How Algae Sensors Work

Principles and Practice in Water Quality Monitoring

Stephanie A. Smith, Ph.D.

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26 May 2020

How Sensors Work:
6-Part Series on Water Quality Monitoring

Once a week, we will discuss why it is important to monitor critical water quality parameters.

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May 26 - Tuesday

June 2 - Tuesday

June 9 - Tuesday

June 16 - Tuesday

June 23 - Tuesday

Dr. Stephanie A. Smith

BACKGROUND

Ph.D. in Microbiology
The Ohio State University

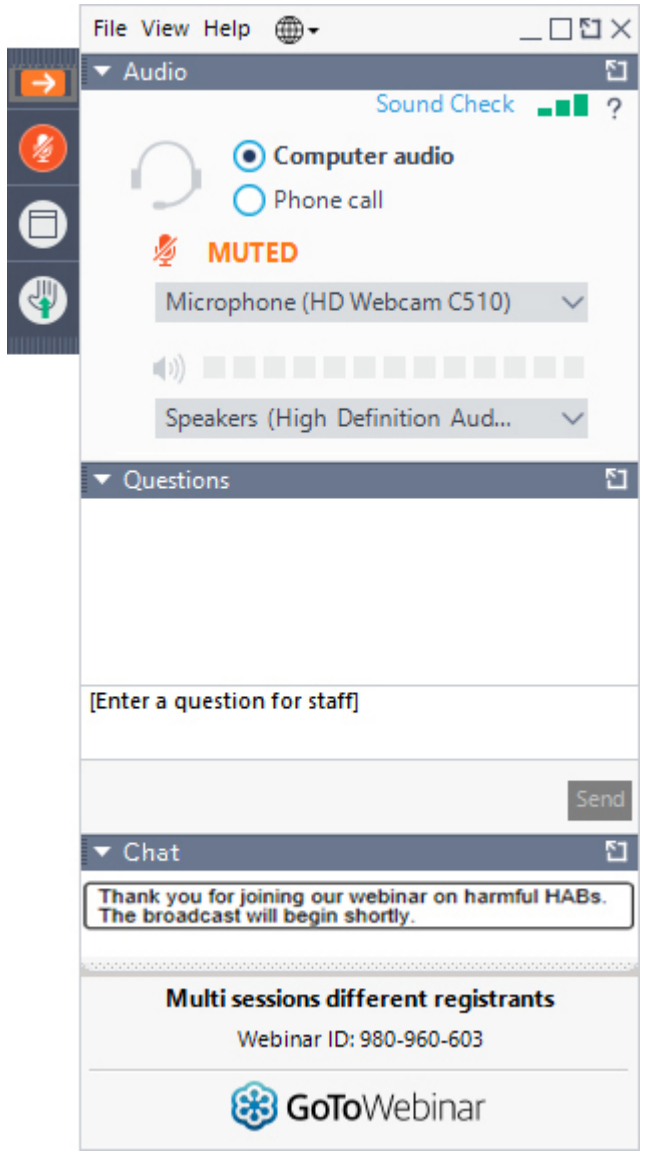
- Expert in blue-green algae and their toxins
- Product Segment Manager, YSI Environmental Solutions
- 20 years working with algae and other photosynthetic microbes

GoTo Webinar

Audio Settings
Make sure you can hear us loud and clear

Ask Questions
We'll try to answer as many as we can during the presentation

Chat
You can also use the Chat panel to ask questions or contact us if you're having technical difficulties



Modify Audio Settings

Please Ask Questions!

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Overview

I. Why Monitor for Algae?
II. Evolution of Algae Monitoring
III. How Algae Sensors Work: Principles
IV. How Algae Sensors Work: Best Practices

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Why Monitor for Algae?

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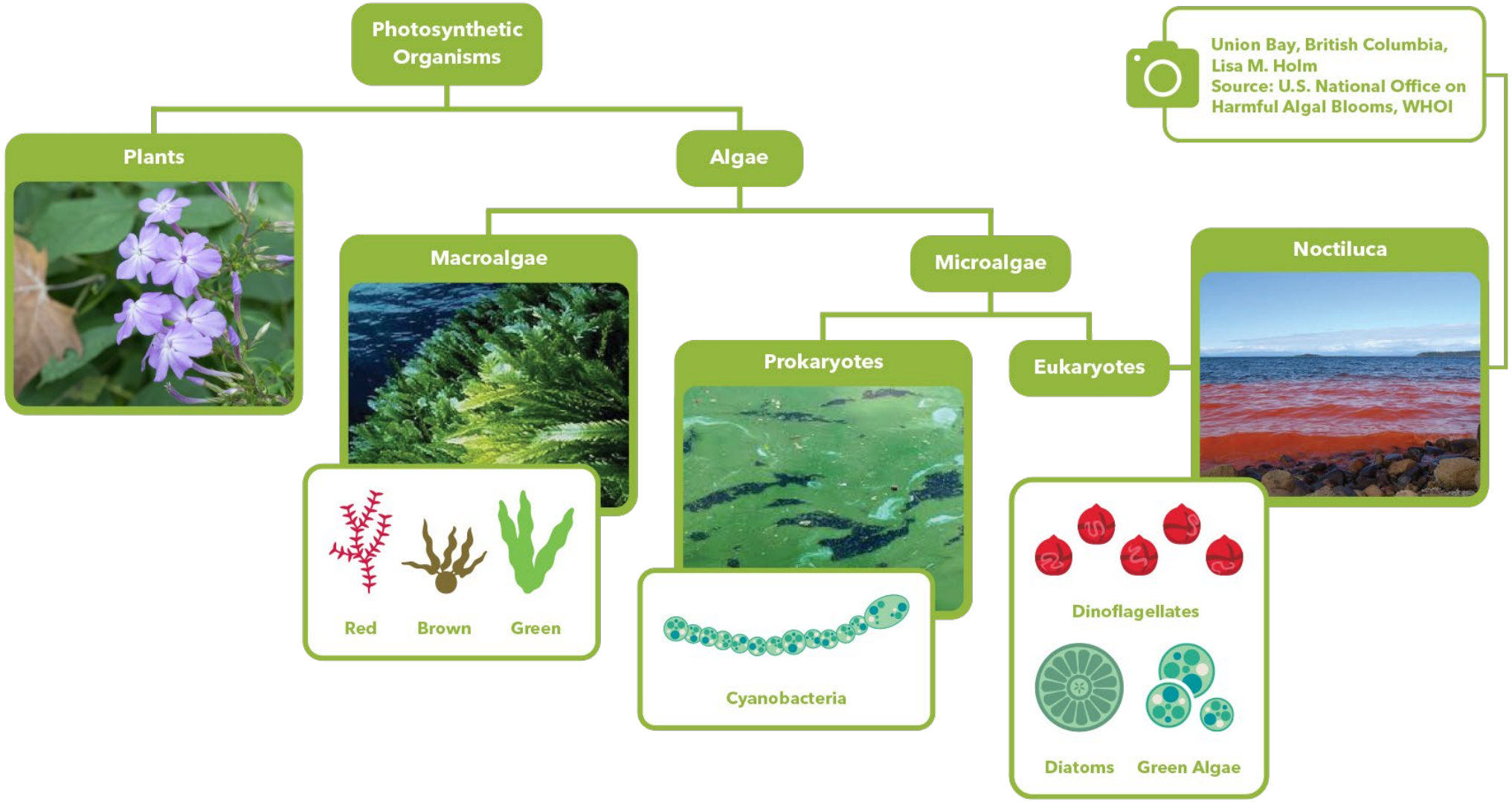
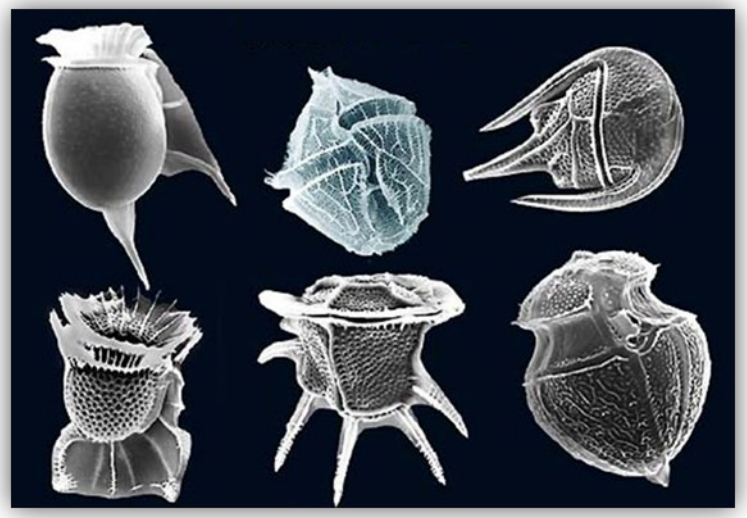
Why are you interested in algae sensors?

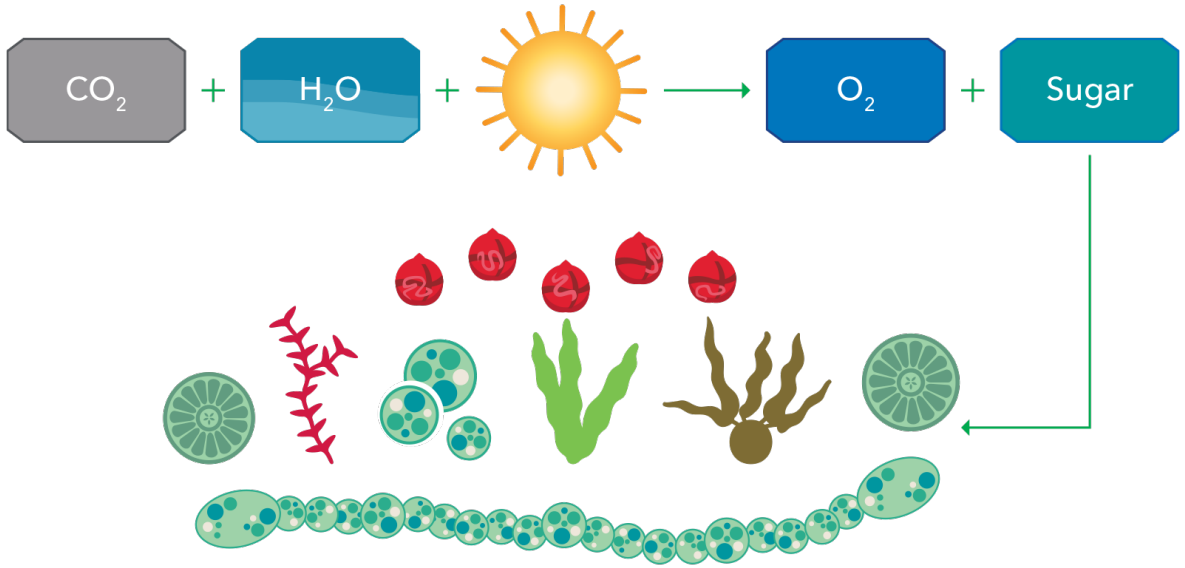
Why Monitor for Algae?



Why Monitor for Algae?

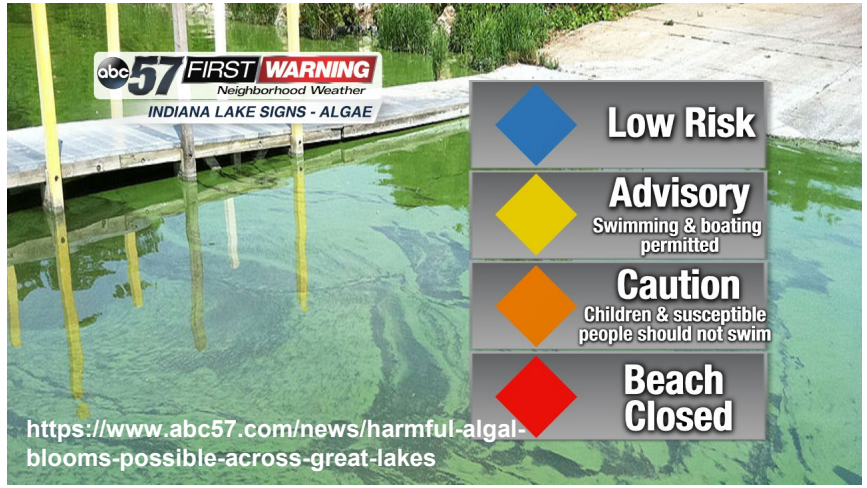
1. Aquatic Ecology Research
   - Primary productivity
   - Ecosystem health and dynamics
   - Interest in types and abundance





Why Monitor for Algae?

1. Aquatic Ecology Research
2. Source Water Protection
   - Harmful Algal Blooms
   - Drinking Water Treatment
   - Interest in changes and products



https://oeconline.org/waters-out-of-whack/

Oregon's state capital was under a drinking water advisory for **1 month**

3 people fell ill from recreating in Lake Billy Chinook during a harmful algae bloom

32 cows died from drinking out of a toxic algae filled pond

4 dogs died from playing in the water on the South Umpqua River

Why Monitor for Algae?

- The scientist: “Has the population of algae changed from ‘healthy’ to ‘unhealthy’ algae?”
- The Treatment Plant Operator: “How do I assure delivery of clean, safe drinking water?”
- The public: “Is it safe to swim here?”

Evolution of Algae Monitoring

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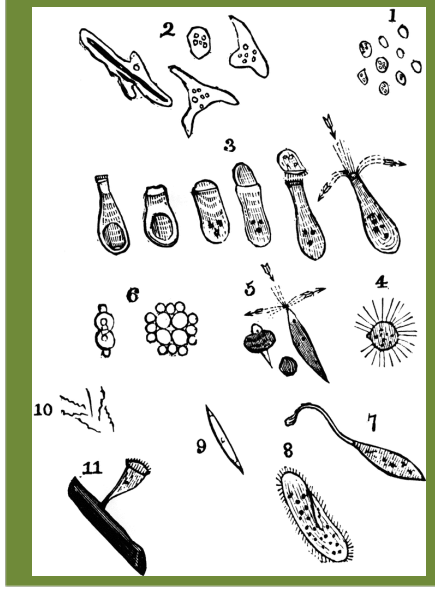
Have you used any of the following monitoring tools for algae?

