Measuring Chl *a* & Nutrient Enrichment Bioassays

Starting Questions

1. What is Chlorophyll?
2. Why do we measure it?

Measuring Chl *a*

1. Filter
2. Extract
3. Fluorometer
4. Acidification
5. Back calculate to account for extraction process

Filter

Filter

Filter

25 ml

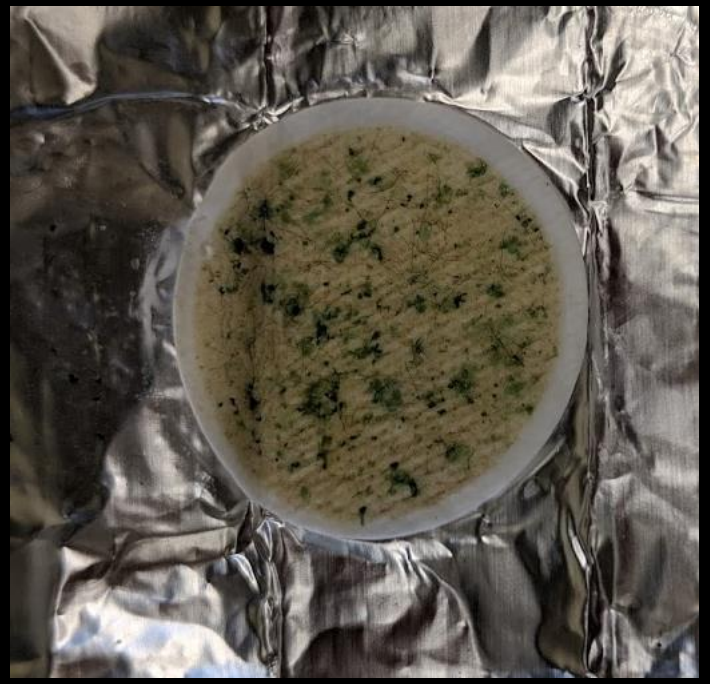
50 ml

100 ml

500 ml

Filter

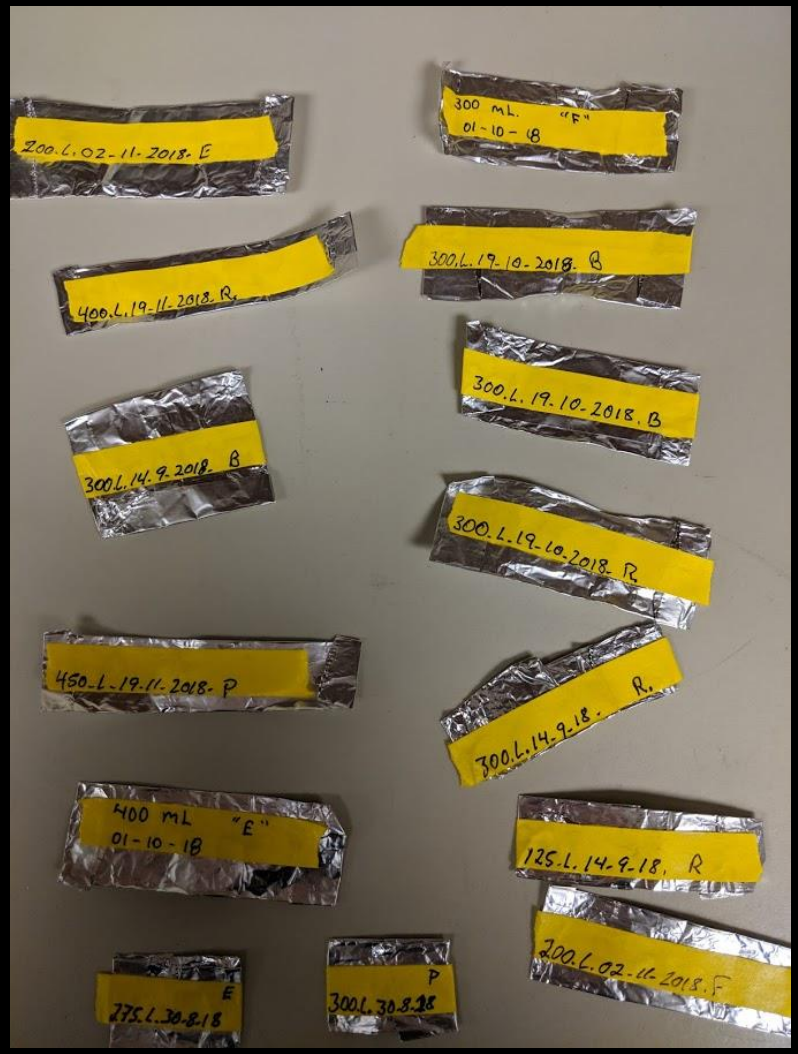
250 ml



250 ml

