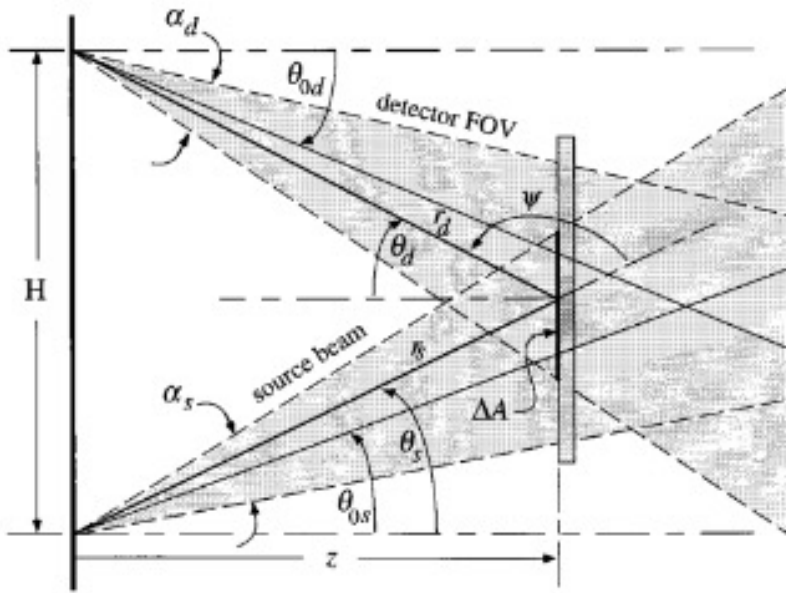
Wavelength response:



Bottom line: significant departures from nominal values.  
Likely varies between sensors and as function of time.

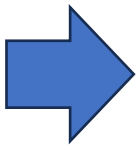
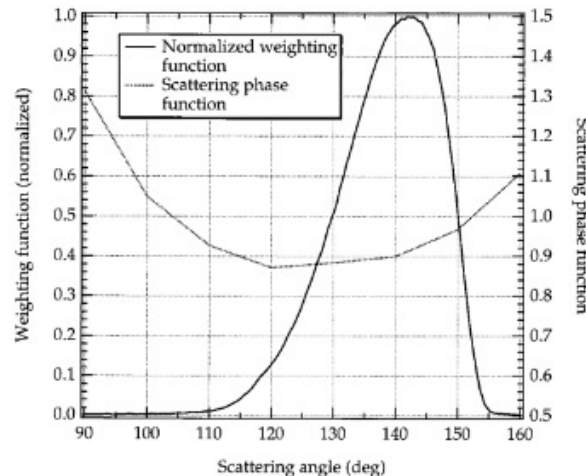
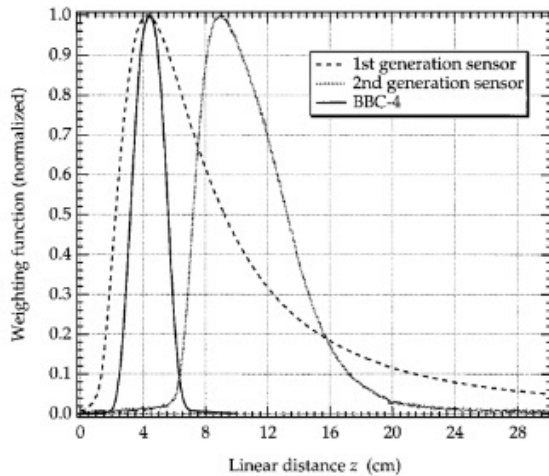


# 2nd way to calibrate optical backscattering sensors:



Need:  
 Reflectance of plaque as  $f(\lambda)$ .  
 NIST traceable.

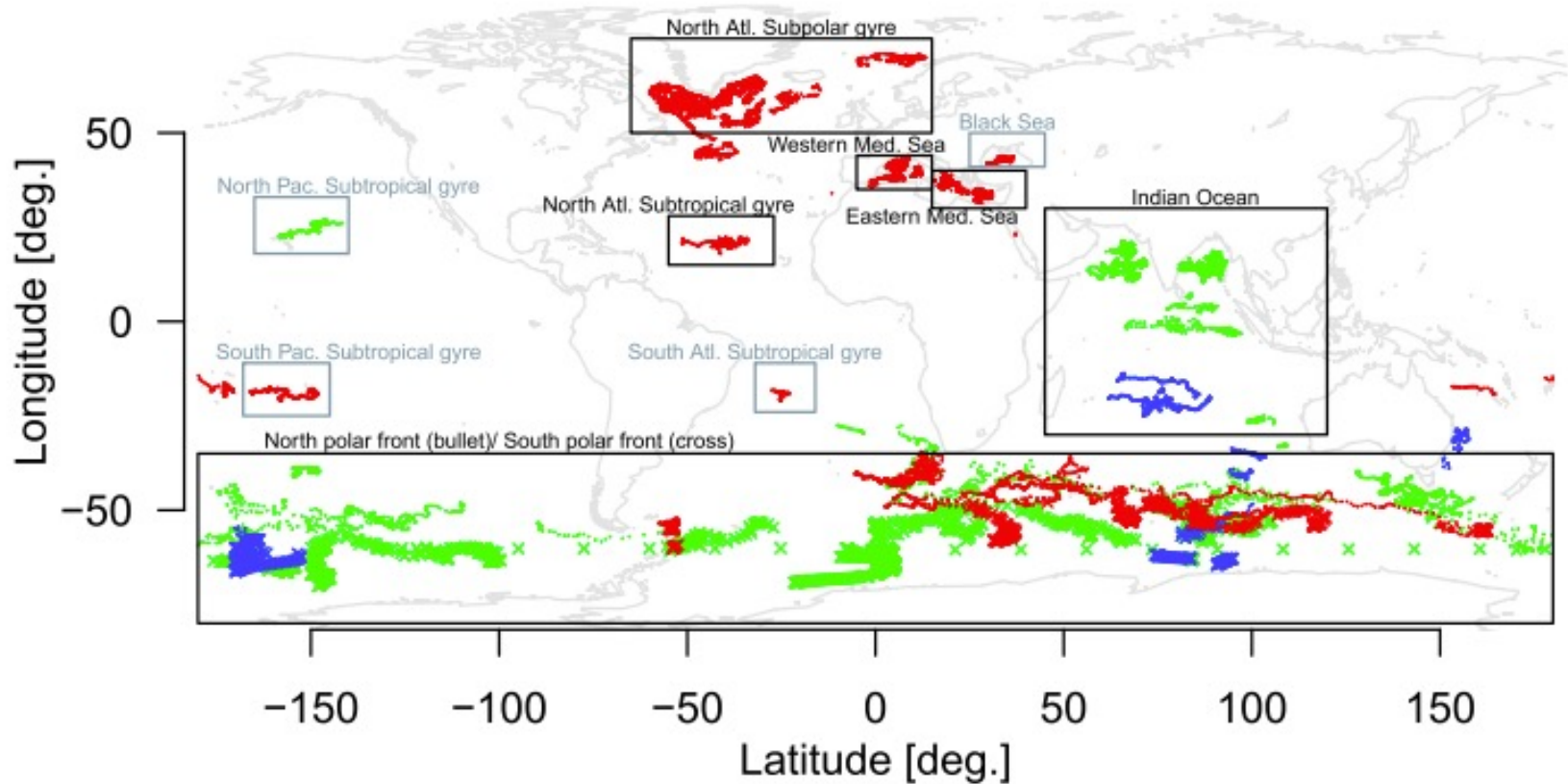
Fig. 5. Optical geometry for analyzing the sensor response to a Lambertian target.