Algae Monitoring Tools

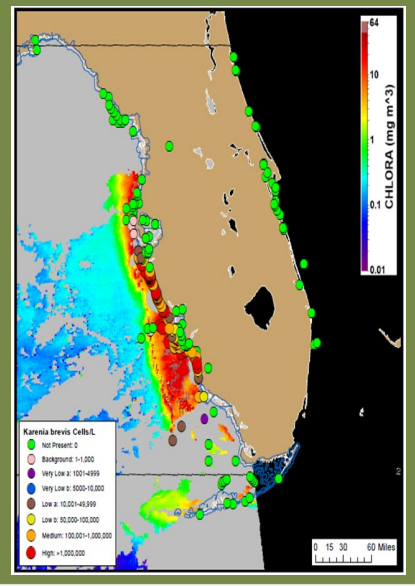
Microscopy



Chlorophyll Analysis

In vivo / in situ Sensors

Satellites



Online analyzers

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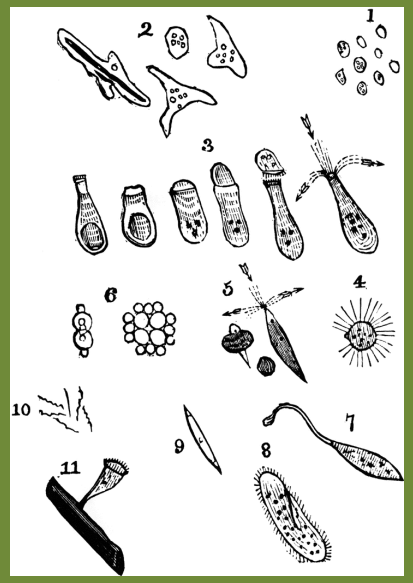
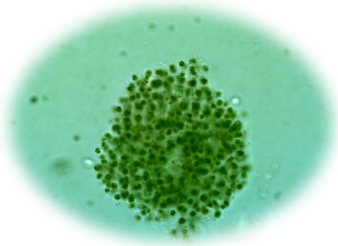
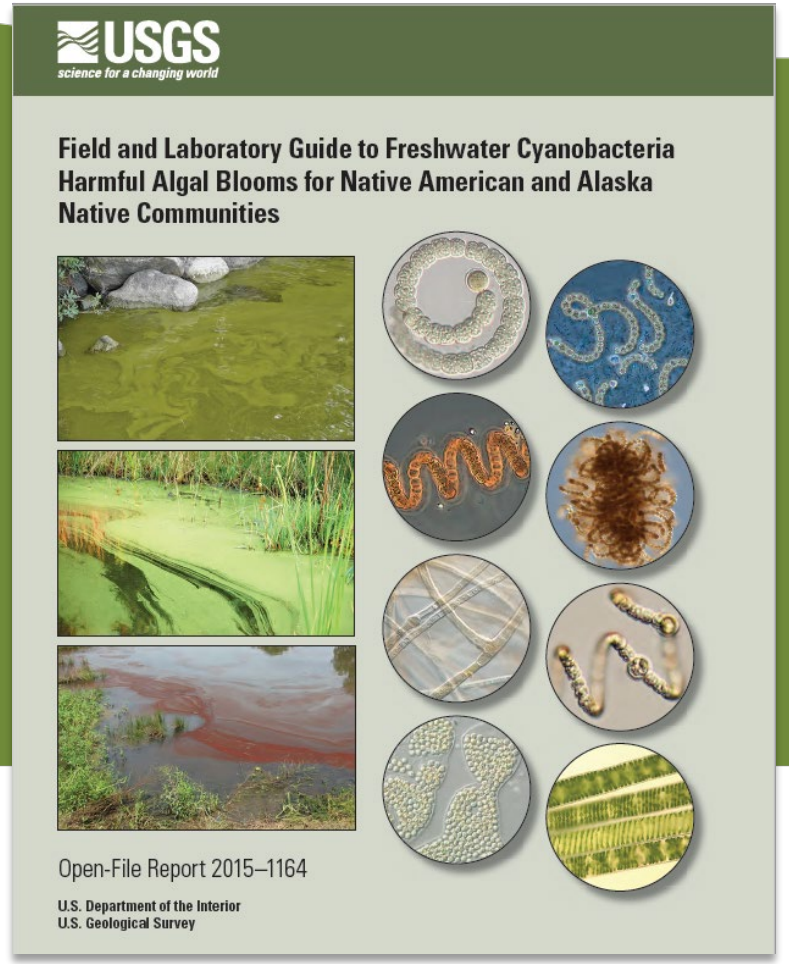
Microscopy



- Low cost, considerable skill
- Still most popular for speciation, enumeration (UOM: cells/mL)
- *Wide variability from one analyst to another*





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Chlorophyll Analysis

EPA Method 445.0:
Fluorometric Analysis

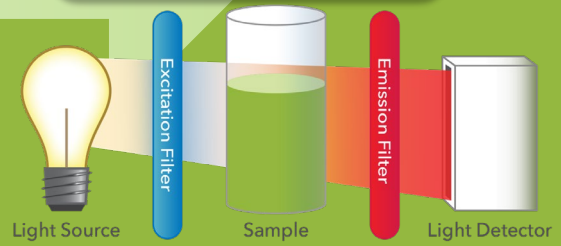
UOM: µg/L or ppb

Sample

Concentrate

Extract

Measure



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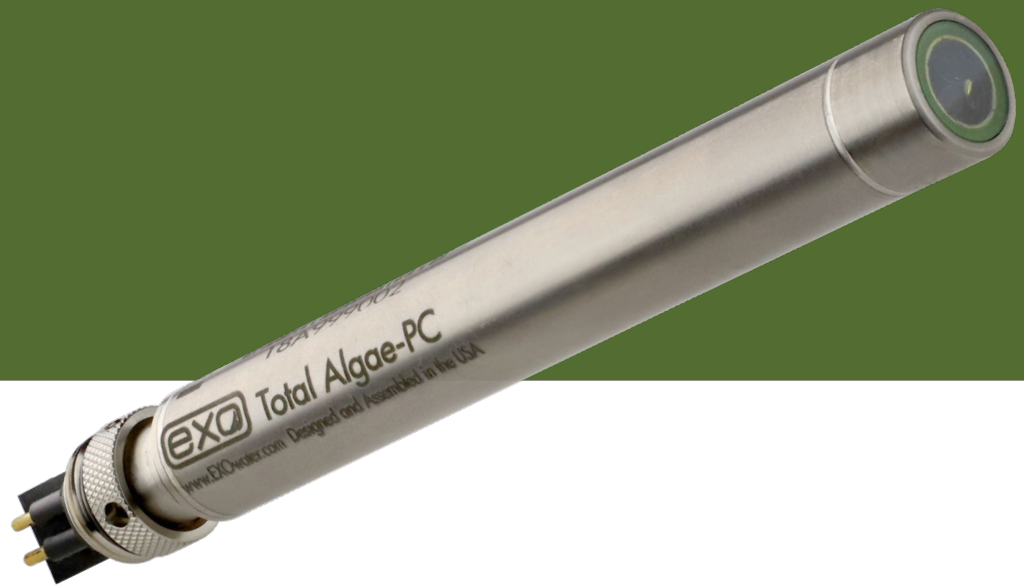
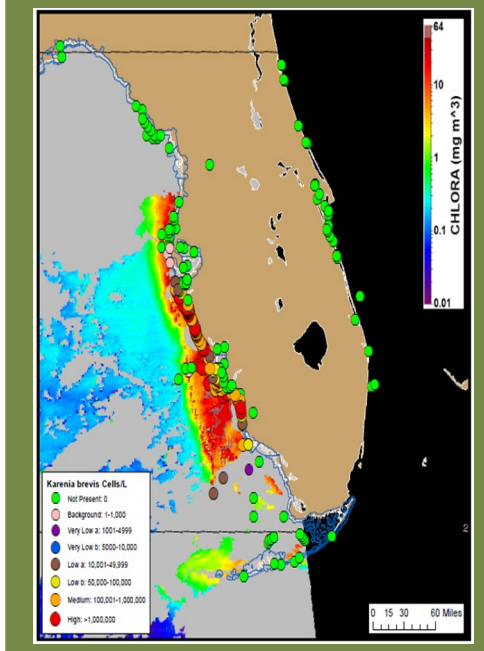
In vivo / in situ Sensors

- Fluorescence *measured directly in the water*
- Correlations with microscopy or extractions depend upon:
  - Population density
  - Population composition
  - Water Quality
  - Lab methods/technicians



Satellites



- Multiple satellites, multiple principles
- NOAA's 2017 Introduction to Remote Sensing of HABs
- NOAA's HAB Bulletins and Forecasts
  - Lake Erie
  - Gulf of Mexico
  - Dead Zone update 10 June 2019!

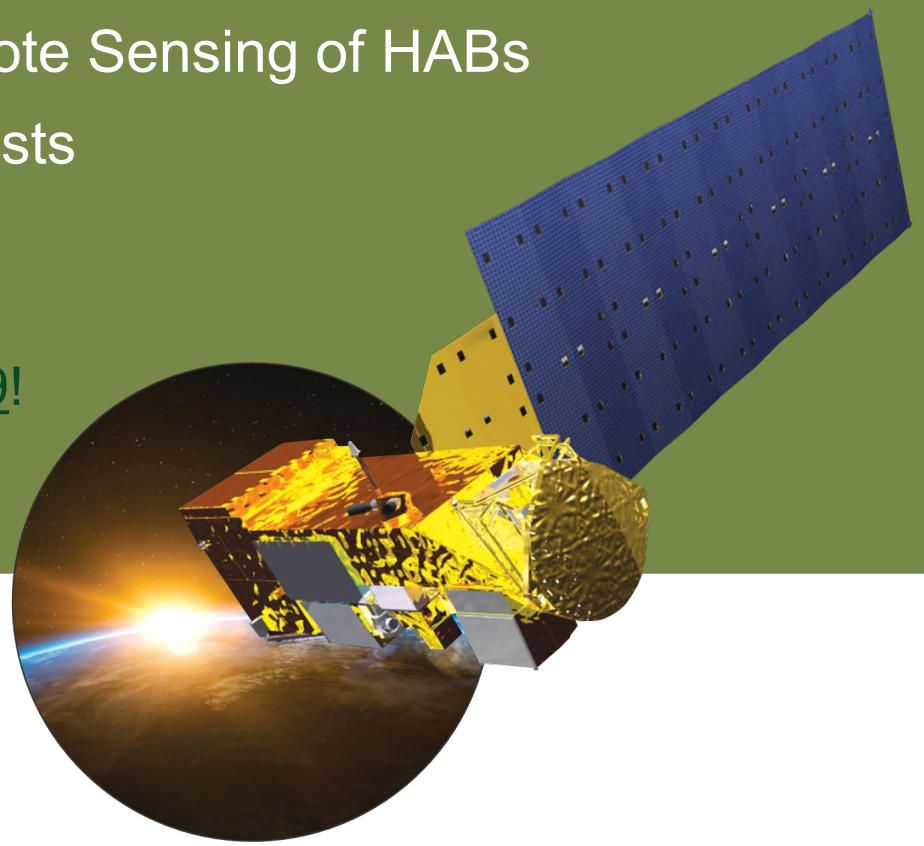
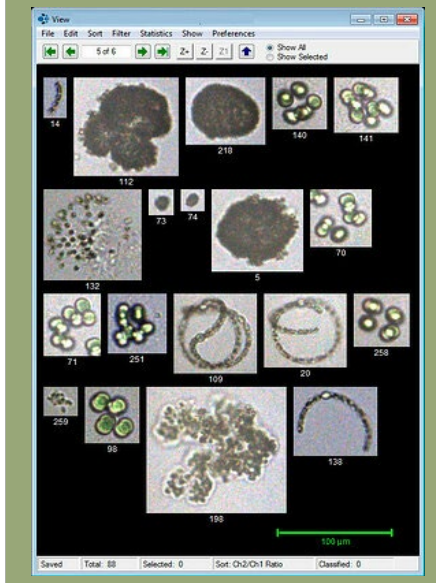


Image Analyzers



- Combines the best of all methods so far
  - Fluorescence-based detection
  - Speciation based on an image library
  - Population counts
- But, $$$

Evolution of Algae Monitoring: Summary

- The maturity of a monitoring tool is not what matters, the monitoring objective is
- The best monitoring programs will leverage a combination of tools
- The future: platforms and tools that provide multiple types of information, in real time
  - Multiparameter platforms
  - Fluid imaging

MISSION: water

ISSUE #6

From Cells To Satellites

Digital Download Now Available!

YSI.com/Mission-Water

How Algae Sensors Work: Principles

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What types of algae are you most interested in monitoring for?

Sensor Principles

I. Fluorescence
II. Fluorescent Pigments of Algae
III. How Algae Sensors Work

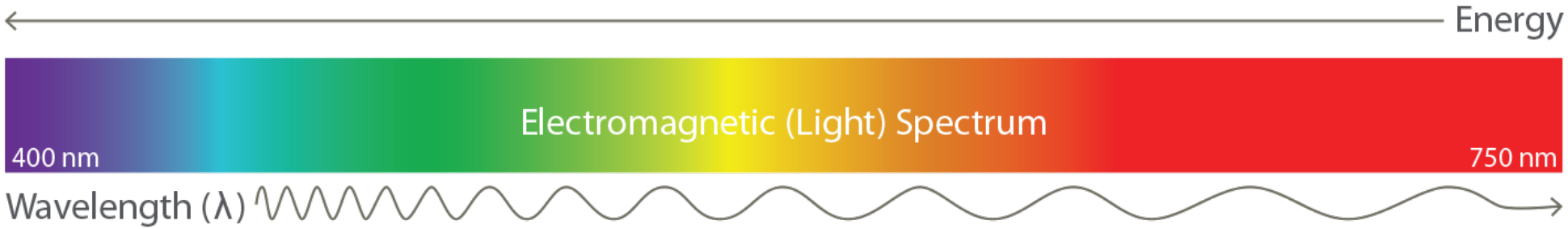
Fluorescence

Excitation

- A molecule absorbs light energy at a specific λ band
- The molecule is “excited” by the energy it has absorbed

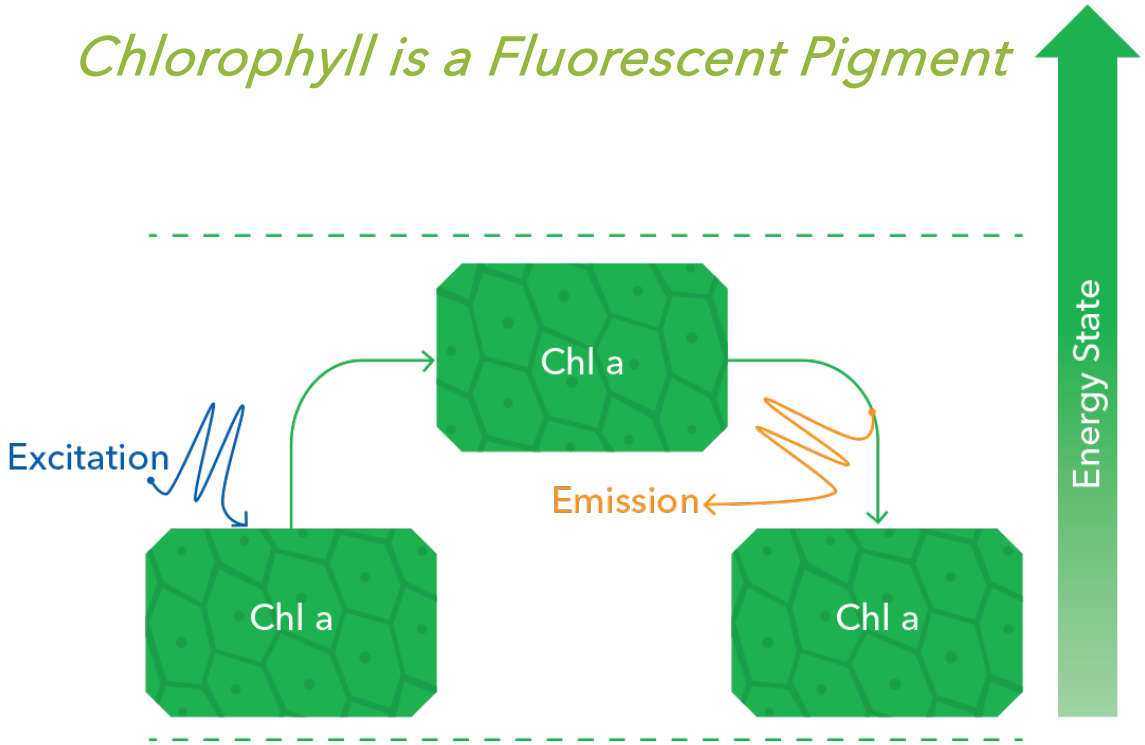
Emission

- The molecule returns to its original energy level by emitting light
- The emitted light is of a longer λ than the molecule originally absorbed (some energy was lost in the process)

