Using eDNA tools to investigate a mysterious algae bloom: An update on Highland (Duck) Lake

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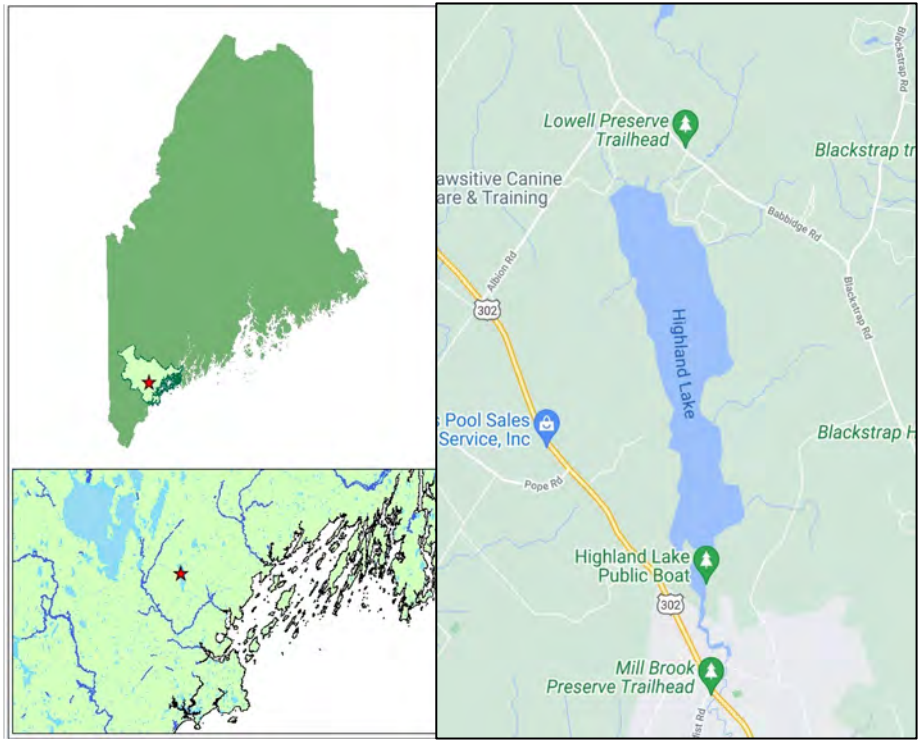
Supported by National Science Foundation award #OIA-1849227 to Maine EPSCoR at the University of Maine

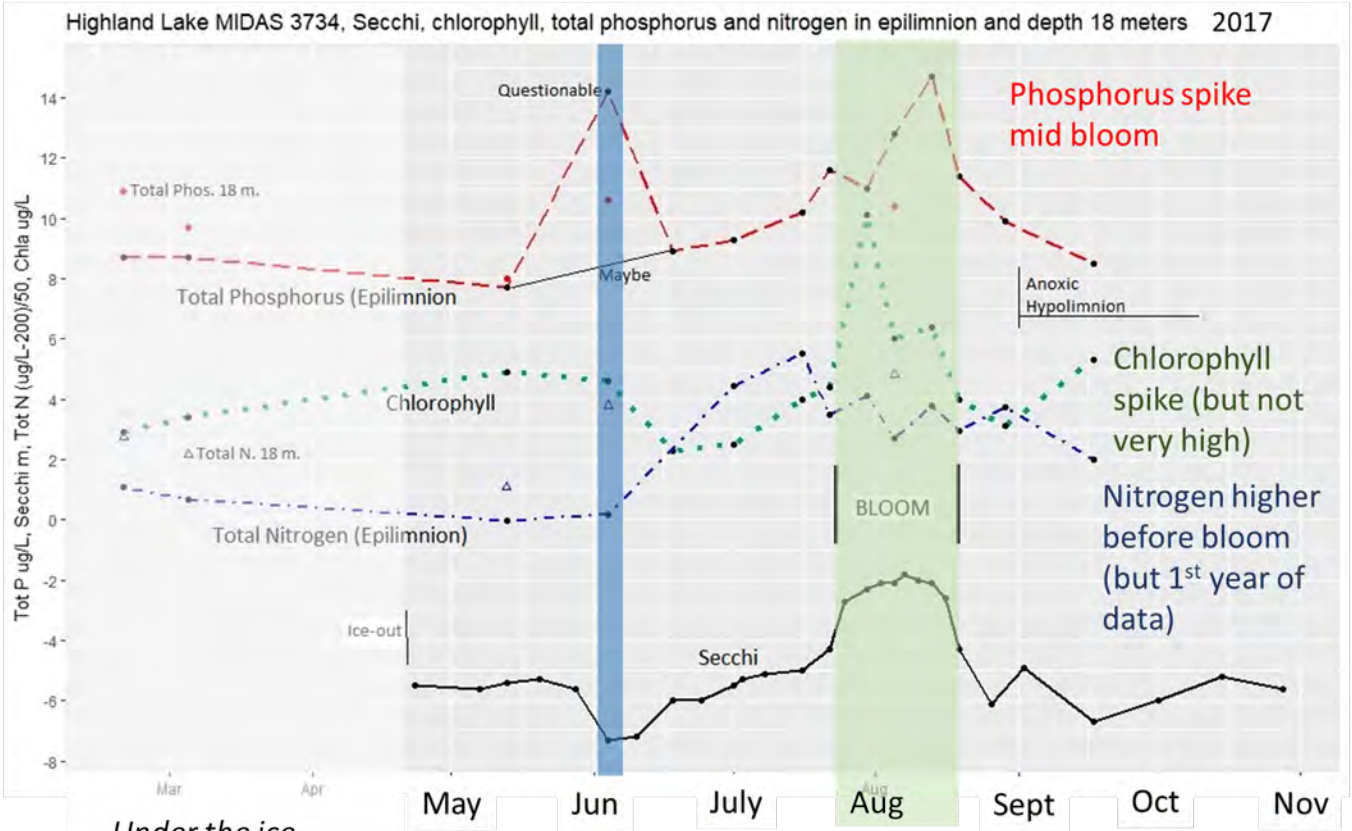
- Highly developed area
- Previously listed (1998-2010) as impaired by Maine-DEP due to deteriorating trophic state
- Active Lake Association water quality team
- Experienced an unusual nuisance bloom, coinciding with the first 4 years of high numbers of spawning anadromous alewife