# VERIFICATION

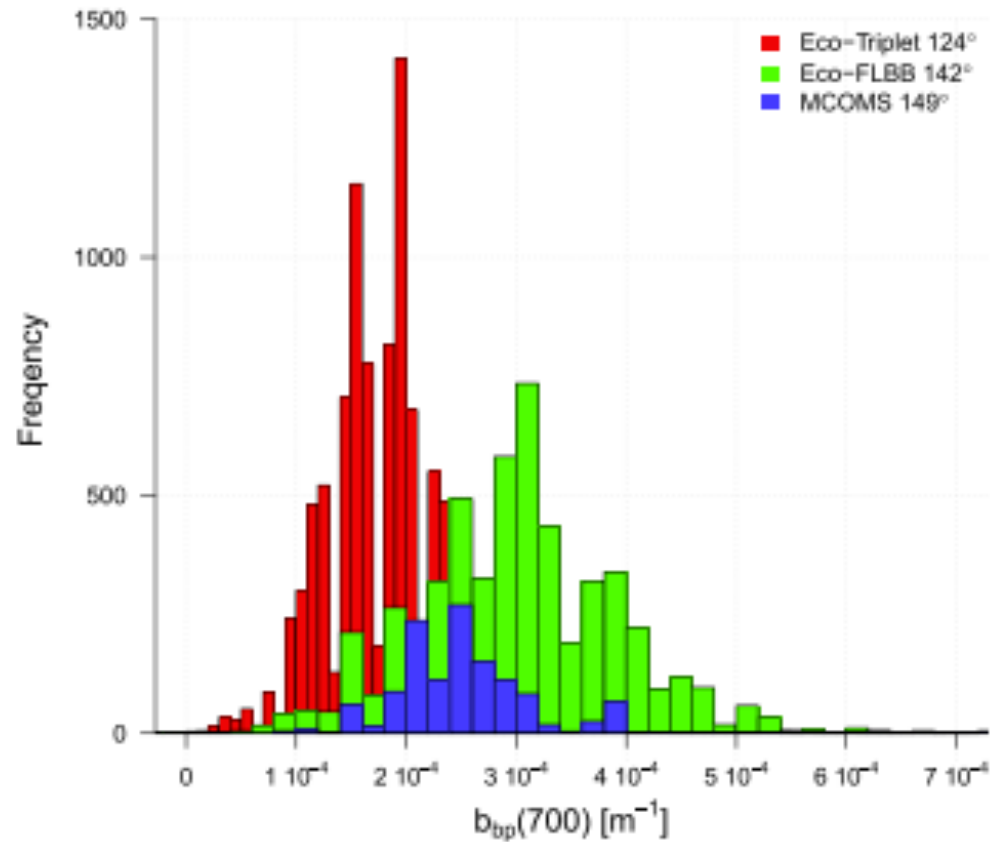
- Verification and validation (also abbreviated as V&V) are independent procedures that are used together for checking that a product, service, or system meets requirements and specifications and that it fulfills its intended purpose.
- Validation is similar to closure – we try to arrive at the same result using different means. Uncertainties are typically large.
- Verification is similar to ‘cross-calibration’, e.g. using an independent bead, plaque or other instrument to check the degree of agreement between calibrated sensors.

# Example 1: VERIFICATION – Poteau et al., 2017 – BGC Argo ( $b_{bp}(700nm)$ , 3 types of sensors)



**Figure 1.** Geographical distribution of all  $b_{bp}(700nm)$  data analyzed in this study. They are colored based on the technology used, Eco-Triplet 124° in red, Eco-FLBB 142° in green, and MCOMS 149° in blue. Black rectangles designate zones in which sufficient floats and profiles exist to perform zonal analysis. The Southern Ocean was further separated into two zones, north and south (identified by crosses) of the Polar Front.