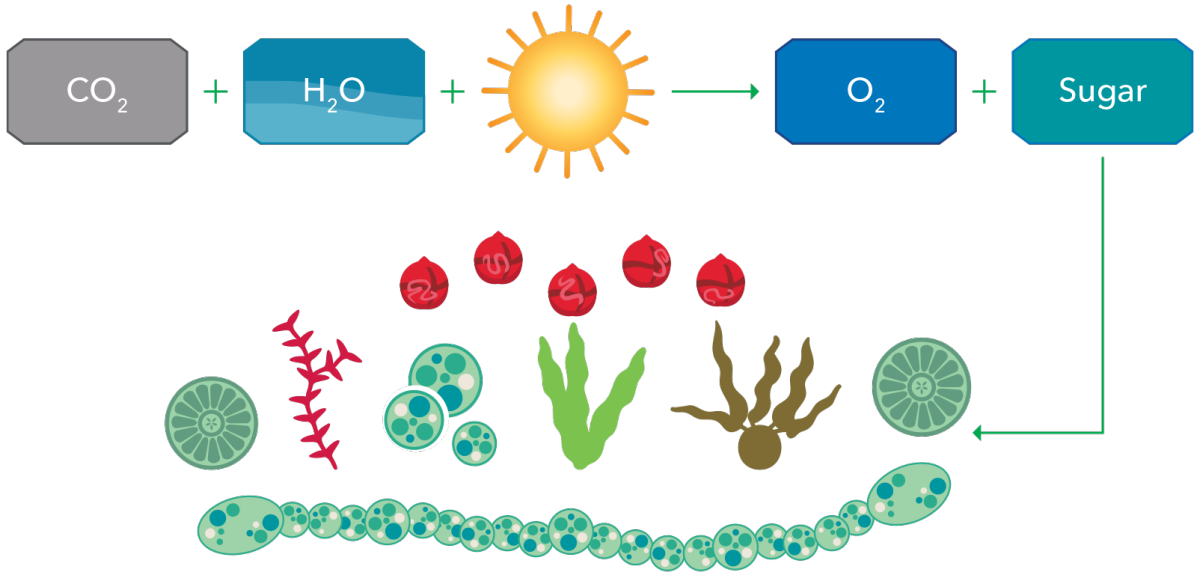


Fluorescent Pigments of Algae

Chlorophyll is a Fluorescent Pigment



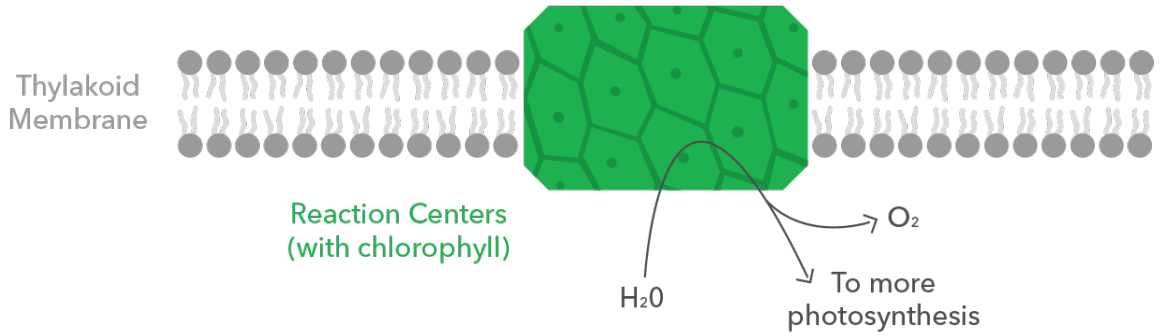
Algae Use Chlorophyll for Photosynthesis



Fluorescent Pigments of Algae

Chlorophyll

- *All* algae



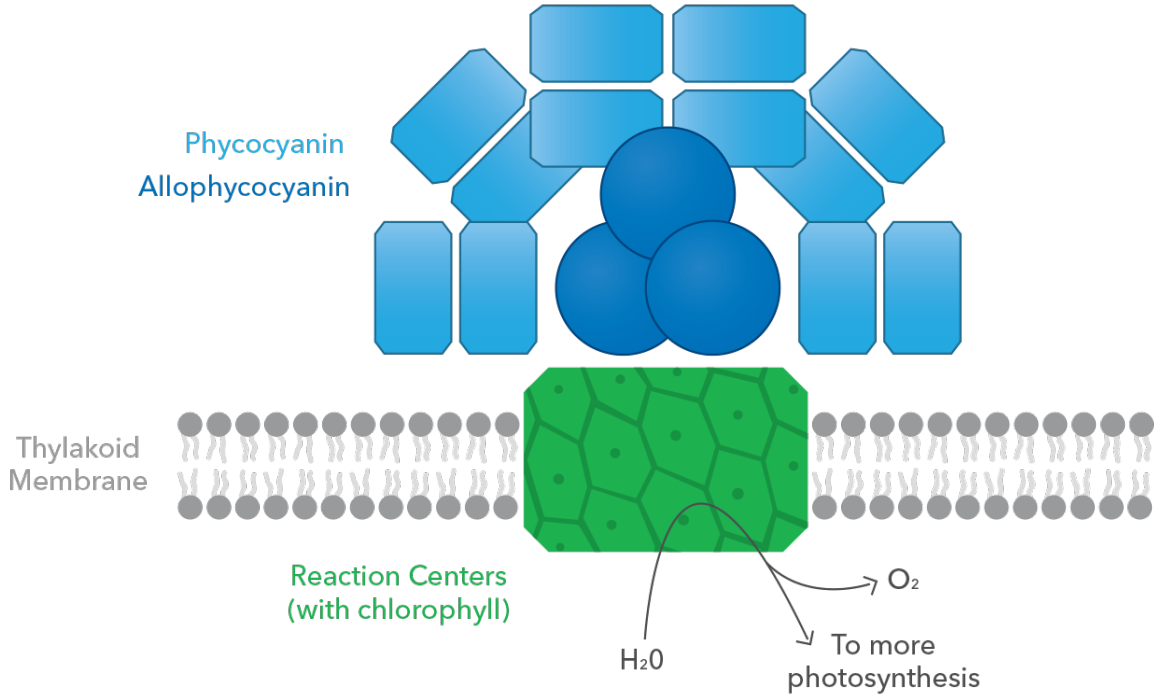
Fluorescent Pigments of Algae

Chlorophyll

- *All* algae

Allophycocyanin/Phycocyanin

- Blue-green algae



Fluorescent Pigments of Algae

Chlorophyll

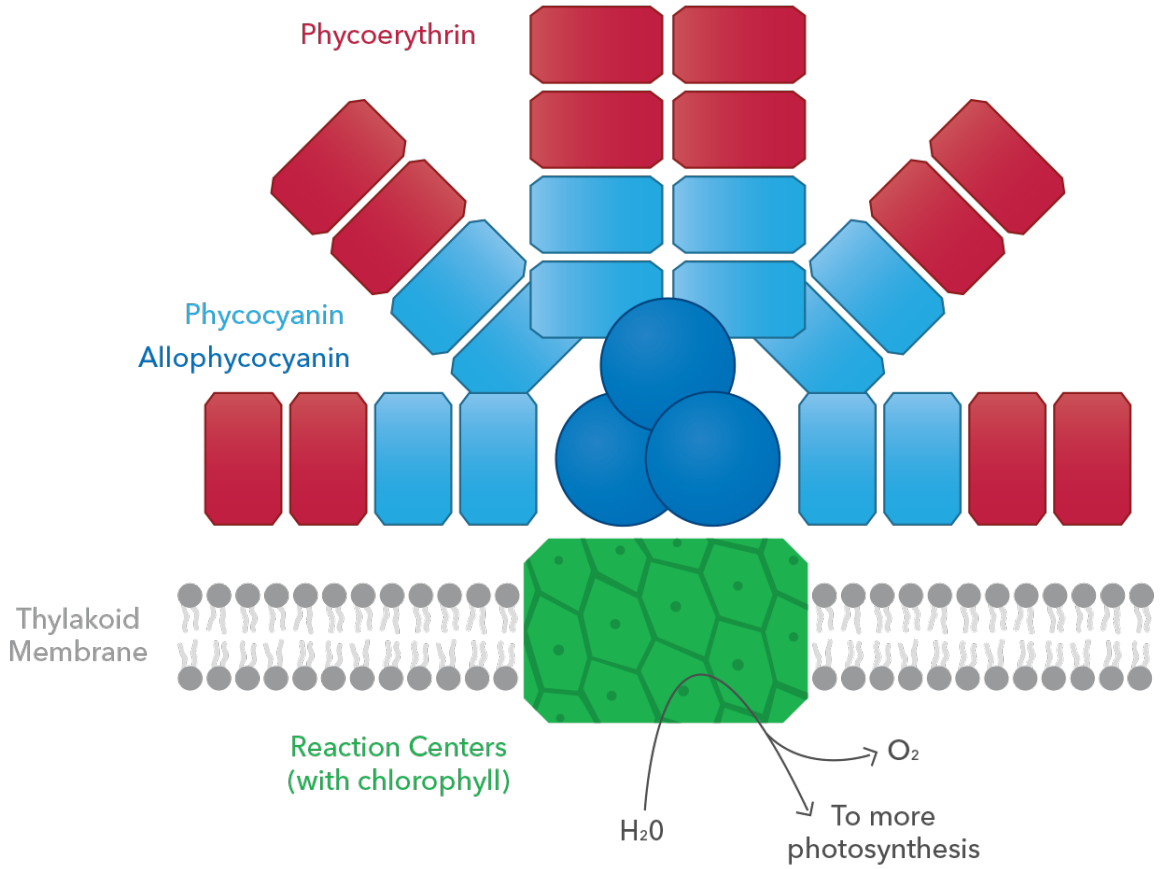
- *All* algae

Allophycocyanin/Phycocyanin

- Blue-green algae

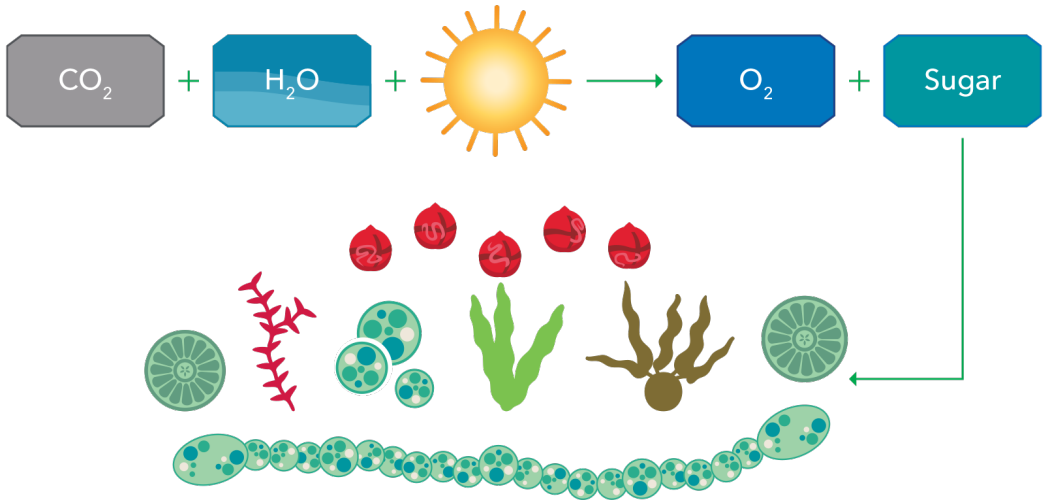
Phycoerythrin

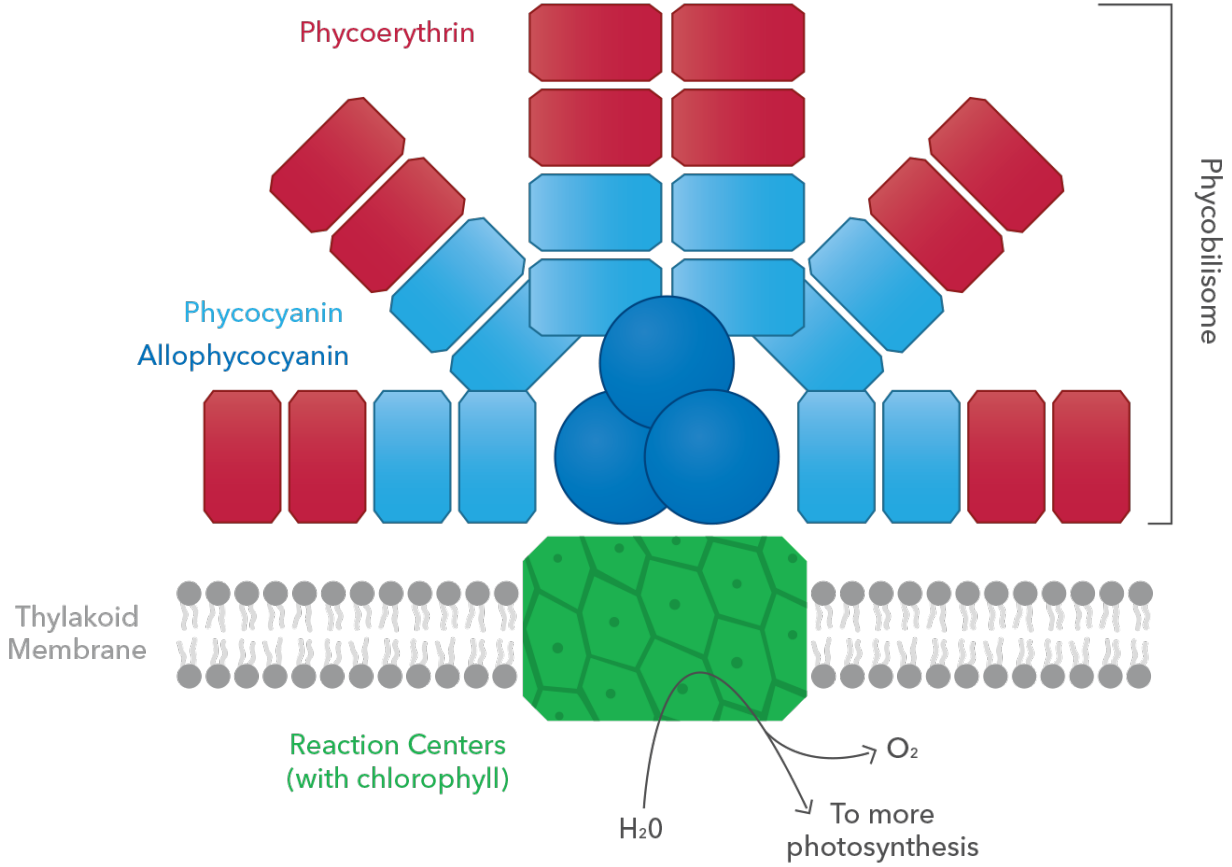
- Blue-green algae native to marine water