Highland (Duck) Pond

MAINE eDNA

Highland Lake experienced extended algae blooms from 2014-2017



Under the ice

Alewife enter

Bloom – preliminarily identified as *Cyanobium* (picocyanobacteria, <3 µm)

Bloom years 2014 - 2017

Persisted for up to 3 weeks, late July in to Aug.

Secchi depth < 2.5

Source: Highland Lake Association (K. Williams); data from HLA, Maine DEP, USM

Cause(s) of algae bloom?

Excess nutrients?

Unusual phytoplankton species?

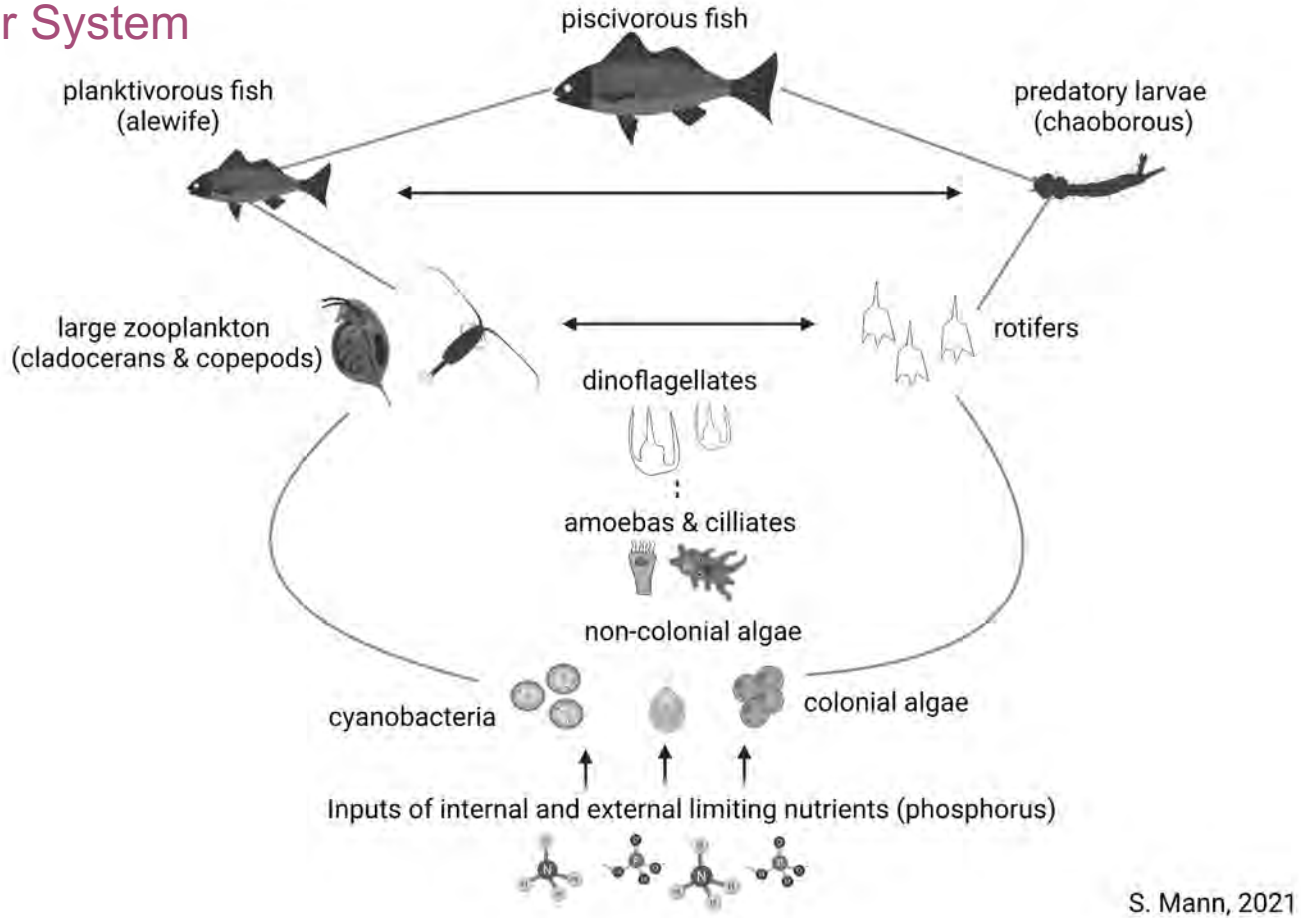
Trophic cascade triggered by consumption of herbivorous zooplankton by alewife?