# Example 1: VERIFICATION – Poteau et al., 2017 – BGC Argo (values @ 900-950m)



**Figure 2.** Histogram of median  $b_{bp}(700)$  measured with each type of sensor for all profiles listed in Table 1.

# Example 1: VERIFICATION – Poteau et al., 2017 – BGC Argo.

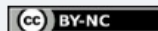
## Correction of scale factors for backscattering channel on ECO sensors mounted on BGC-Argo floats

DATE	2021-02-18
AUTHORS	Barnard Andrew <sup>1</sup>
AFFILIATIONS	1. Sea-Bird Scientific, Research and development department
DOI	10.17882/54520
PUBLISHER	SEANOE

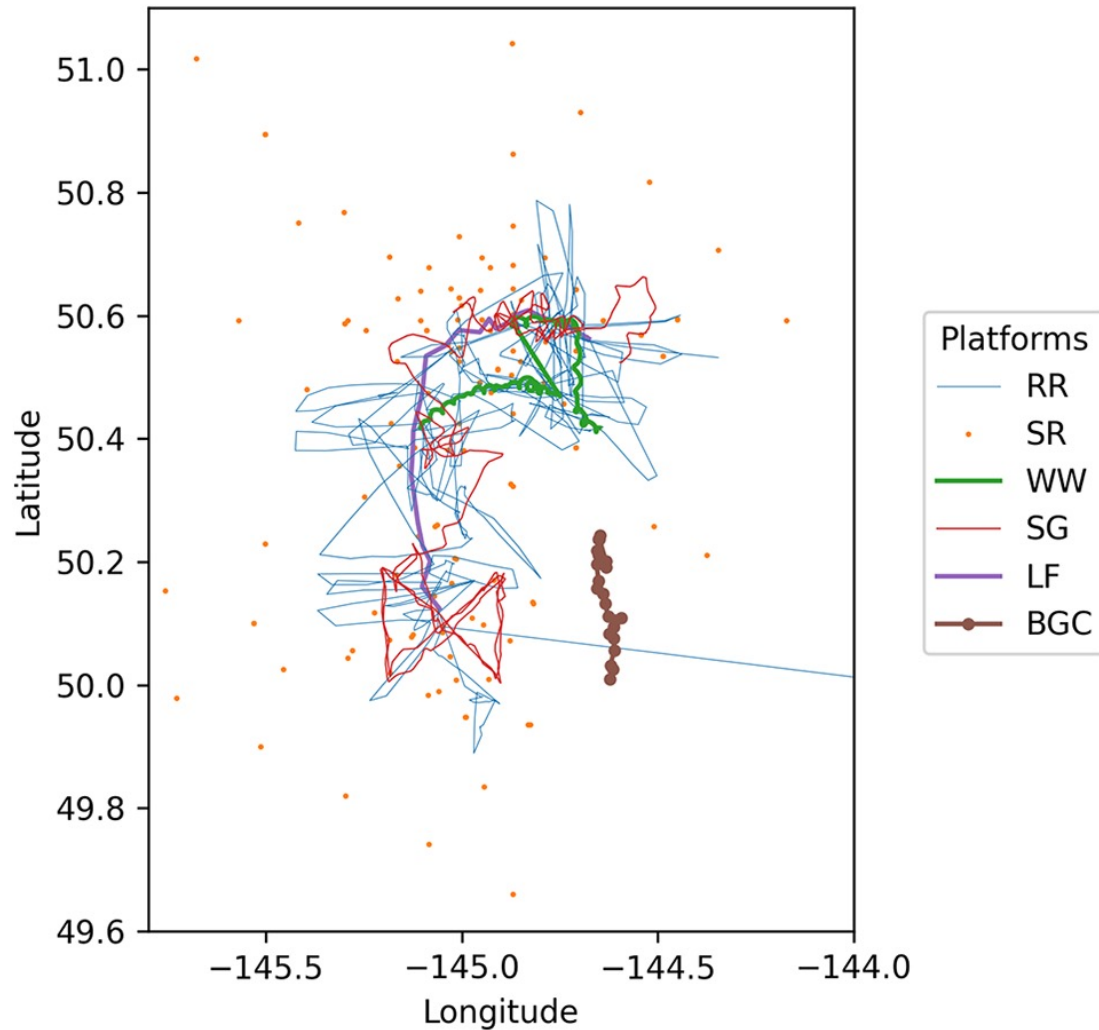
WET Labs investigated the bias found in Poteau et al. 2017: <http://dx.doi.org/10.1002/2017gl073949> and provides a matrix of affected sensors with scale factors for the backscattering channels using a correct weighted phase function constant values for ECO sensors mounted on BGC-Argo floats.

DISCIPLINES	Biological oceanography, Physical oceanography
LOCATION	90N, -90S, 180E, -180W