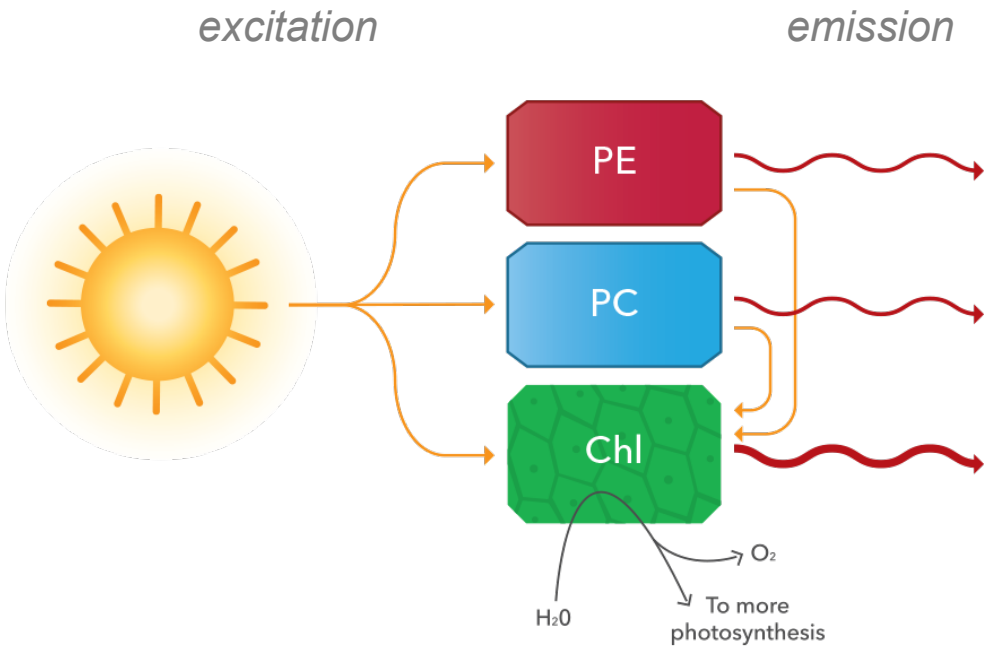
Fluorescent Pigments of Algae

The Phycobilisome channels light energy to chlorophyll for photosynthesis





Fluorescent Pigments of Algae



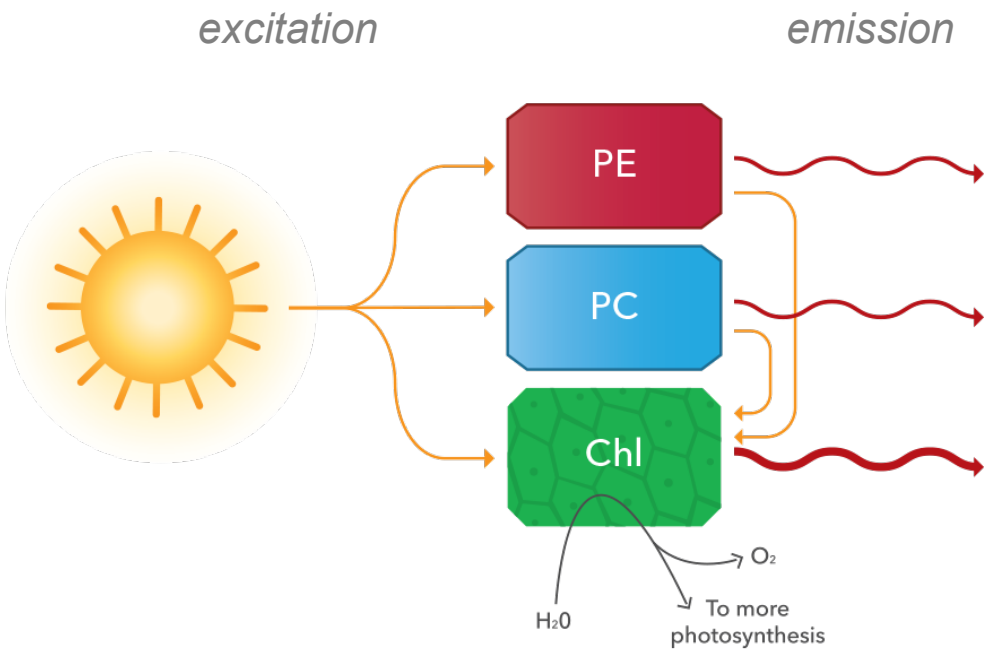
- Each pigment can absorb energy from sunlight (excitation)

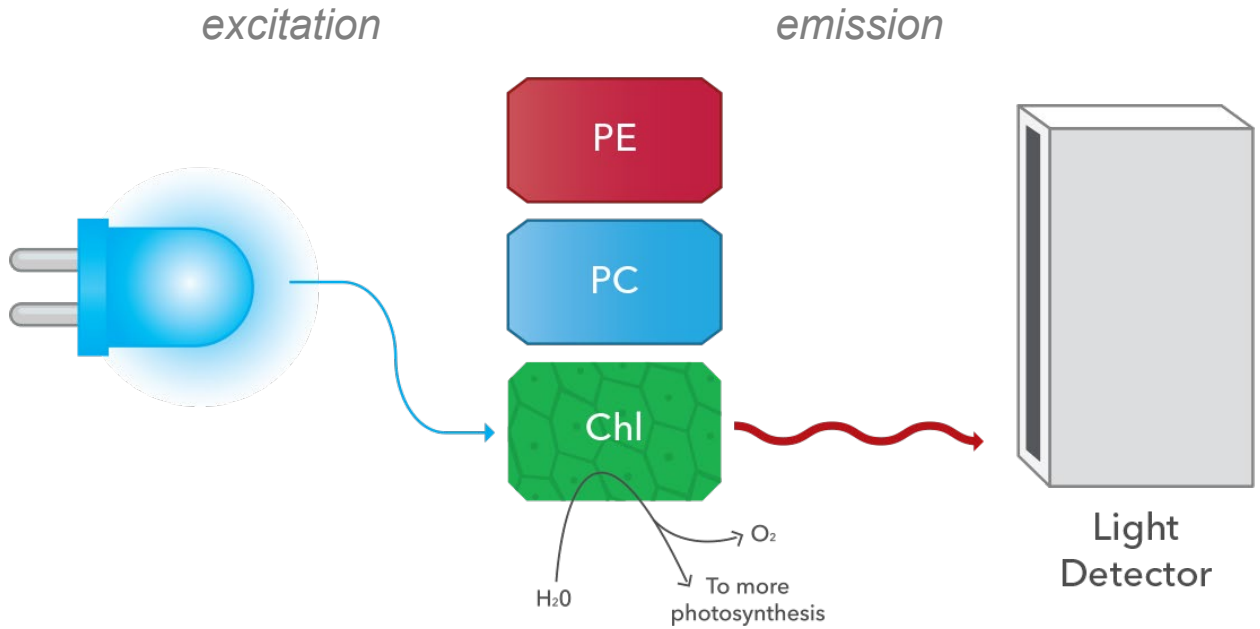
- Each pigment emits energy
- Light emitted by one pigment can excite other pigments
- Transfer to other pigments is not 100% efficient

- Ultimate goal: channel the energy to chlorophyll to drive photosynthesis

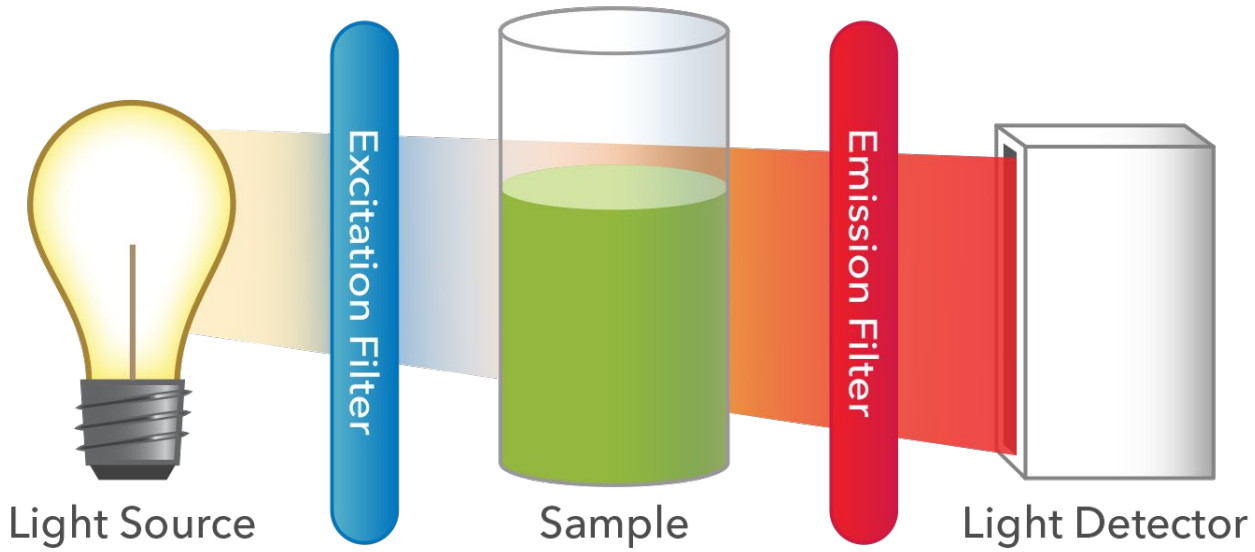
How Algae Sensors Work

A sensor uses an LED to excite pigments, and a detector to see the emissions





Measuring Fluorescence: Fluorometers



- Light Source
- Excitation filter: to select wavelengths of light that will excite only the molecules of interest
- Emission filter: to select wavelengths of light that only the molecules of interest are known to emit
- Detector will see the light that passes through the emission filter

How Algae Sensors Work

EXO™ Continuous Monitoring

ProDSS Spot Sampling

TAL Dual-channel Sensors

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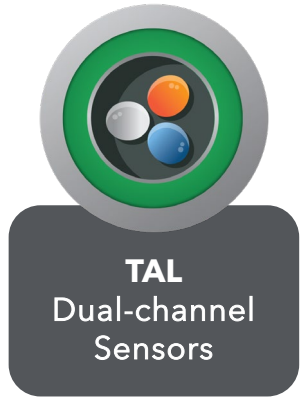
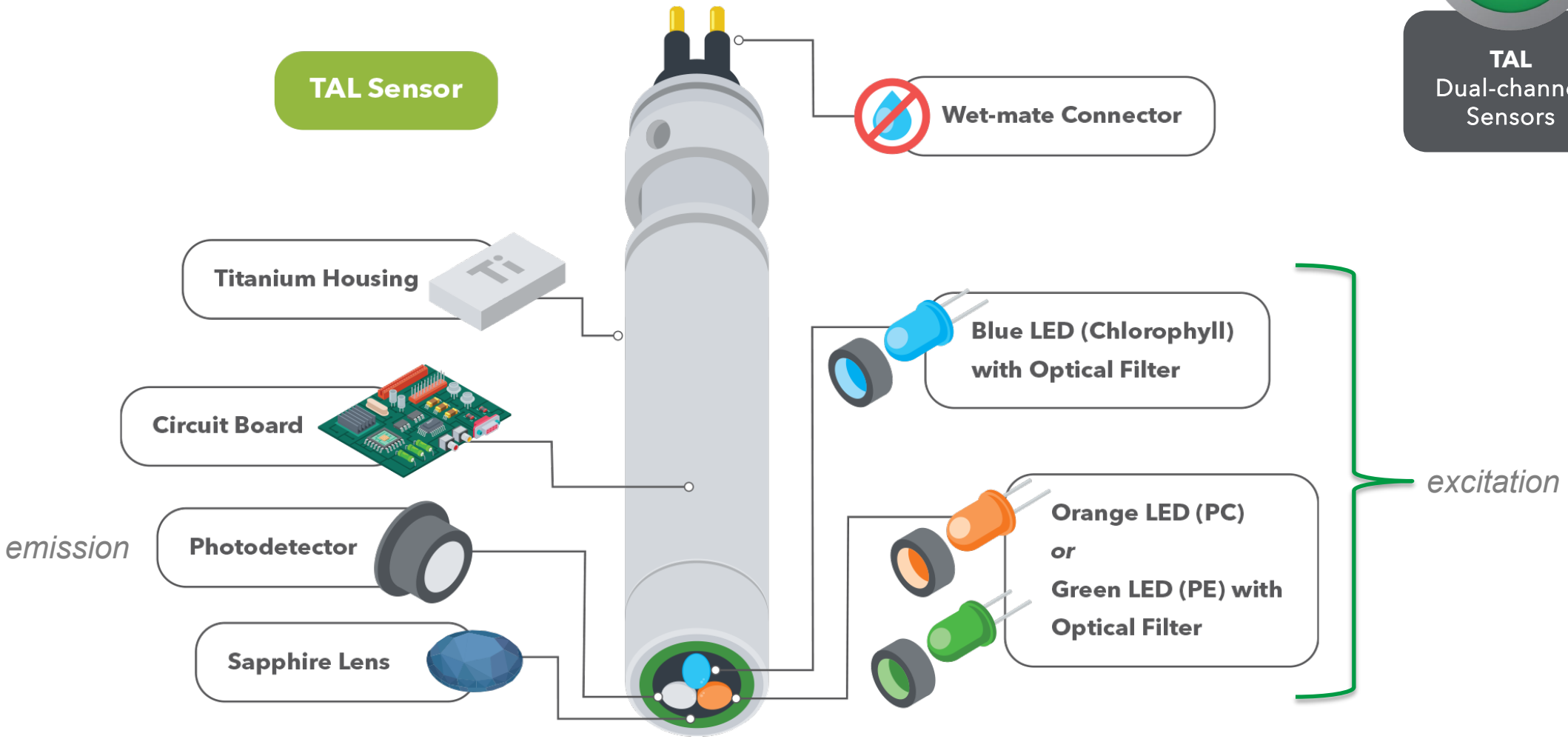
Anatomy of YSI's TAL Sensor