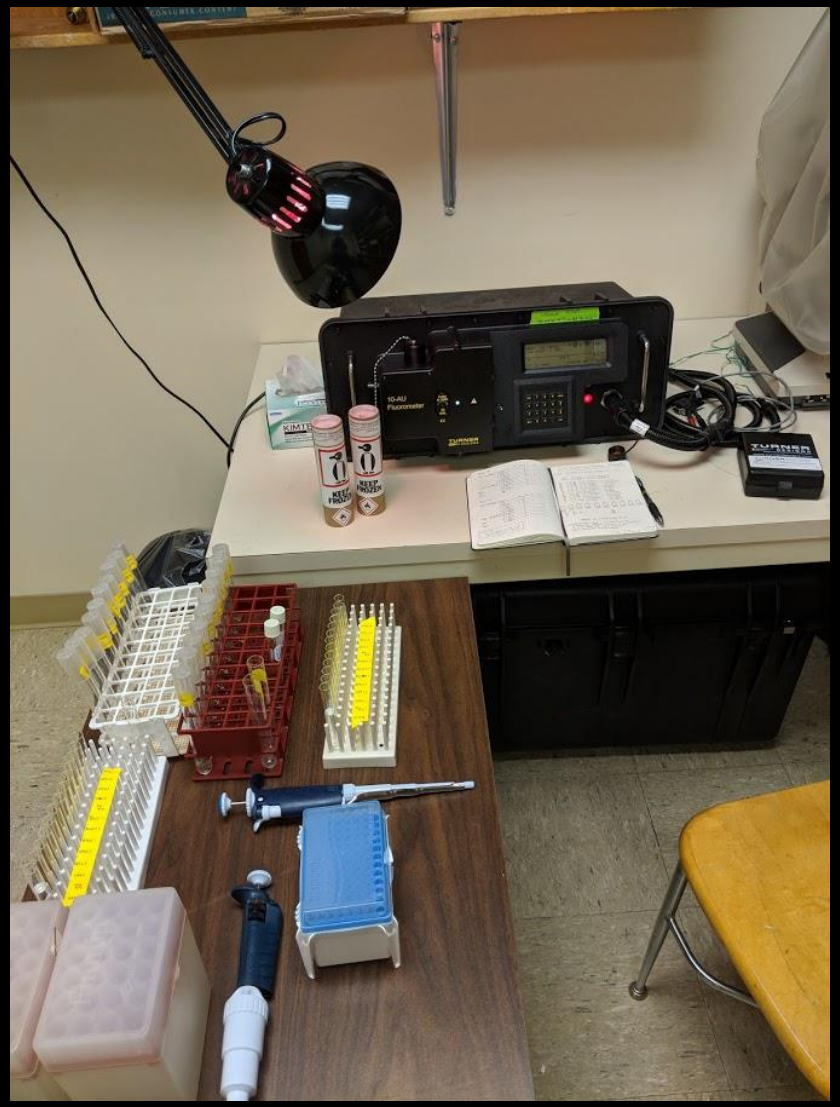
250 ml

Transport


200.L.02-11-2018. E
300 mL "F"
01-10-18
400.L.19-11-2018. R
300.L.19-10-2018. B
300.L.19.10-2018. B
300.L.14.9.2018. B
300.L.19-10-2018. R
450.L.19.11.2018. P
300.L.14.9.18. R.
400 mL "E"
01-10-18
125.L.14.9.18. R
200.L.02.11.2018. F
E
275.L.30.8.18
P
300.L.30.8.18

Extract in Ethanol or Acetone for 24 hrs

Read samples on a Fluorometer



How does a Fluorometer work?