LICENCE

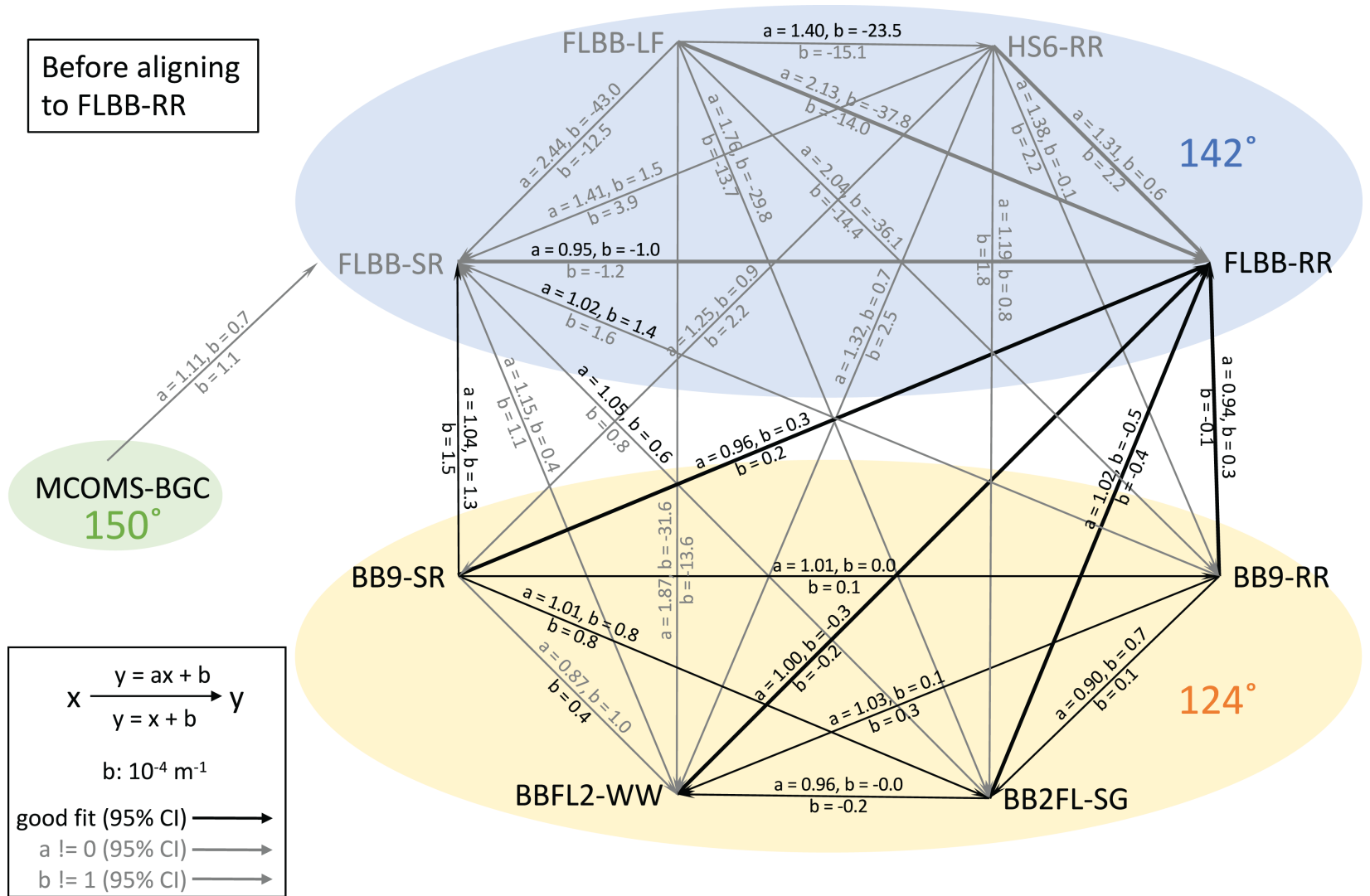


## Example 2: VERIFICATION – Erikson et al., 2022 – EXPORTS NP



Many platform compared at different times and locations.

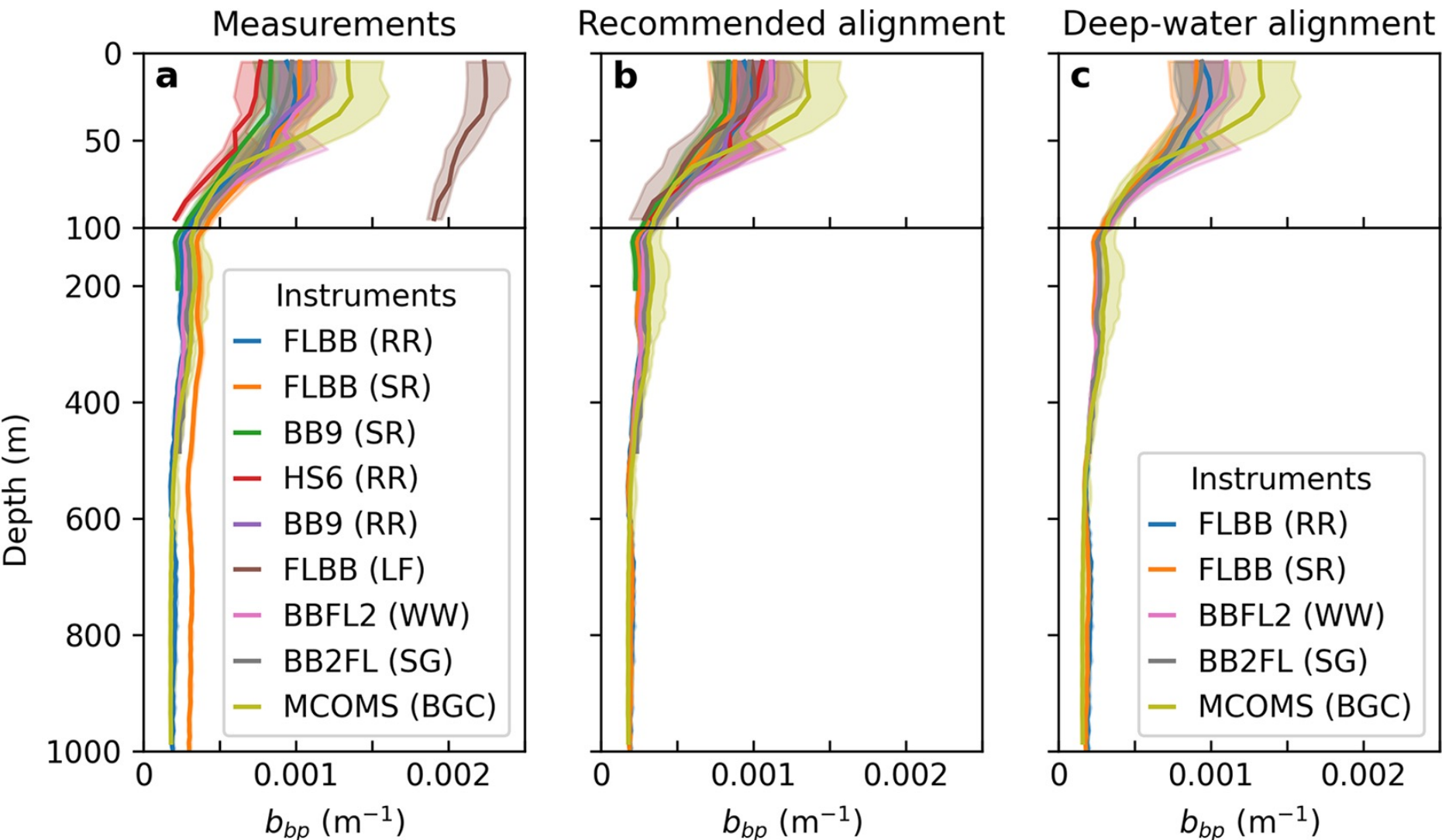
# Example 2: VERIFICATION – Erikson et al., 2022 – EXPORTS NP



Bottom line: much larger uncertainties than expected!