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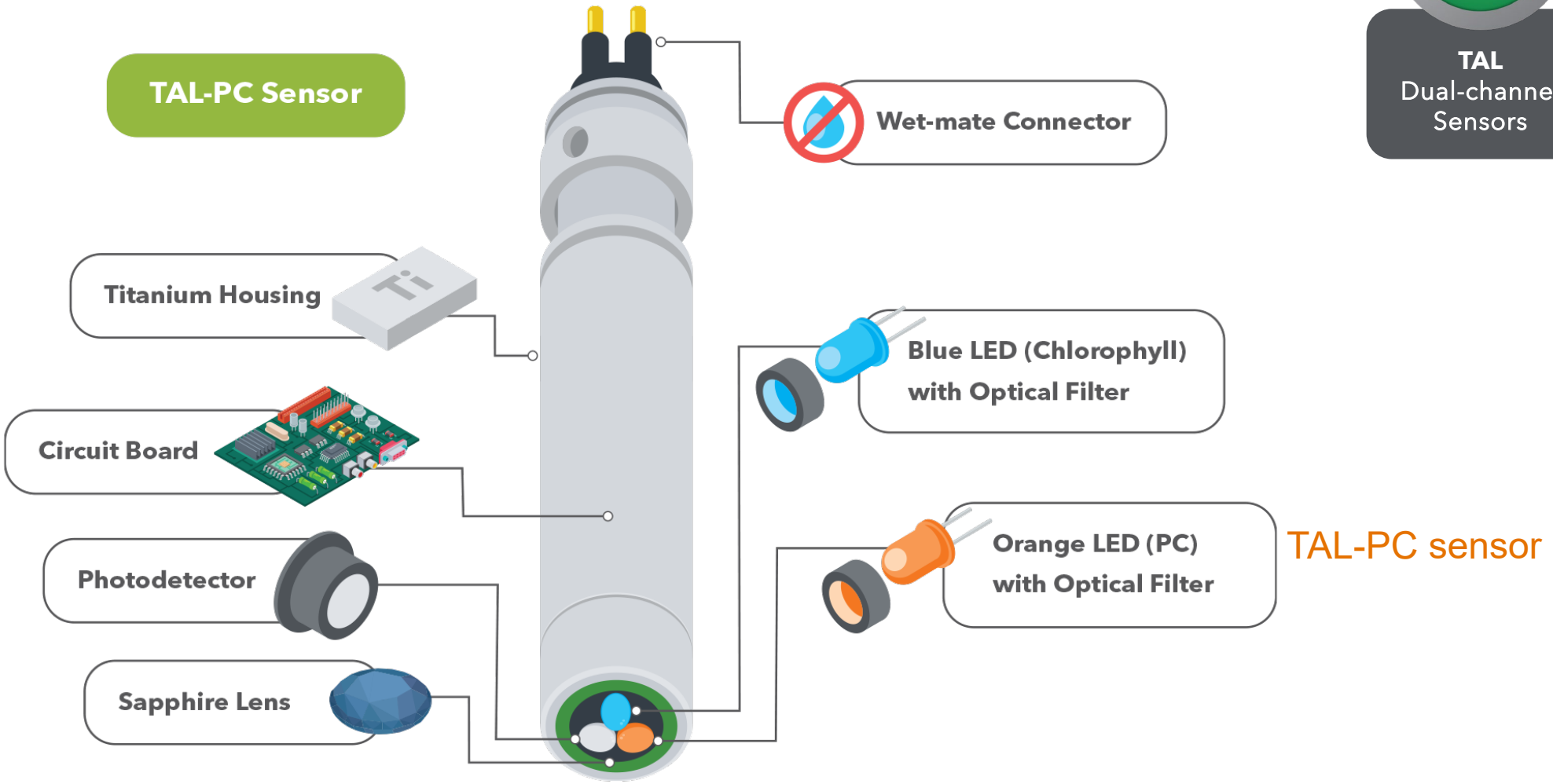
Anatomy of YSI's TAL-PC Sensor



TAL
Dual-channel
Sensors

TAL-PC sensor

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Anatomy of YSI's TAL-PC Sensor

TAL
Dual-channel
Sensors

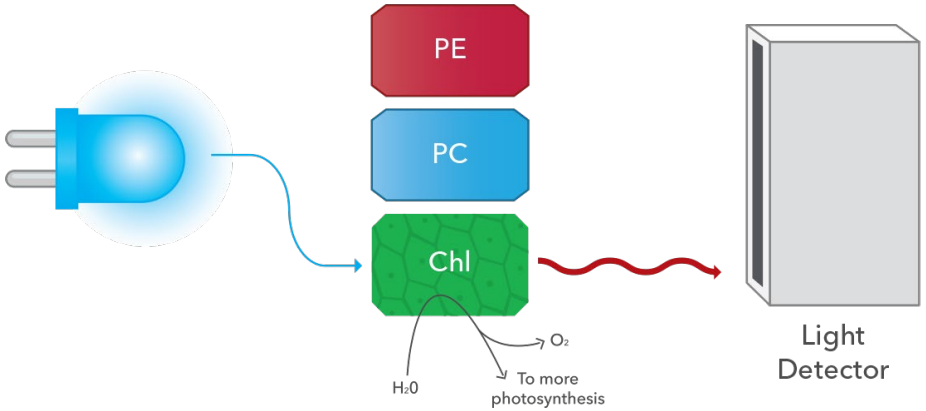
Two excitation channels, One Photodector

TAL-PC Sensor

Chlorophyll Channel

	PC	Chl
excitation	LED λ	470 ± 15 nm
emission	PD λ	685 ± 20 nm

TAL-PC



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Anatomy of YSI's TAL-PC Sensor

Two excitation channels, One Photodector

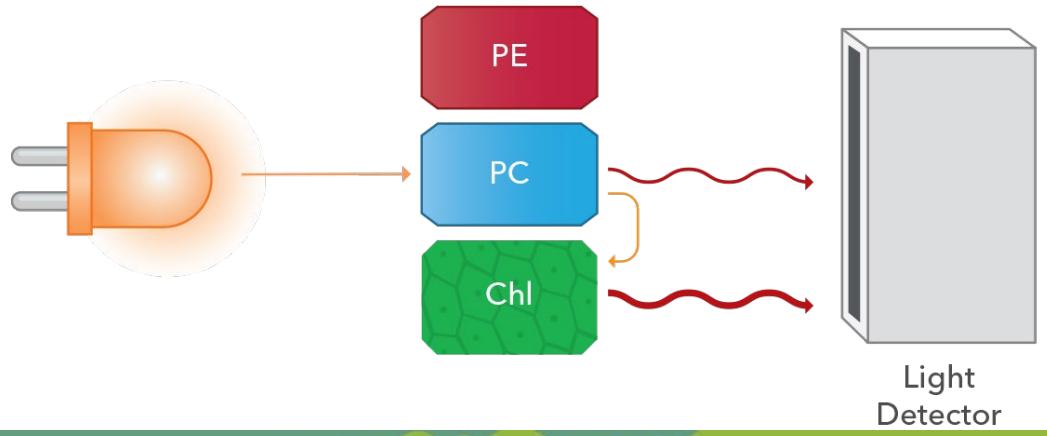
TAL-PC Sensor

Chlorophyll Channel

Phycocyanin Channel

		PC	Chl
excitation	LED λ	590 ± 15 nm	470 ± 15 nm
emission	PD λ		685 ± 20 nm

TAL-PC



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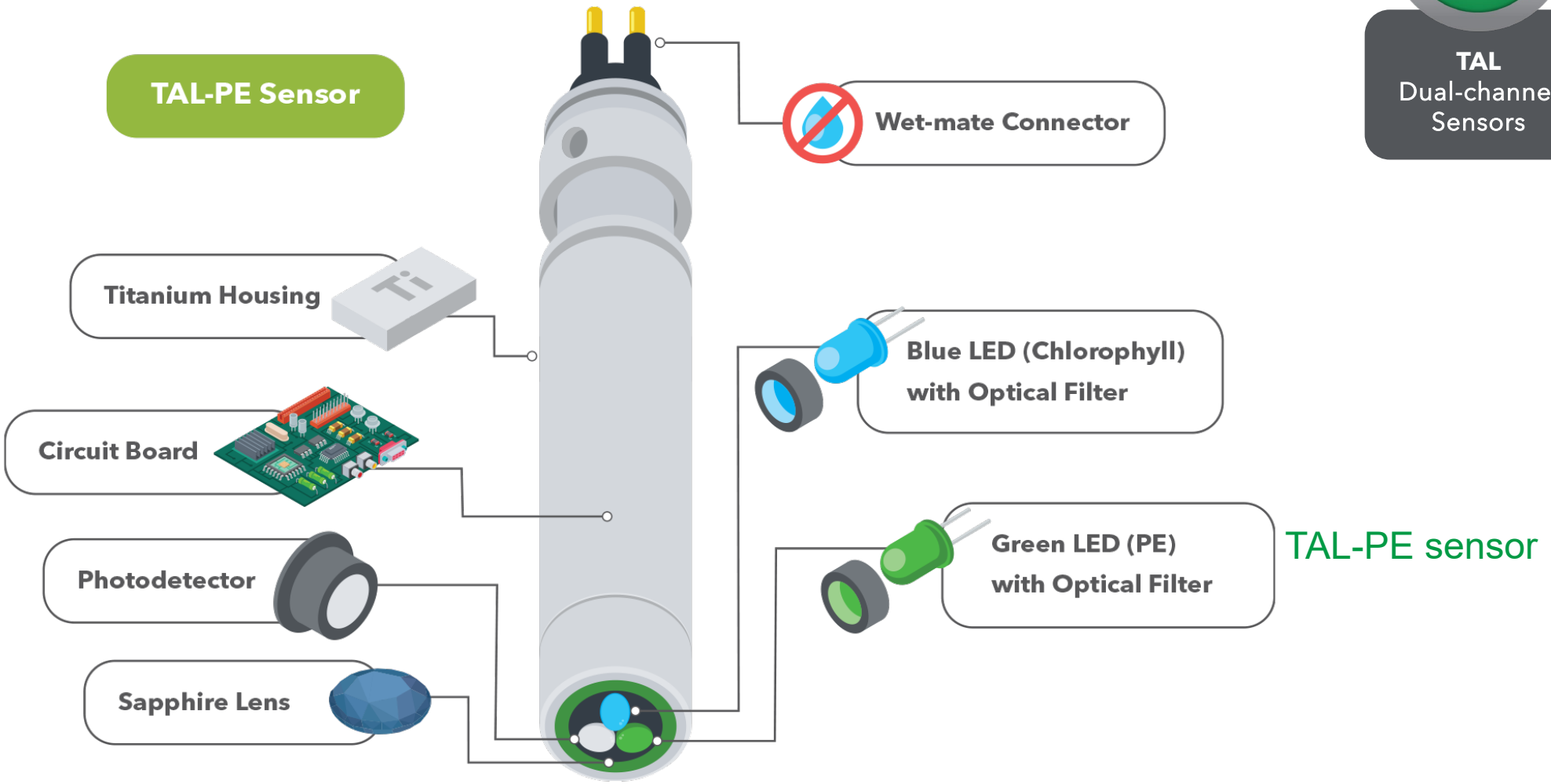
Anatomy of YSI's TAL-PE Sensor



TAL
Dual-channel
Sensors

TAL-PE sensor

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Anatomy of YSI's TAL-PE Sensor

TAL
Dual-channel
Sensors

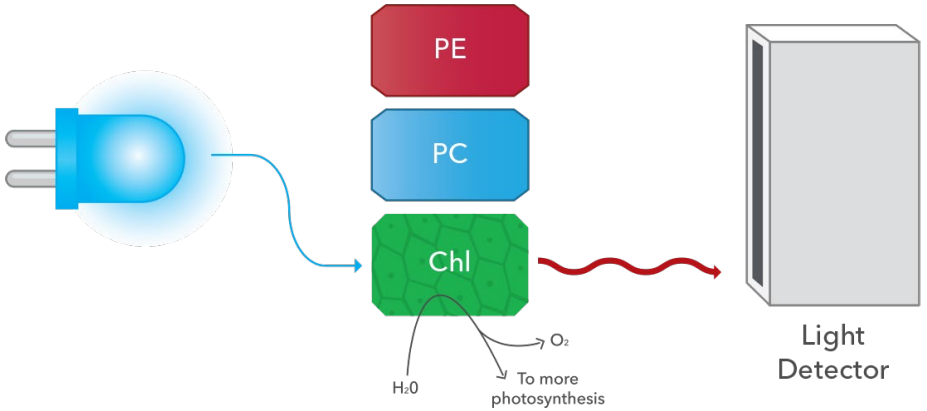
Two excitation channels, One Photodector

TAL-PE Sensor

Chlorophyll Channel

	Chl	PE
excitation LED λ	470 ± 15 nm	
emission PD λ	685 ± 20 nm	

TAL-PE



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Anatomy of YSI's TAL-PE Sensor

TAL
Dual-channel
Sensors

Two excitation channels, One Photodector

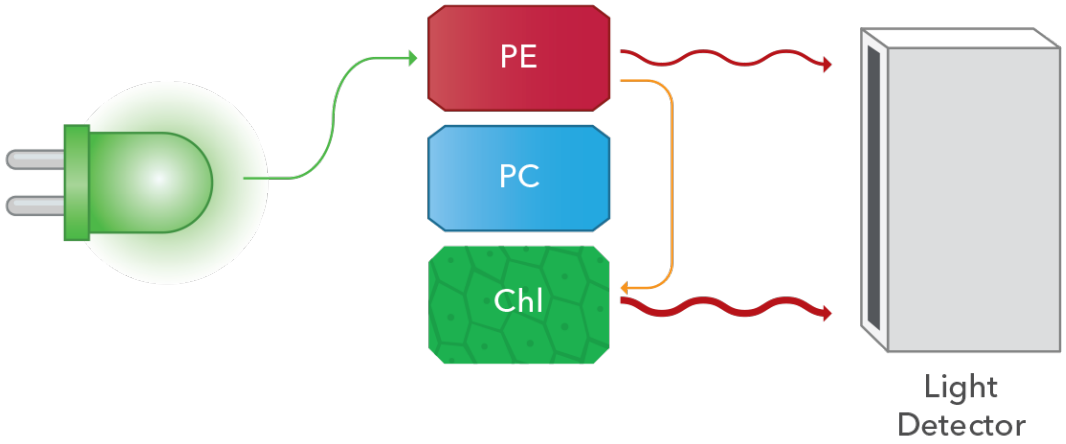
TAL-PE Sensor

Chlorophyll Channel

Phycoerythrin Channel

		Chl	PE
excitation	LED λ	470 ± 15 nm	525 ± 15 nm
emission	PD λ	685 ± 20 nm	

TAL-PE



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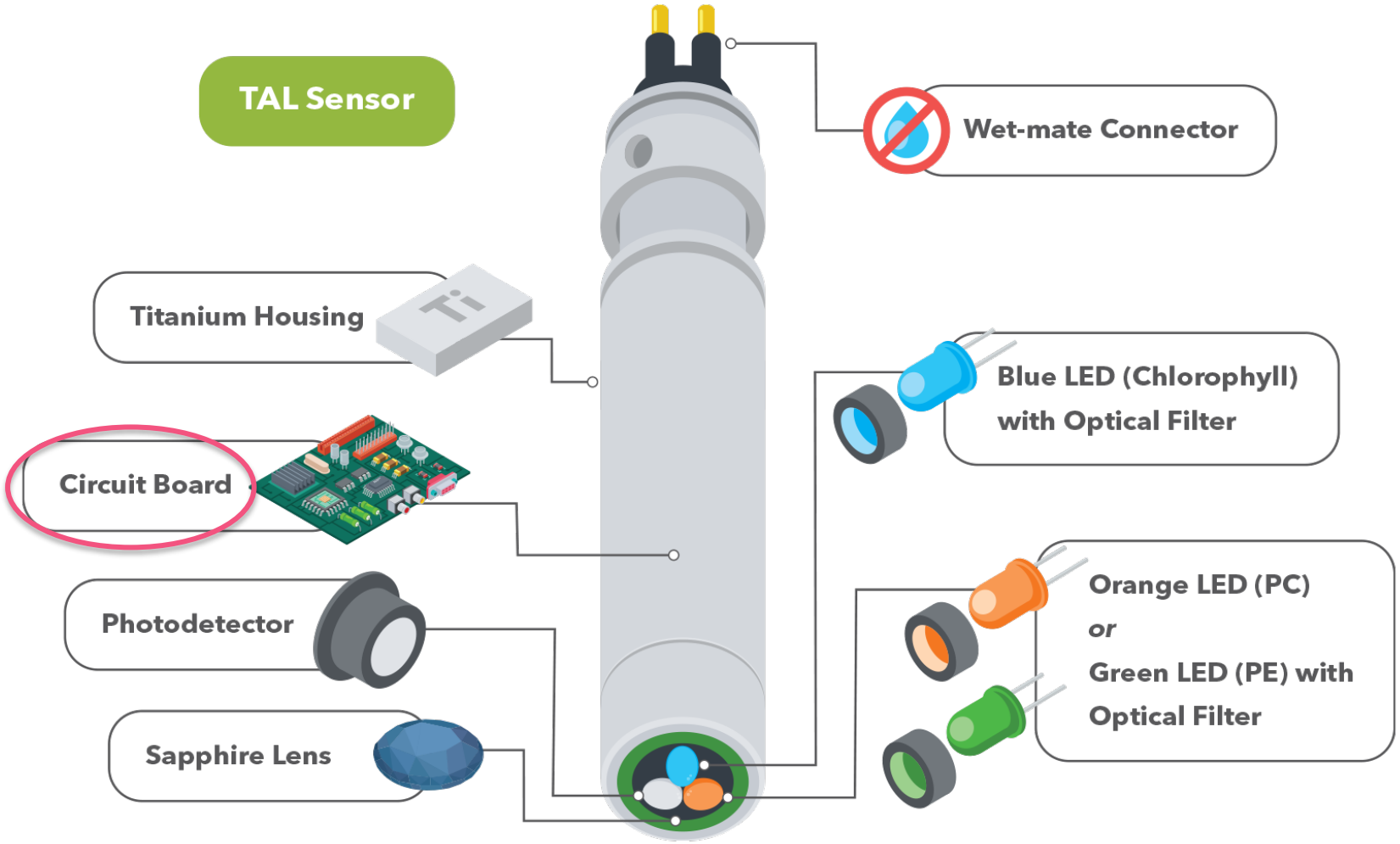
Anatomy of YSI's TAL Sensor