- Fluorometers measure ‘Fluorescence’

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- Fluorescence is when a compound absorb a specific wavelength of light and almost instantaneously emits a longer wavelength of light

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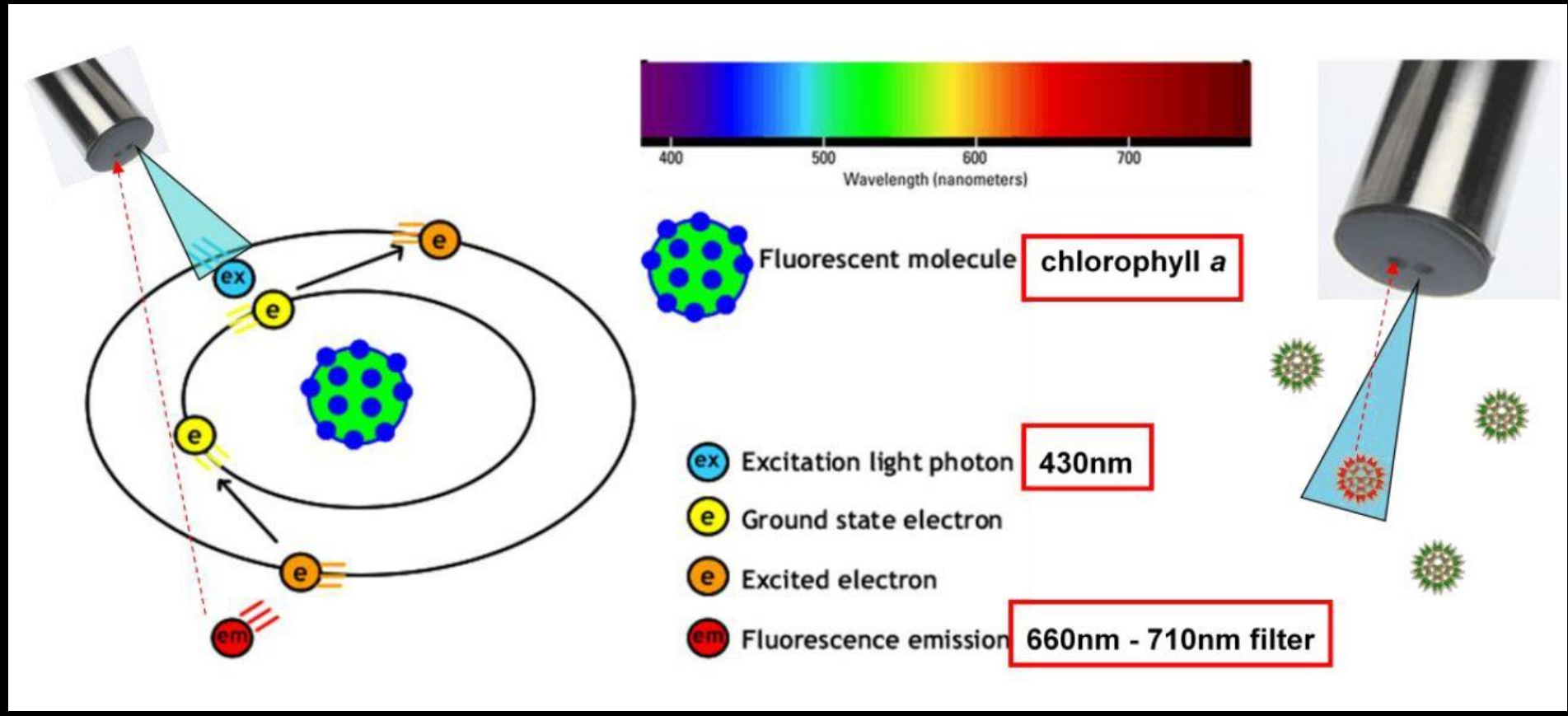

Our fluorometer is “shooting” 440nm and “reading” 685nm

Acidification

Separates the intact algae from the "dead pieces" by converting *Chlorophyll* to *Chlorophyllide.*

Chlorophyllide does not fluores.