# Example 2: VERIFICATION – Erikson et al., 2022 – EXPORTS NP



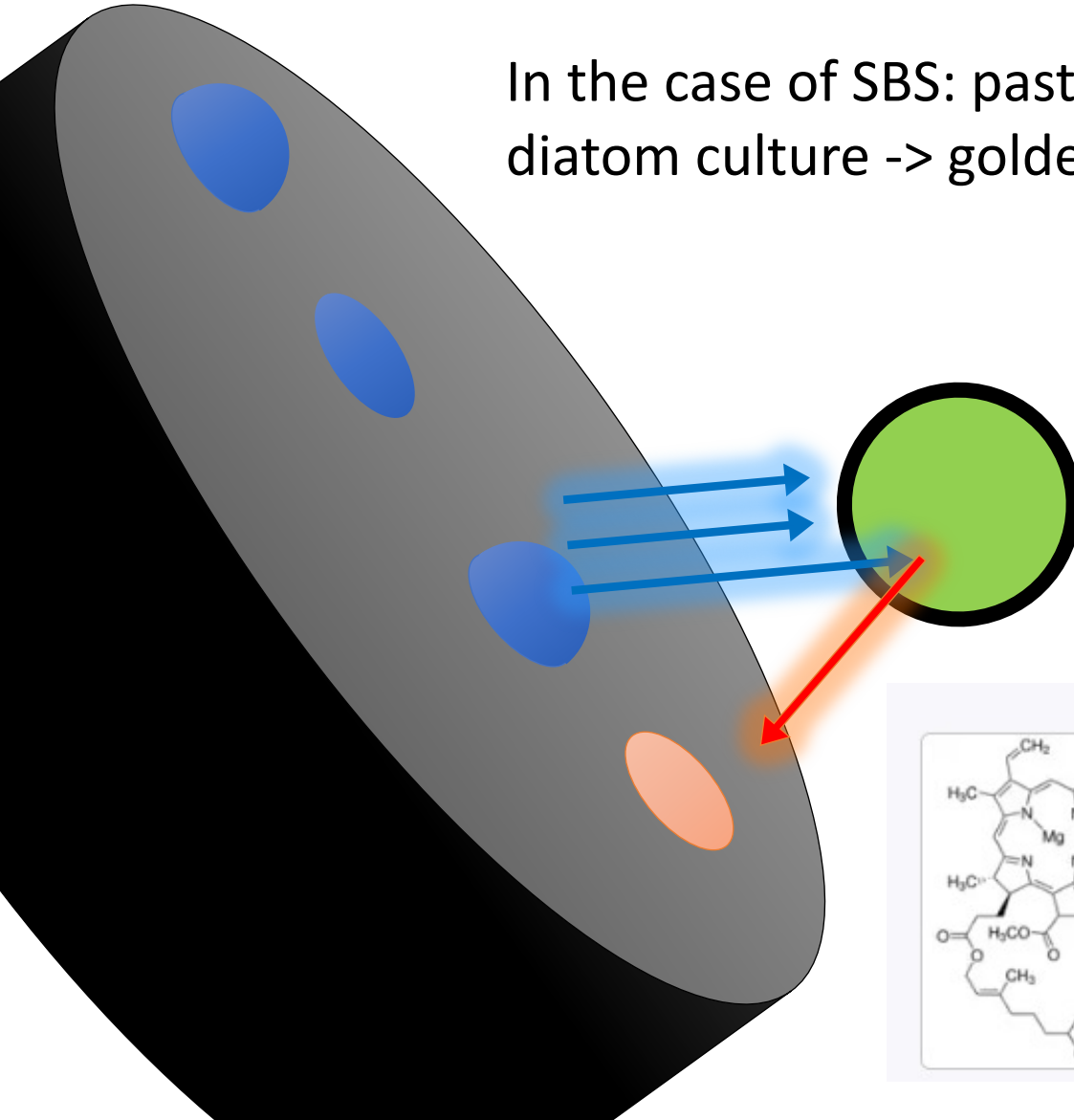
Bottom line: much larger uncertainties than expected!



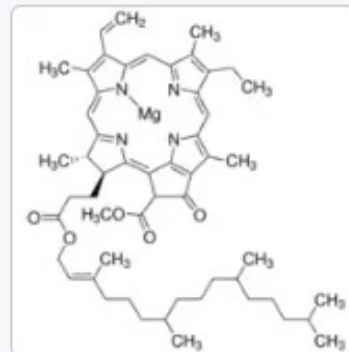
# How are fluorometer calibrated?

Standards: Purified chlorophyll from spinach.

In the case of SBS: past data collected with a diatom culture -> golden sensor.



Chlorophyll  
fluorescence



C5753 ▶ **Sigma-Aldrich.**

**Chlorophyll a**

★★★★★ (0) [Write a review](#)

from spinach

Empirical Formula (Hill Notation):

$C_{55}H_{72}MgN_4O_5$

# How are fluorometer calibrated?

## What are the advantages and disadvantages of using such a standard?

- Advantage: same model sensors calibrated similarly on land will provide consistent output in the field (we can directly cross-compare output from sensors on different platforms).
- Disadvantage: not really the chlorophyll we are after.

Summary up to this point:

When you get data to analyze, DO NOT ASSUME IT IS GOOD DATA.

Let the data convince you first it is usable by subjecting it to different consistency checks (we call these validation exercises - closure).

Do not assume stated uncertainties (e.g. in papers describing the calibration procedure) account for everything. Beware of unknown unknowns.

*But why do we use these sensors in the first place?*

- Typically, we are interested in ocean biogeochemistry.
- We use the sensors to interpolate between biogeochemical measurements within a cruise.

Parameters:

$b_{bp}$ : POC,  $C_{phyto}$ , TSM.

$F_{chl}$ : [Chl *a*], physiology (quenching).

Ratio: growth rate (NPP), physiology.

*What are the 'standards' to calibrate sensors with to obtain biogeochemical parameters with?*

With backscattering sensors:

POC – issue – what is the blank (DOC adsorbs to the filter and affects reading)?

$C_{\text{phyto}}$  – sorting FCM vs. FCM + assumptions regarding carbon and volume of phytoplankton.