TAL
Dual-channel
Sensors

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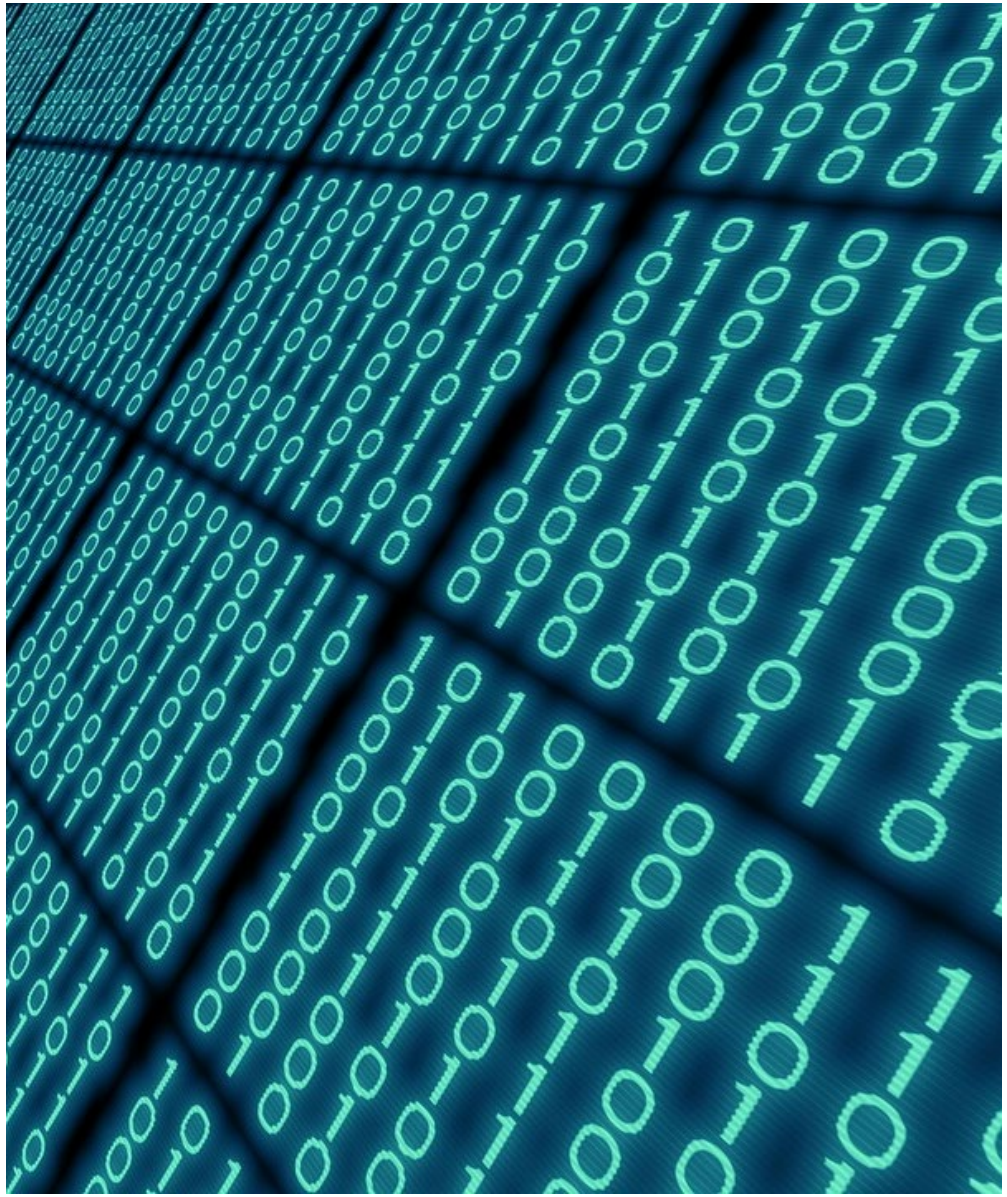
Anatomy of YSI's TAL Sensor

- Board design: minimize electrical "noise"
  - Signal-to-noise ratio

- Firmware:
  - Modulation of optical frequencies
  - Additional filtering of excitation and emission wavelengths
  - Modulation of power to optimize range



Anatomy: Form Drives Function

This Feature…	Translates To…
Quality of the LEDs	Less drift, less power, longer life
Quality of the filters and PD	High specificity for signal of interest
Board design/electronics	Better SNR, better sensitivity
Mechanical construction with reduced detection angles	Fewer environmental interferences, better SNR
Firmware	Even better SNR, better sensitivity

Anatomy: Form Drives Function

Feature	6-Series PC	EXO TAL-PC	So?
Excitation optics	590 ± 20 nm	590 ± 15 nm	More specific target excitation, needs less power
Emission optics	640 ± 40 nm	685 ± 20 nm	Less prone to interferences and non-specific signals
Data Processing	0.1 RFU DL	0.01 RFU DL	More sensitive

Sensor Principles: Summary

- Form drives function—understand how your sensor works!
- Firmware is as important as hardware
  - The signals you see are *post-processed*
- The best sensors:
  - Balance specificity with sensitivity
  - Optimize SNR
  - Are ruggedized for field applications

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How Algae Sensors Work: Best Practices

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What would you guess is the biggest challenge people face when monitoring for algae?

Best Practices

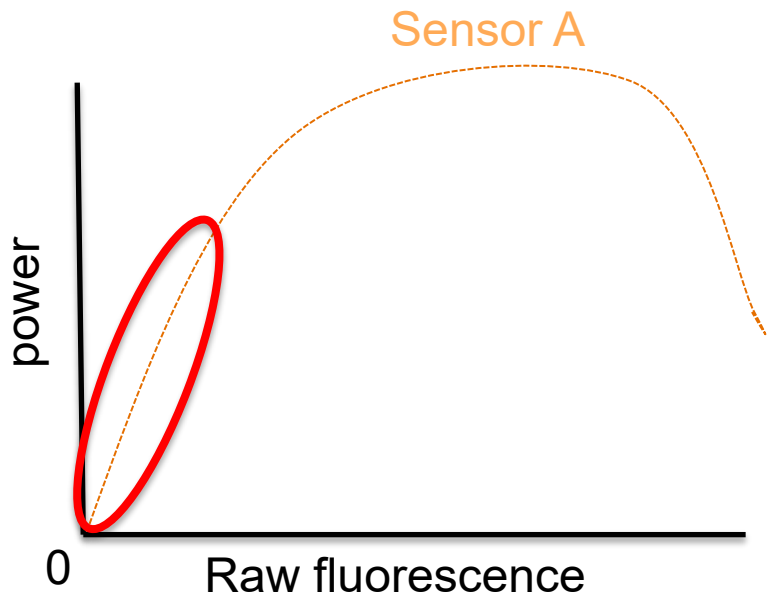
I. Sensor Calibration
II. Prevent Biofouling
III. Use RFU

https://www.ysi.com/ysi-blog/water-blogged-blog/2019/02/answers-to-the-top-5-hab-monitoring-questions

https://www.ysi.com/ysi-blog/water-blogged-blog/2019/07/how-algae-sensors-work-answers-to-four-challenging-questions

Sensor Calibration: Why

- Each sensor is “tuned” so its 0-100% relative fluorescence unit (RFU) scale is within its linear output range



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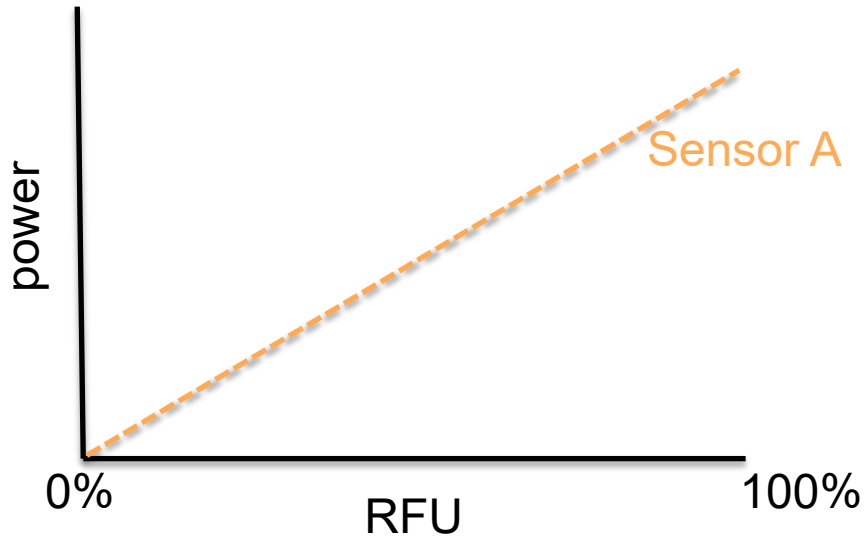
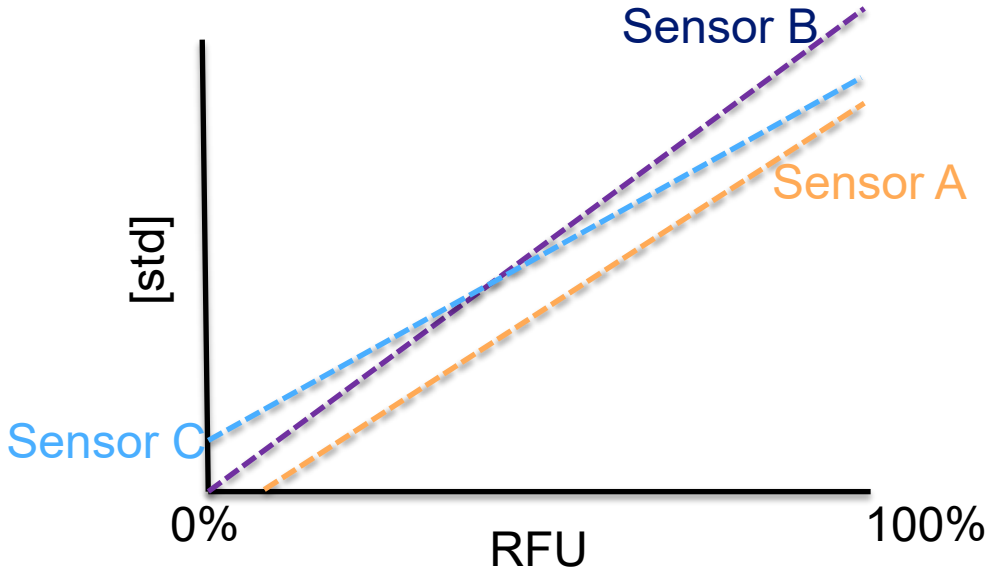


Sensor Calibration: Why

- Each sensor is “tuned” so its 0-100% relative fluorescence unit (RFU) scale is within its linear output range
- RFU scale is not identical between sensors at the point of manufacture, and
- As sensors are used, they may drift so that their scales are no longer identical



Sensor Calibration: Why

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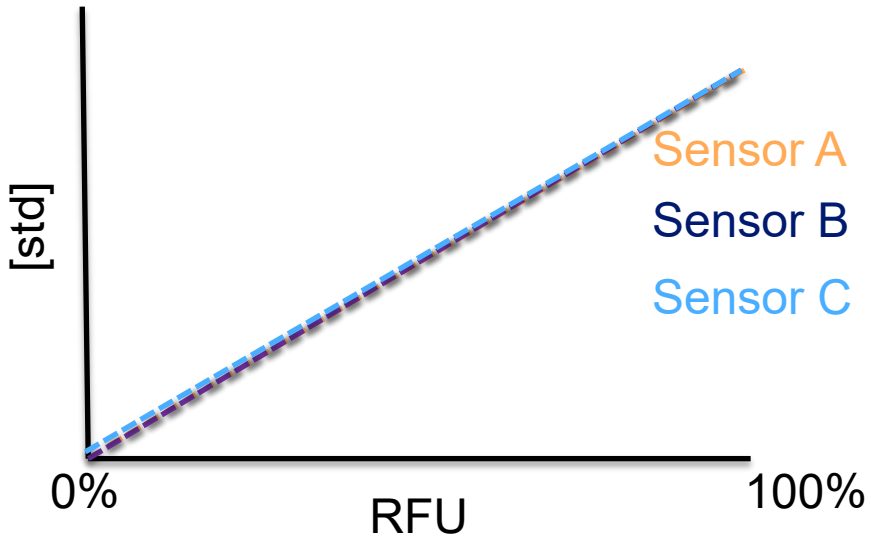
- RFU scale is not identical between sensors at the point of manufacture, and
- As sensors are used, they may drift so that their scales are no longer identical

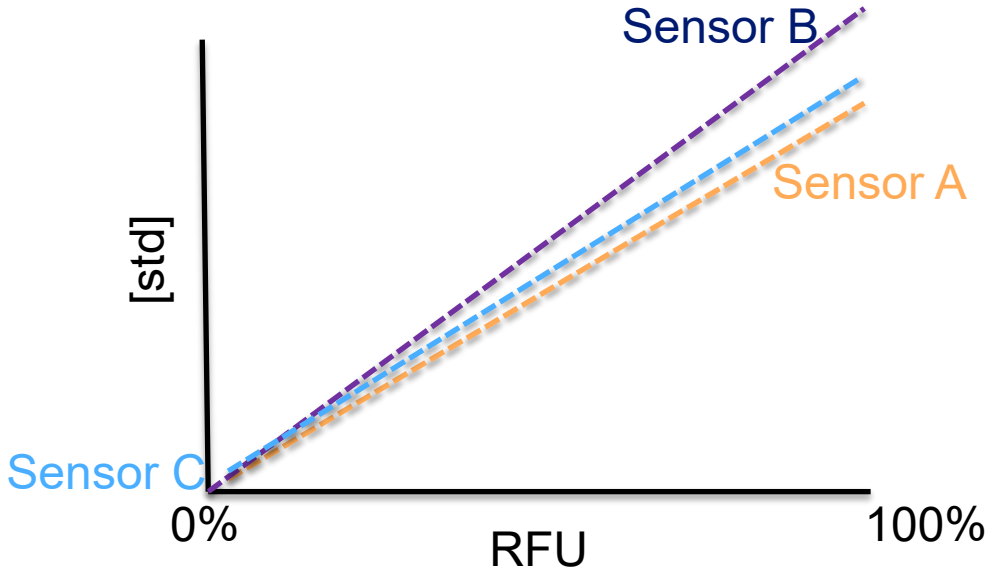
Calibration of RFU against a standard allows sensor outputs to be compared