Fluorescence measures *Chlorophyll* and *Phaeophytin*

Chlorophyll = Living and contributing to the system

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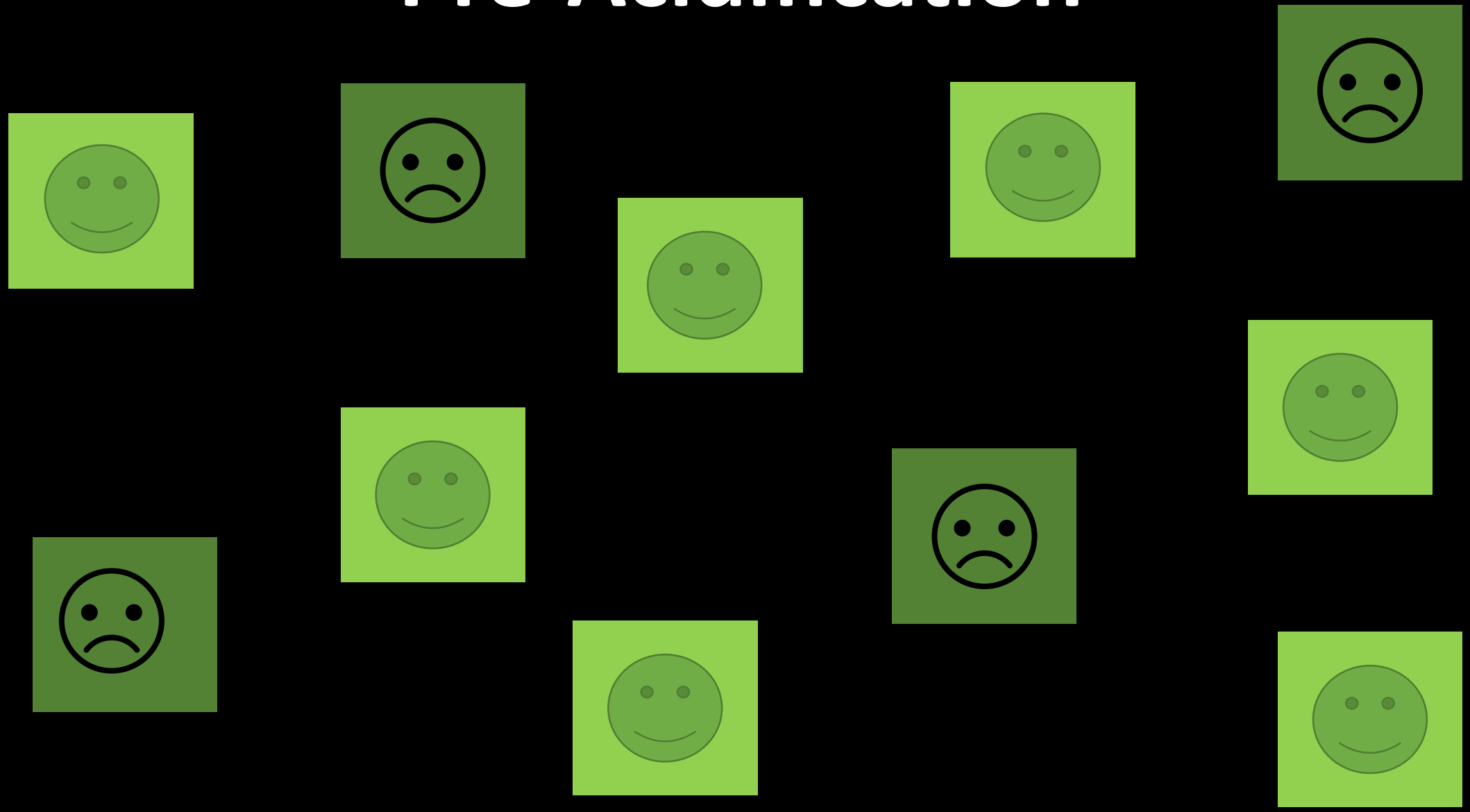
Chlorophyll = Living and contributing to the system