For Chlorophyll a:

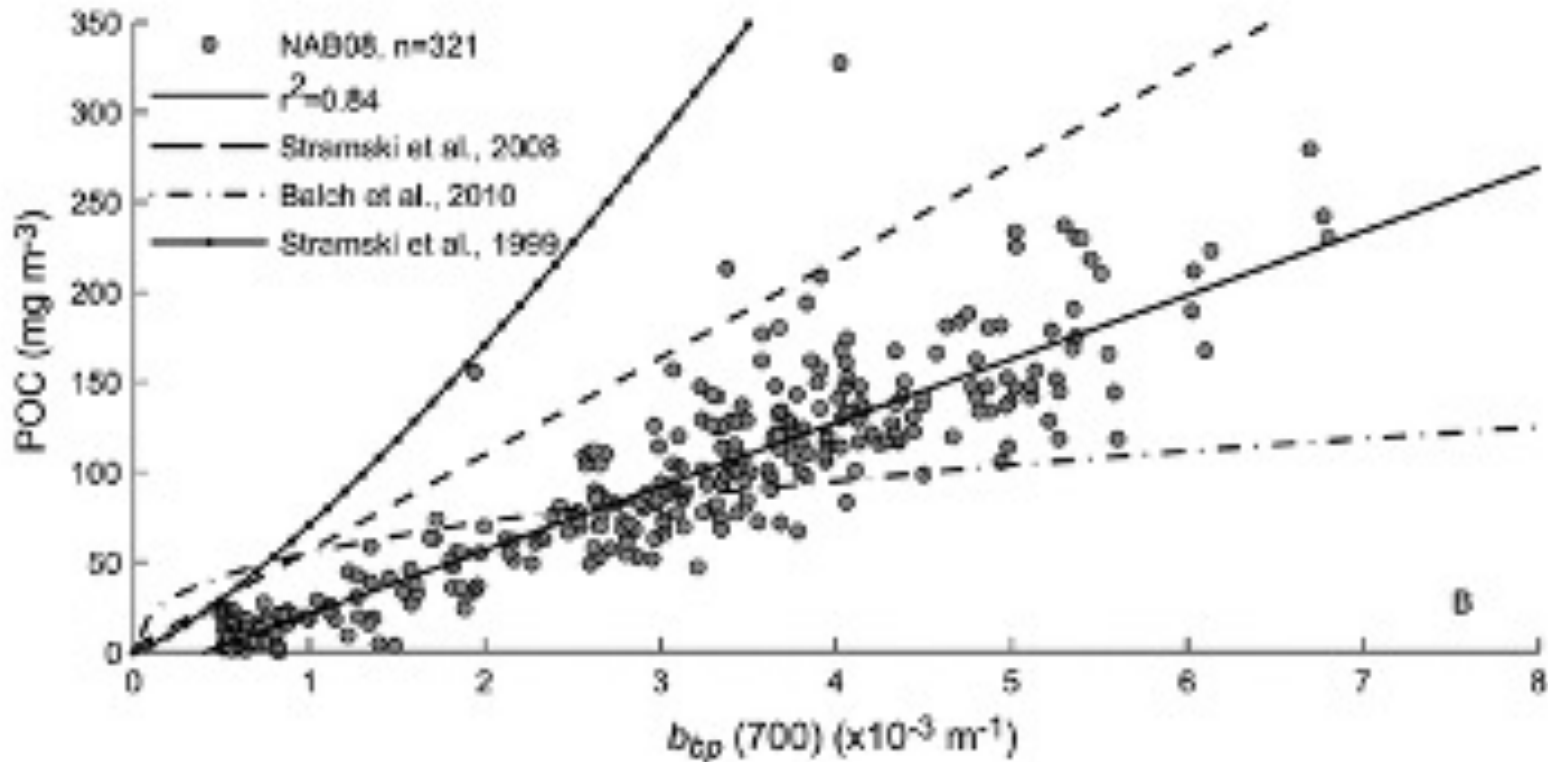
HPLC, fluorometric (Turner), absorption-based, and photometric methods.