Sensor Calibration: "One-Point Calibration"

- Water standard
- A, B, and C will give similar readings at the lower RFU scale, but become more dissimilar at higher RFUs

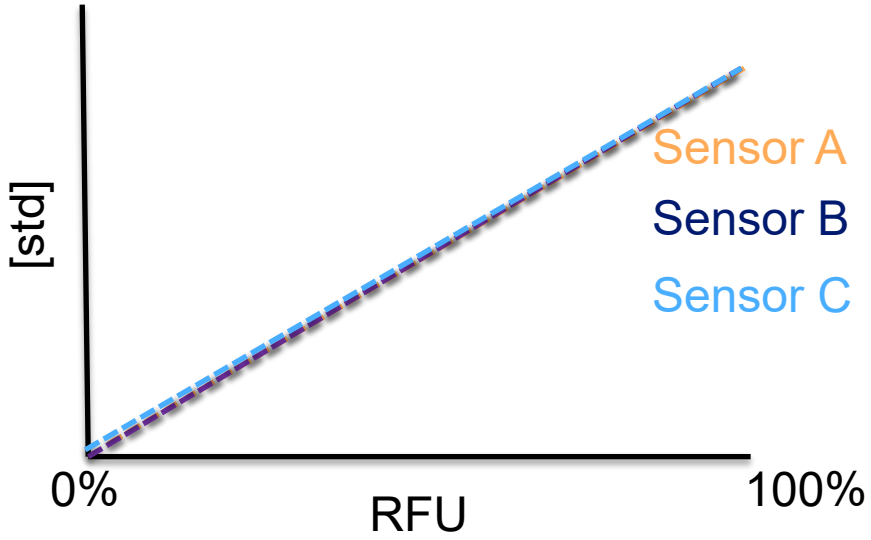
A "one-point cal" is a zero reset, not a true calibration of the full scale of an optical sensor



Sensor Calibration: "Two-Point Calibration"

- Water standard + a standard that reads somewhere beyond zero
- All sensors now have the same behavior across their RFU ranges

A "two-point cal" allows sensor data to be compared across the entire RFU scale



Sensor Calibration: How

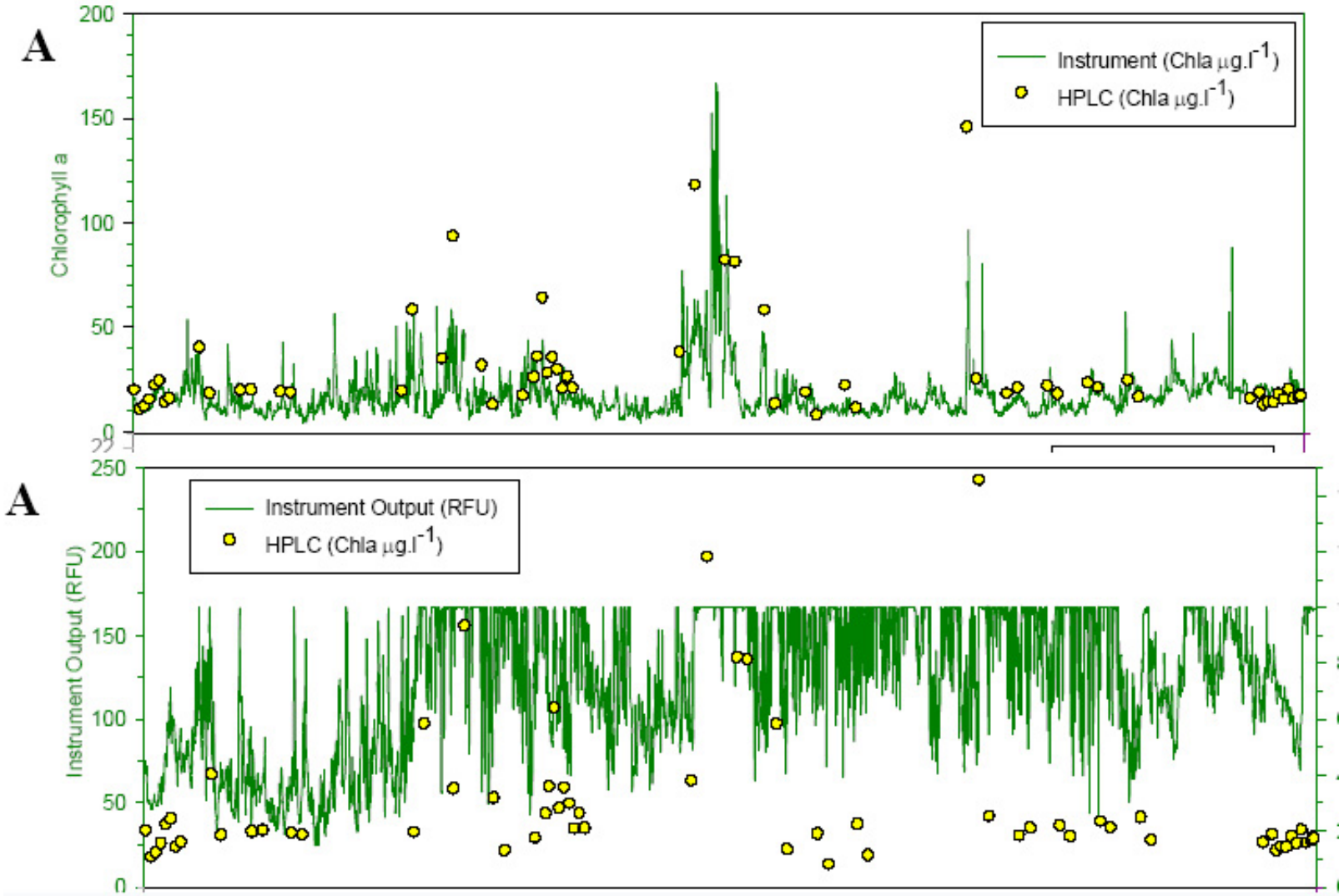
- Clean, clean, clean!
  - Glassware
  - Calibration cup
  - Sensor faces
  - Remove wiper
- Multical
  - Multiple sensors at once
  - One batch of standard
  - Lessens variability

YSI EXO2
Sonde
+ CT
+ 5xTAL-PC

Prevent Biofouling

Biofouling is the #1 enemy of optical sensor data during continuous monitoring!

Prevent Biofouling



Prevent Biofouling

Pre-deployment
EXO Sonde with copper sensor guard.

Post-deployment
Heavy fouling prevented by copper sensor guard.

Post-deployment
Wiped sensing area free of biofouling.

How Anti-Fouling Works

- Freshwater Fouling
- Marine Fouling
- Evolution and Principles of Antifouling Technology
- Recommended Cleaning Procedures

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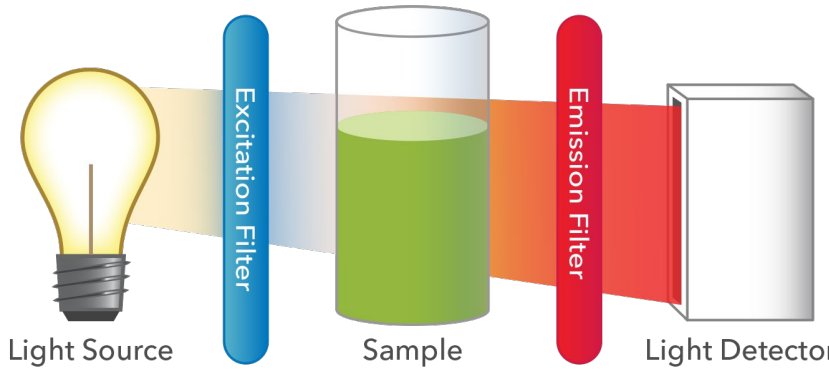
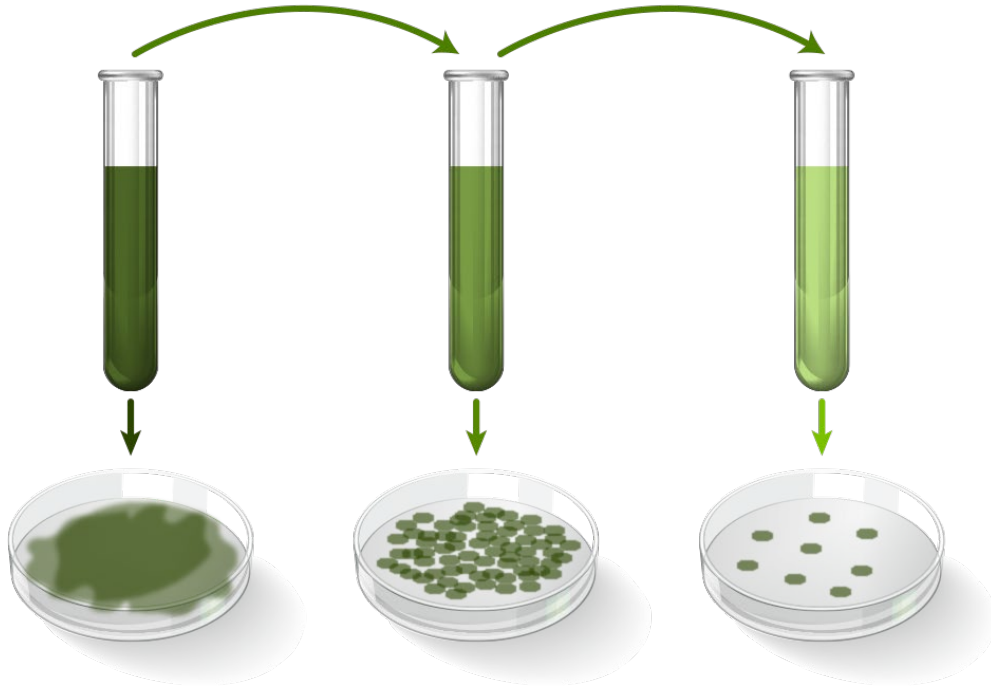
Sensor Units of Measurement

- µg/L of pigment (ppb)
- Cells/mL or CFU/mL
- Biovolume
- RAW
- Relative Fluorescence Units (RFU)

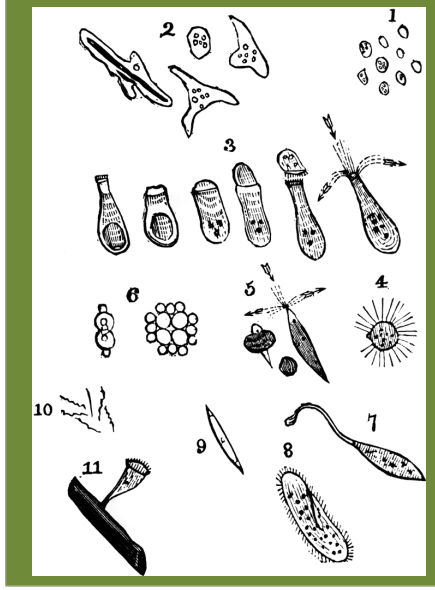




Algae Monitoring Tools

Microscopy

Cells/mL

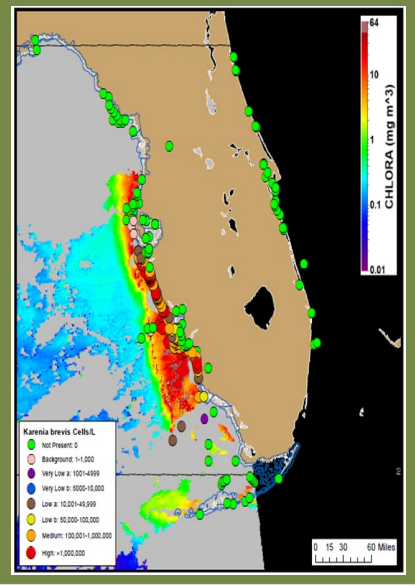


Chlorophyll Analysis

µg/L

***In vivo / in situ* Sensors**

Satellites



Online analyzers

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Chlorophyll Analysis

EPA Method 445.0:
Fluorometric Analysis

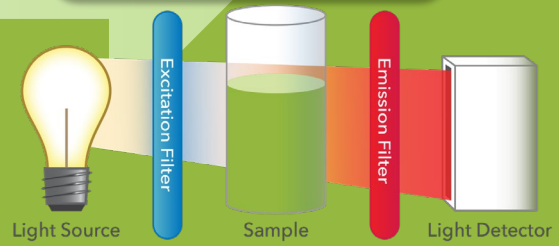
UOM: µg/L or ppb

Sample

Concentrate

Extract

Measure



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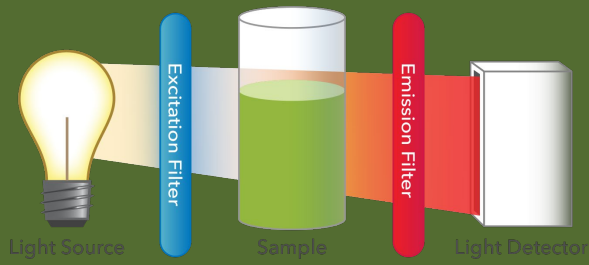
In vivo / in situ Sensors