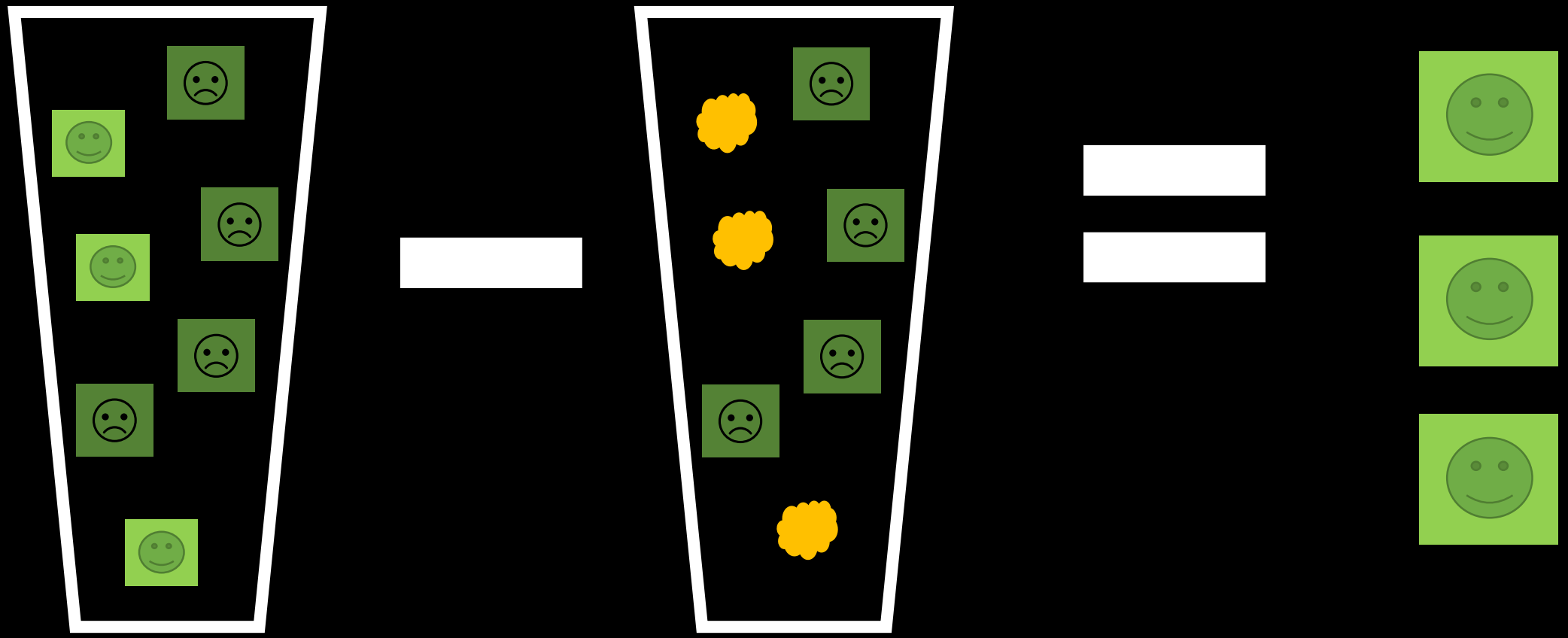
Phaeophytin = Chemical compound in Photosystem II

Pre-Acidification



Post-Acidification

Pre-Acidification - Post-Acidification = Chl *a*



Final Calculations

$$\frac{\text{Volume of water filtered (ml)}}{\text{Volume of extraction liquid used (ml)}} = \text{Concentration Factor}$$

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$$\frac{\text{Volume of water filtered (ml)}}{\text{Volume of extraction liquid used (ml)}} = \text{Concentration Factor}$$

$$\frac{\text{(Pre-Acid)} - \text{(Post-Acid)}}{\text{Concentration Factor}} = \text{Final Chl a } (\mu g/L)$$