*What are sources of uncertainties associated with these standards?*

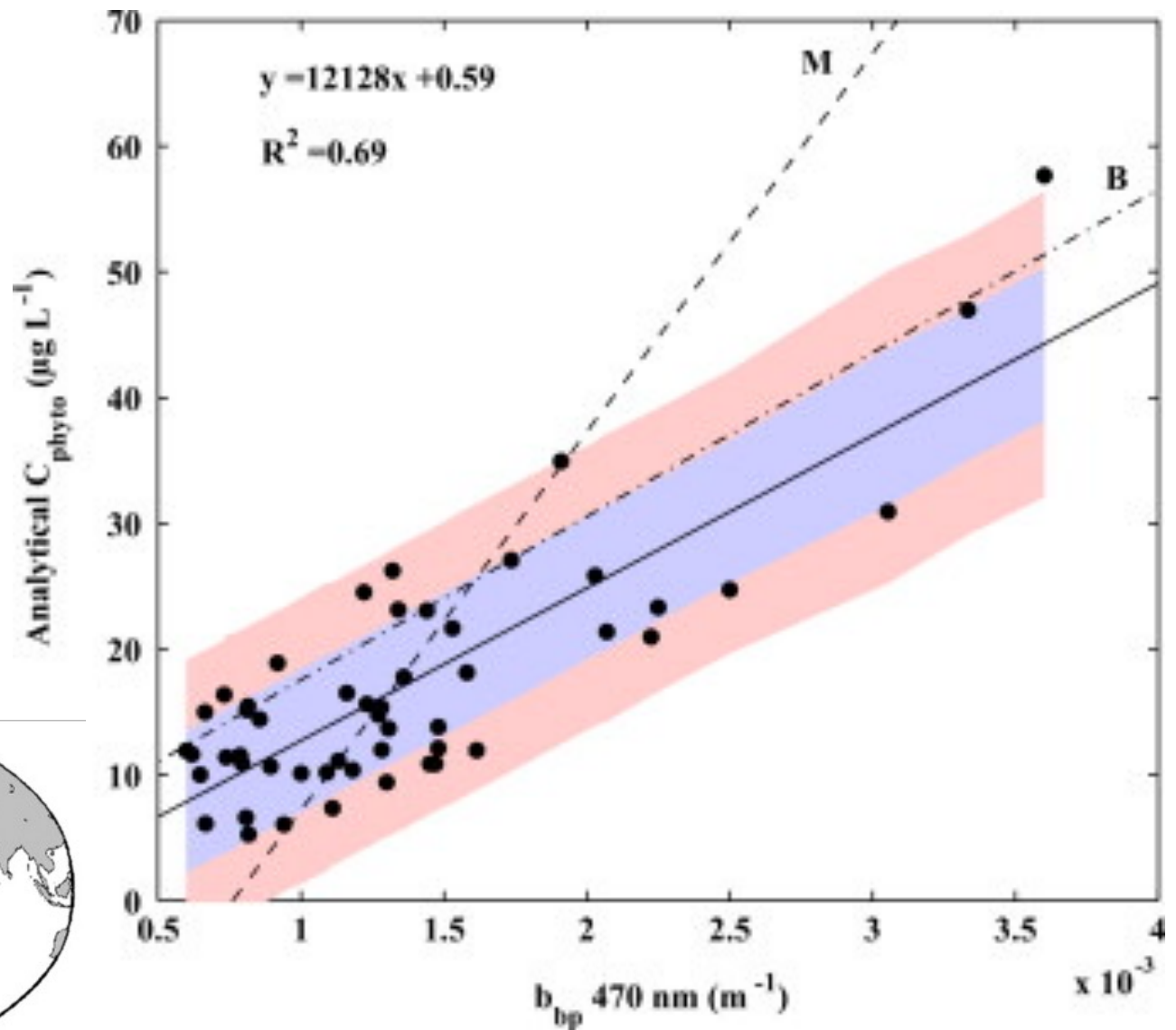
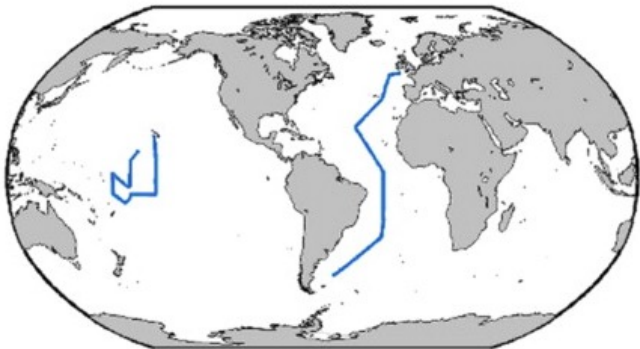
1. WRT to  $b_{bp}$ : Variability in assembly in terms of  $C_{phyto}/POC/TSM$ .
2. WRT to chlorophyll – variability between fluorescence and pigments.
3. Where is the data coming from? How representative is it in space and time? Extrapolation vs. interpolation (particularly wrt AI).

# Some examples:

$b_{bp}$  to POC:



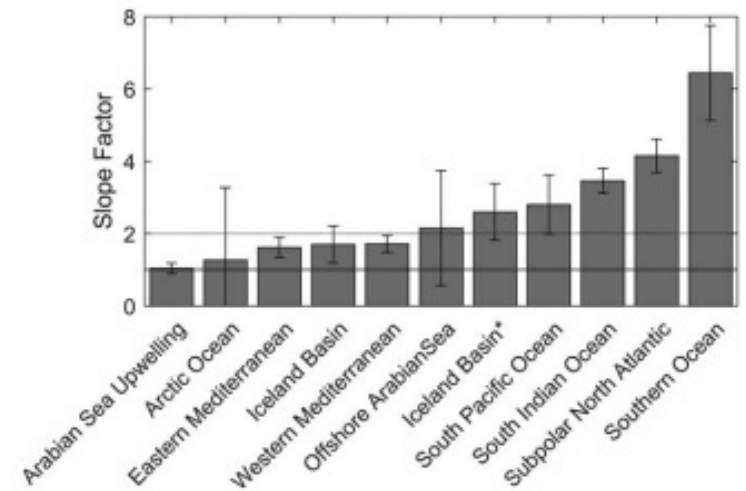
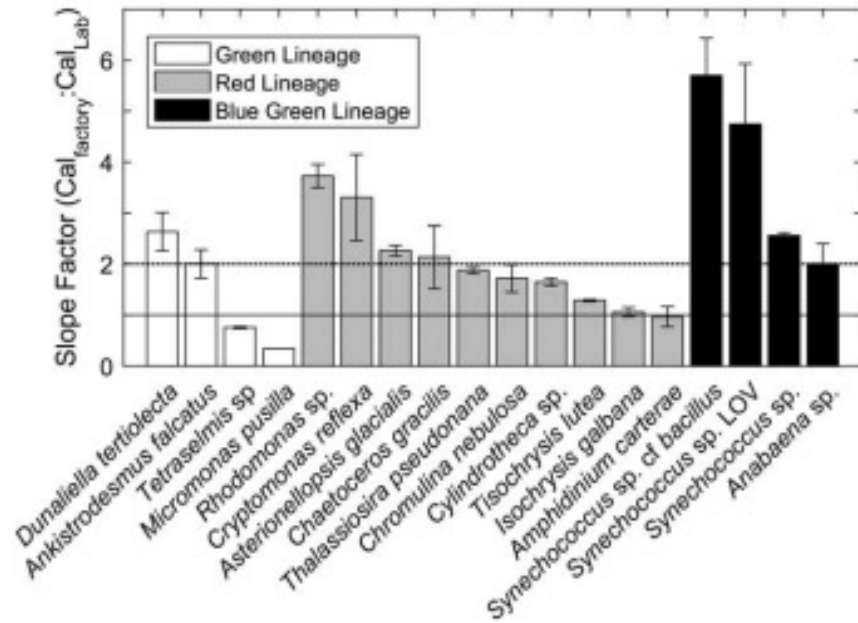
$b_{bp}$  to  $C_{phyto}$ :