Sample

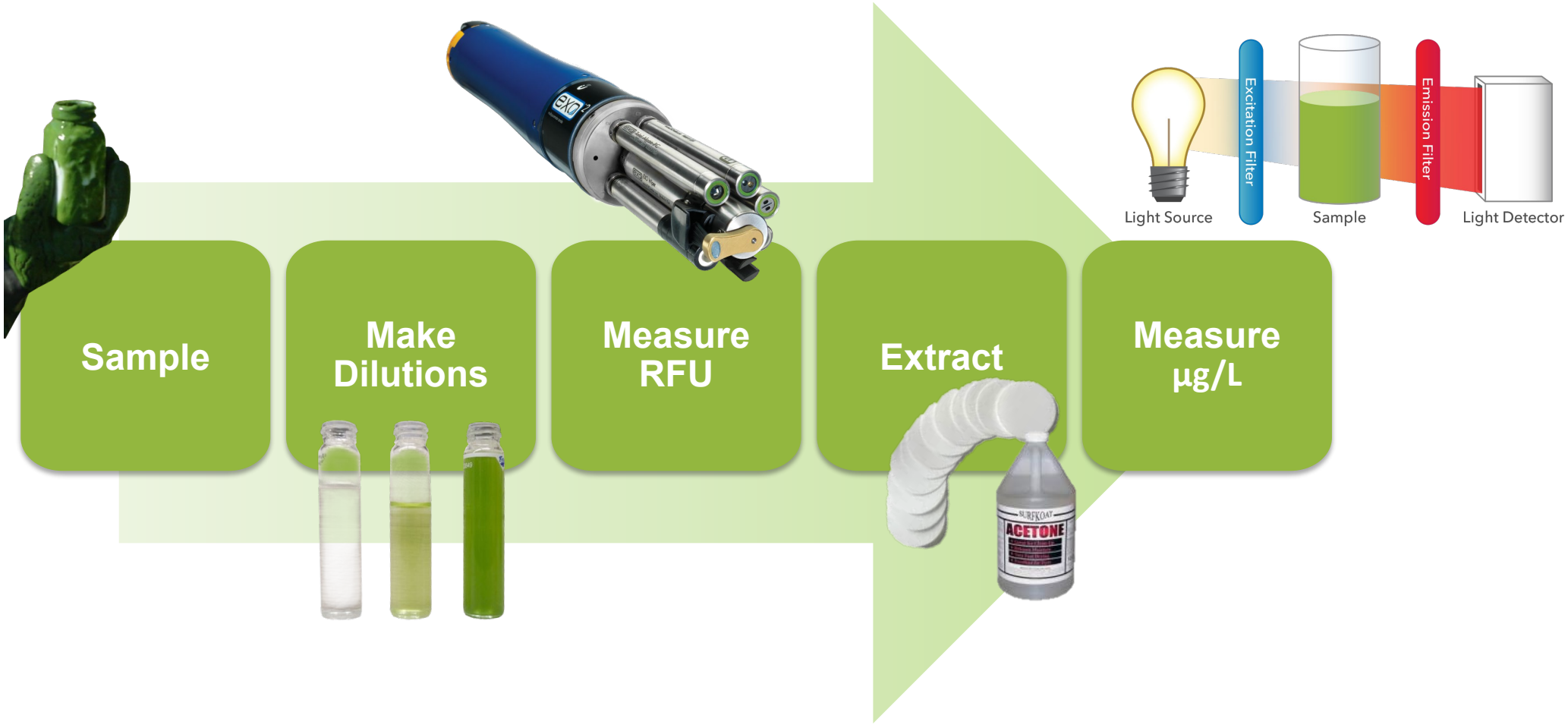
Make Dilutions

Measure RFU

Extract

Measure µg/L

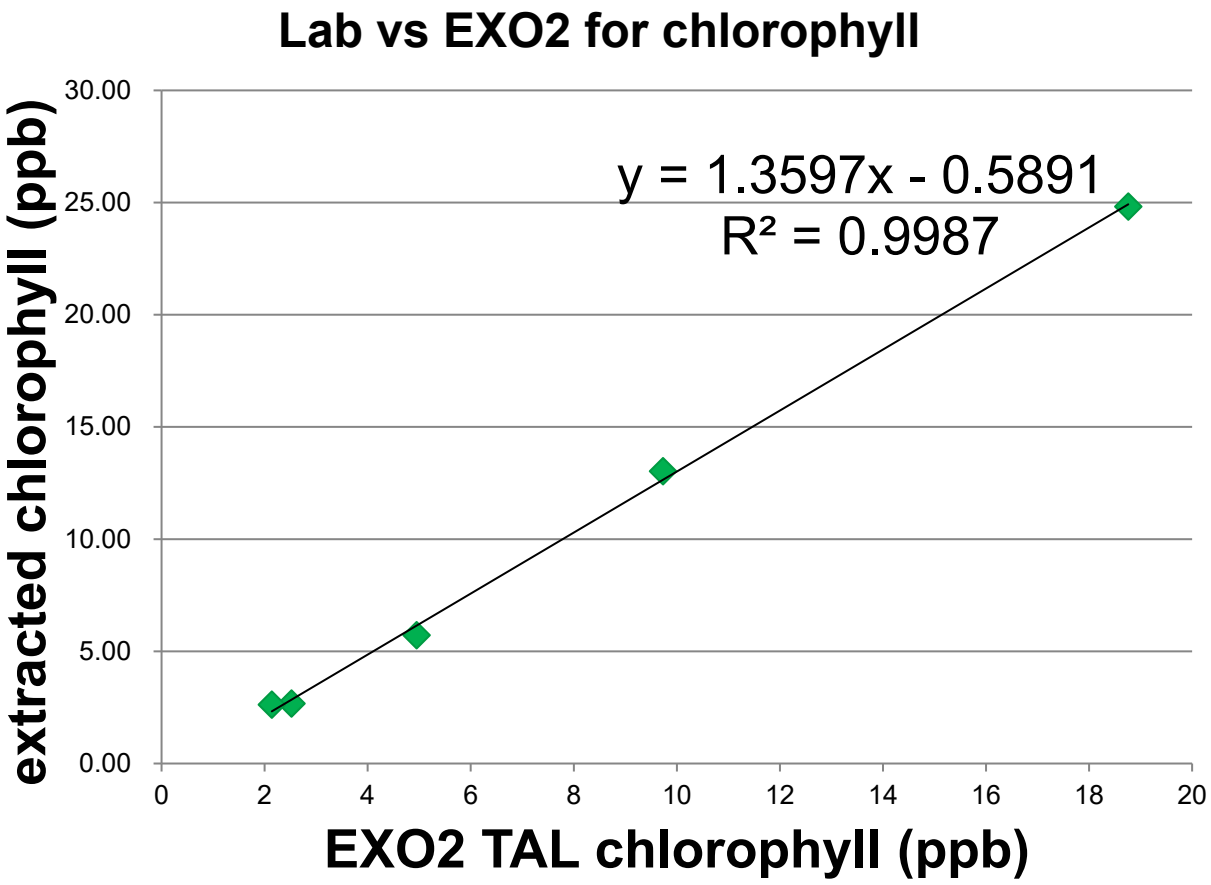
Sensor Units of Measurement: Trust, but Verify!!



Sensor Units of Measurement: Trust, but Verify!!

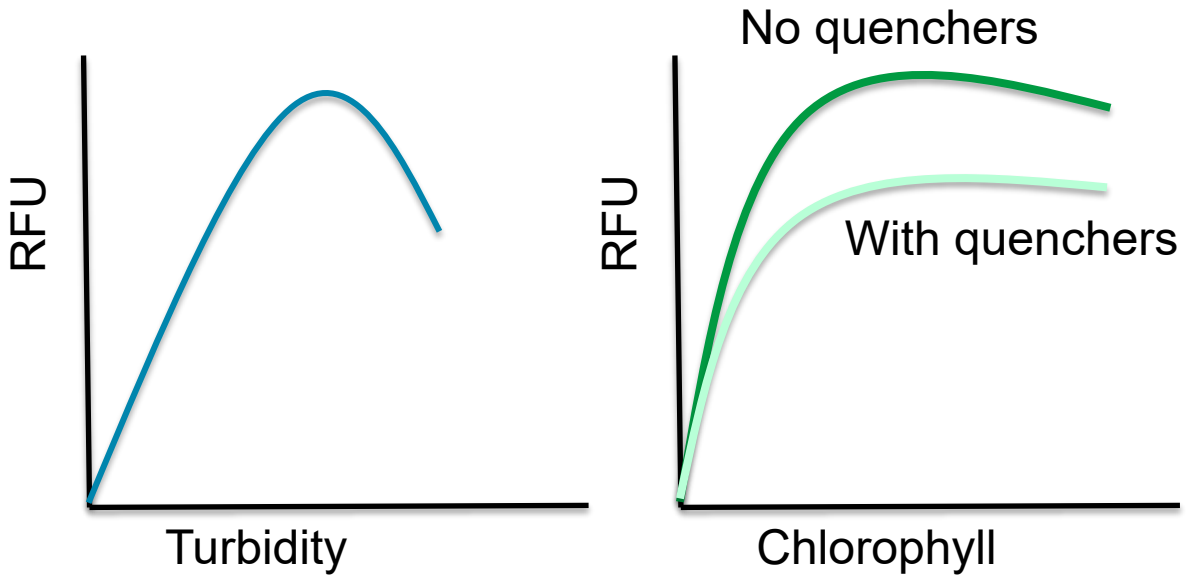
- **Geihu Lake, China**
- Followed process on prior slide
- Used bench fluorometer
- Strong correlation in this case…
- Will use EXO ppb output

Thank you James Chen
of Xylem Beijing!

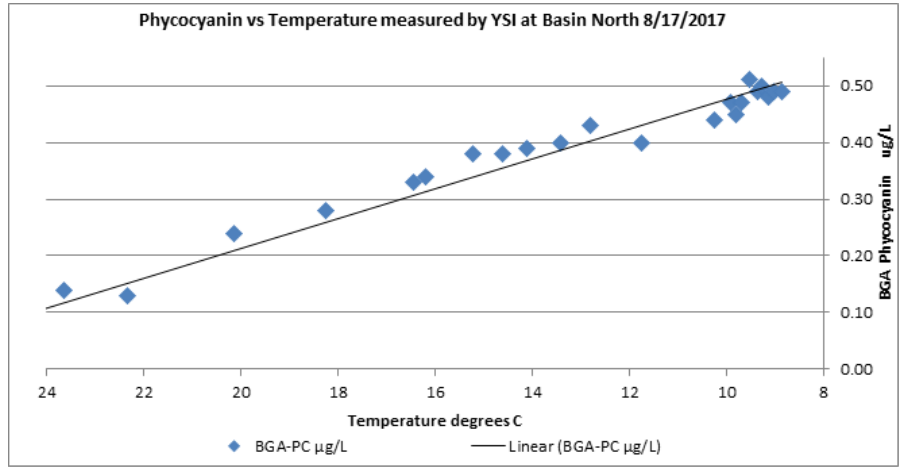


Are You Ready?

1. Environmental Factors
   a) Turbidity
   b) IFE
   c) Temperature effects



http://video.ysi.com/ysi-webinar-are-you-ready-harmful-algal-blooms



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Are You Ready?

1. Environmental Factors
2. Algal Physiology
    1. Pigments are in membranes
    2. Pigment turnover
    3. Algae move in the water column

http://video.ysi.com/ysi-webinar-are-you-ready-harmful-algal-blooms

Plant Physiol. (1975) *56*, 791–796

Temperature Dependence of Chlorophyll *a* Fluorescence in Relation to the Physical Phase of Membrane Lipids in Algae and Higher Plants[1]

Received for publication June 9, 1975 and in revised form August 20, 1975

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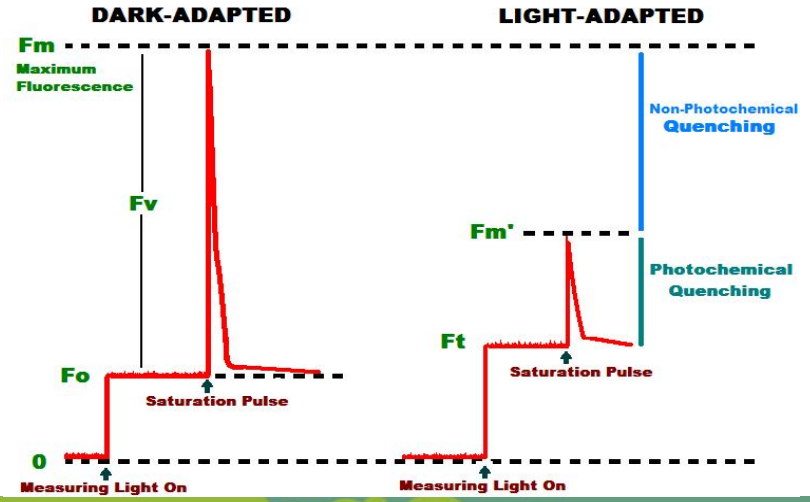

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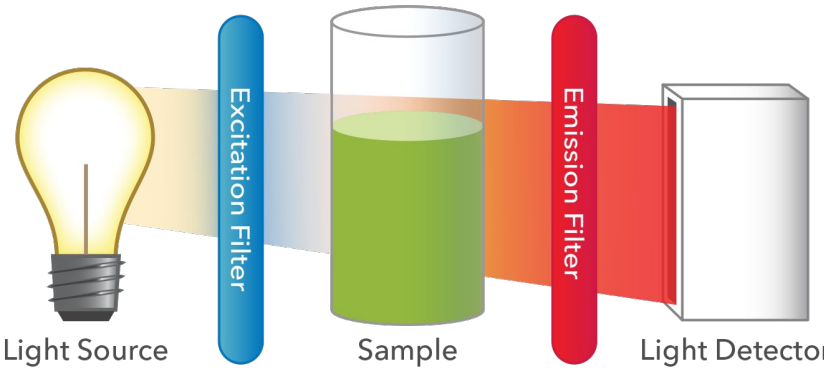
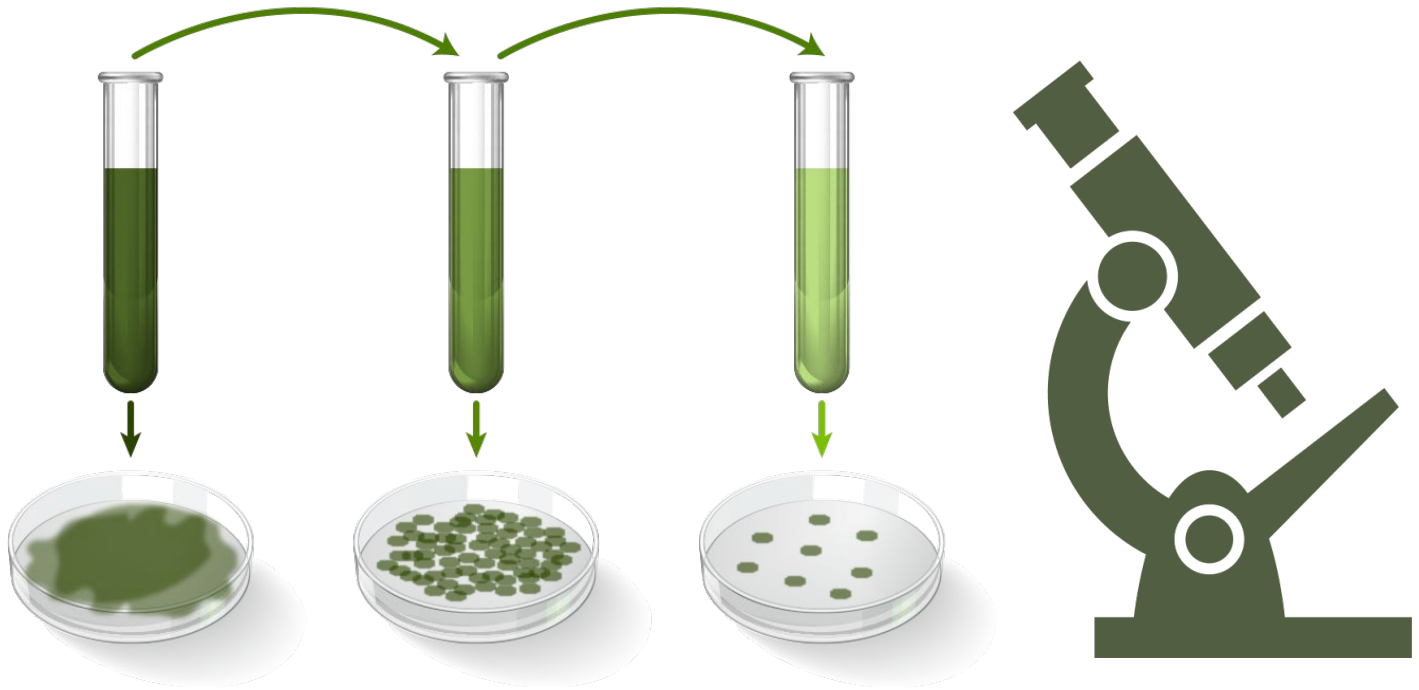
Sensor Units of Measurement

- ~~RAW~~
- Relative Fluorescence Units (RFU)
- ~~µg/L of pigment (ppb)~~
- ~~Cells/mL or CFU/mL~~
- ~~Biovolume~~





Best Practices: Summary

- Check calibration
- Whenever possible, do two-point calibration
- Prevent Fouling
- Use RFU and build your system understanding

Do you want someone from YSI to contact you to discuss algae sensors?

Questions?

Contact us:

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June 2nd / www.xylem-analytics.asia

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