Conclusion Questions

1. What are the basic steps in measuring Chlorophyll *a*?

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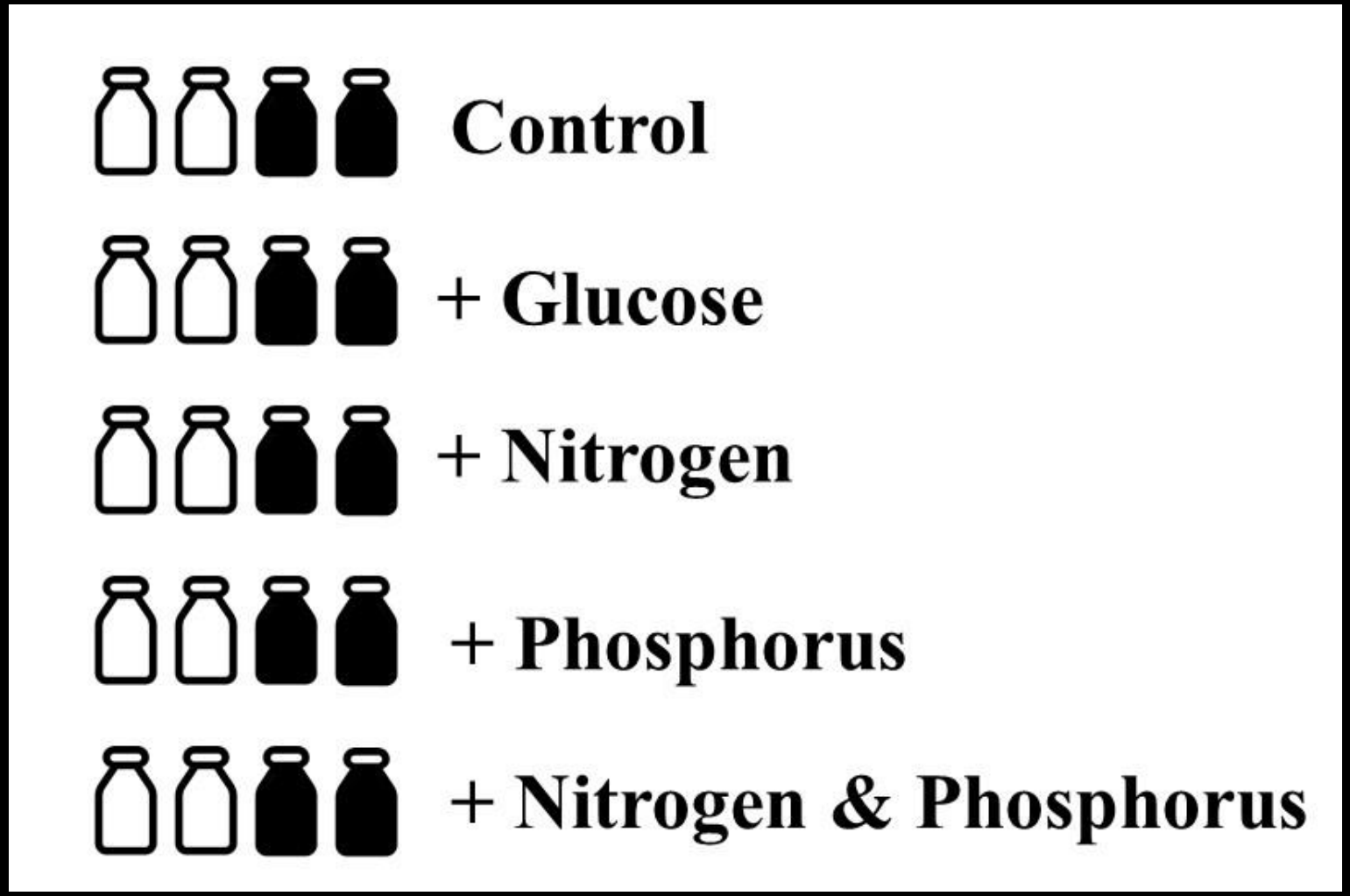
2. What is the difference between Chlorophyll, Chlorophyllide and Phaeophytin?

Conclusion Questions

1. What are the basic steps in measuring Chlorophyll *a*?
2. What is the difference between Chlorophyll, Chlorophyllide and Phaeophytin?
3. Briefly explain the acidification step.

Nutrient Enrichment Bioassay



Nutrient Enrichment Bioassay

- Very similar to the Metabolism Lab but instead of measuring GPP, NEP and Respiration, we measure Chl *a.*
- Can be done in the lab an in the field.

Nutrient Enrichment Bioassay

Nutrient Enrichment Bioassay: Honduras style

Ctl	Ctl	N 1	NP 10	P 10	P 1	N 2	Ctl	P 2	N 10	N 5	P 5	NP 10	Ctl	NP 2	Ctl
NP 2	N 5	Ctl	P 1	NP 10	NP 2	N 10	P 5	P 10	N 1	Ctl	P 2	N 2	NP 10	Ctl	Ctl
Ctl	P 10	NP 10	NP 2	Ctl	P 2	P 5	NP 2	NP 2	N 2	N 10	Ctl	P 1	NP 10	N 5	N 1

Without Grazers

	Controls	1 times	2 times	5 times	10 times
	■	■	■	■	■
Plus N	■	■	■	■	■
	■	■	■	■	■
	■	■	■	■	■
Plus P	■	■	■	■	■
	■	■	■	■	■
Plus N and P			■		■
			■		■
			■		■

Nutrient Enrichment Bioassay

1. What questions can we answer with nutrient bioassays?

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LETTER

Global analysis of nitrogen and phosphorus limitation of primary producers in freshwater, marine and terrestrial ecosystems

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Abstract

The cycles of the key nutrient elements nitrogen (N) and phosphorus (P) have been massively altered by anthropogenic activities. Thus, it is essential to understand how photosynthetic production across diverse ecosystems is, or is not, limited by N and P. Via a large-scale meta-analysis of experimental enrichments, we show that P limitation is equally strong across these major habitats and that N and P limitation are equivalent within both terrestrial and freshwater systems. Furthermore, simultaneous N and P enrichment produces strongly positive synergistic responses in all three environments. Thus, contrary to some prevailing paradigms, freshwater, marine and terrestrial ecosystems are surprisingly similar in terms of N and P limitation.

Keywords

Ecosystem, meta-analysis, nitrogen, nutrient limitation, phosphorus, primary production.