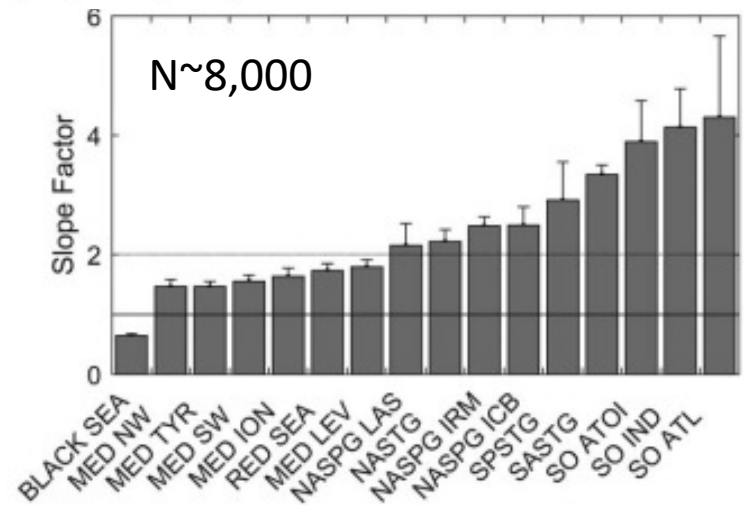
Graff et al., 2015



# $F_{chl}$ to Chl:

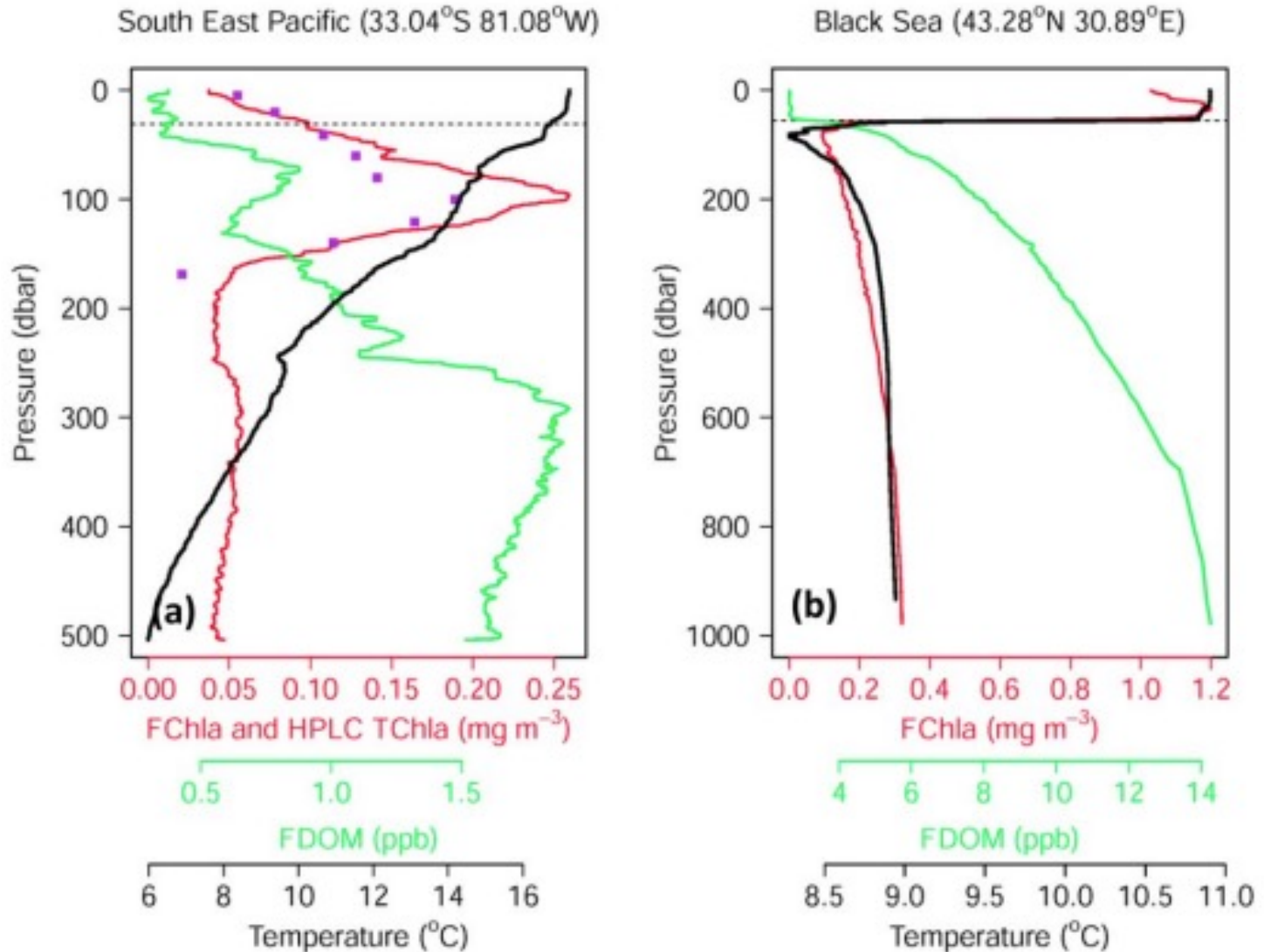


**Fig. 2.** Mean slope factors derived from observations of paired HPLC and in situ Chl fluorescence from major oceanographic regions (Table 1). Error bars indicate 95% confidence limits on slope from linear regression of all observations within each region. Lines indicate slope factors of 1 (solid) and 2 (dotted).



**Fig. 4.** Mean slope factors derived from ratio of factory calibrated Chl fluorescence to radiometrically-derived Chl (see text for details) obtained from profiling biogeochemical Argo floats described in Table 2. Error bars indicate 95% confidence limits on slope derived from regression of all observations within each region. Lines indicate slope factors of 1 (solid) and 2 (dotted).

# FYI: Contamination of $F_{chl}$ by CDOM.



## Additional consistency check:

1. *What is your expectation for the value of  $C_{\text{phyto}}/\text{chl}$  in the upper ocean?*
2. *Below the euphotic depth?*
3. *What affects this ratio?*

Parting words:

A full accounting of uncertainties is a never ending task.

Bias detection is key – bias does not get smaller the more data we collect.

Be honest. Small error bars are not a sign of good science, but rather a sign of an optimistic scientist.

Validation/closure should be part of every method section of a paper you write.