Unlikely: TP levels lower than those associated with nuisance blooms

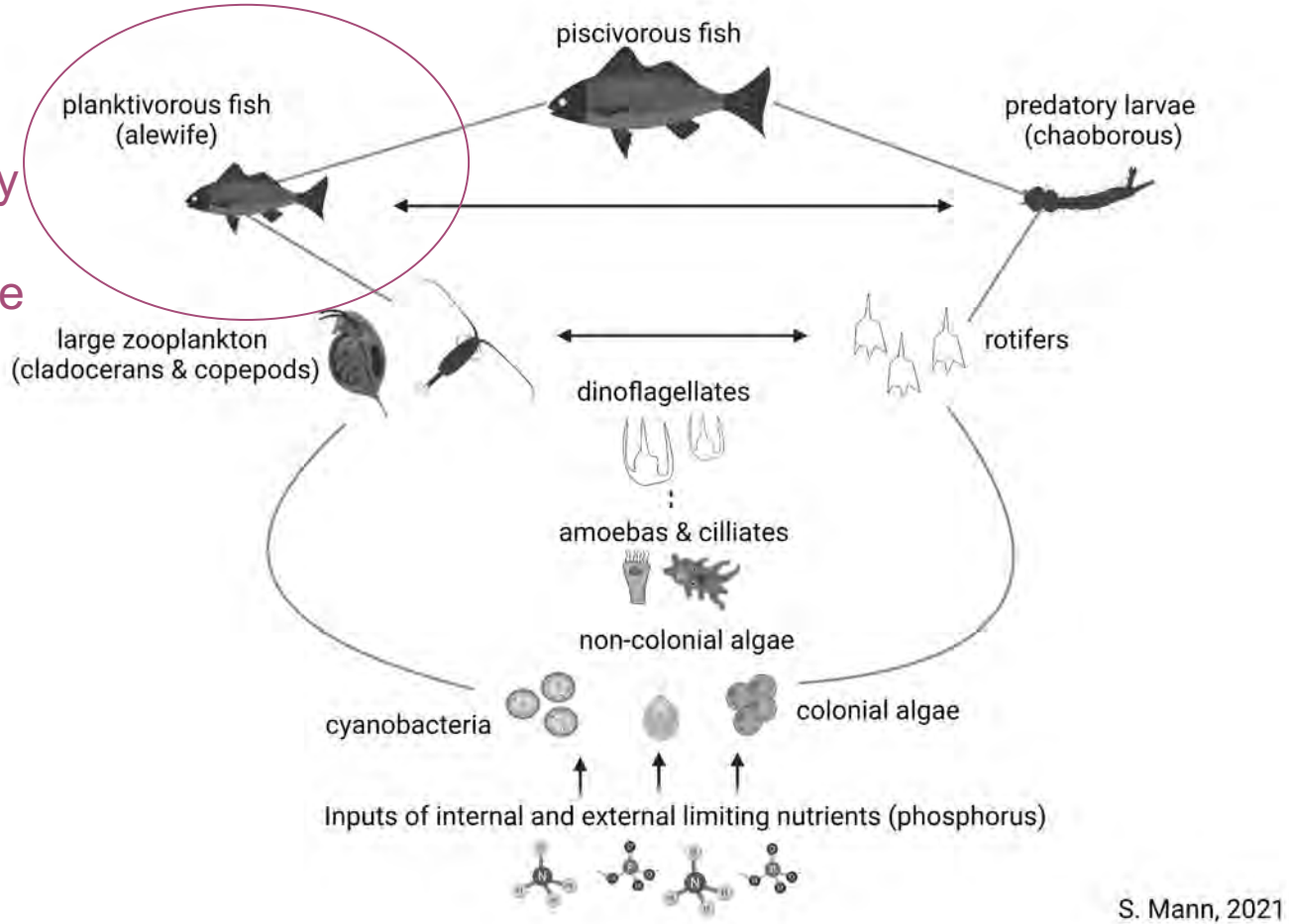
Preliminary observations suggested that the bloom was caused by a picocyanobacteria - Today's talk!

Coming soon!

Freshwater System

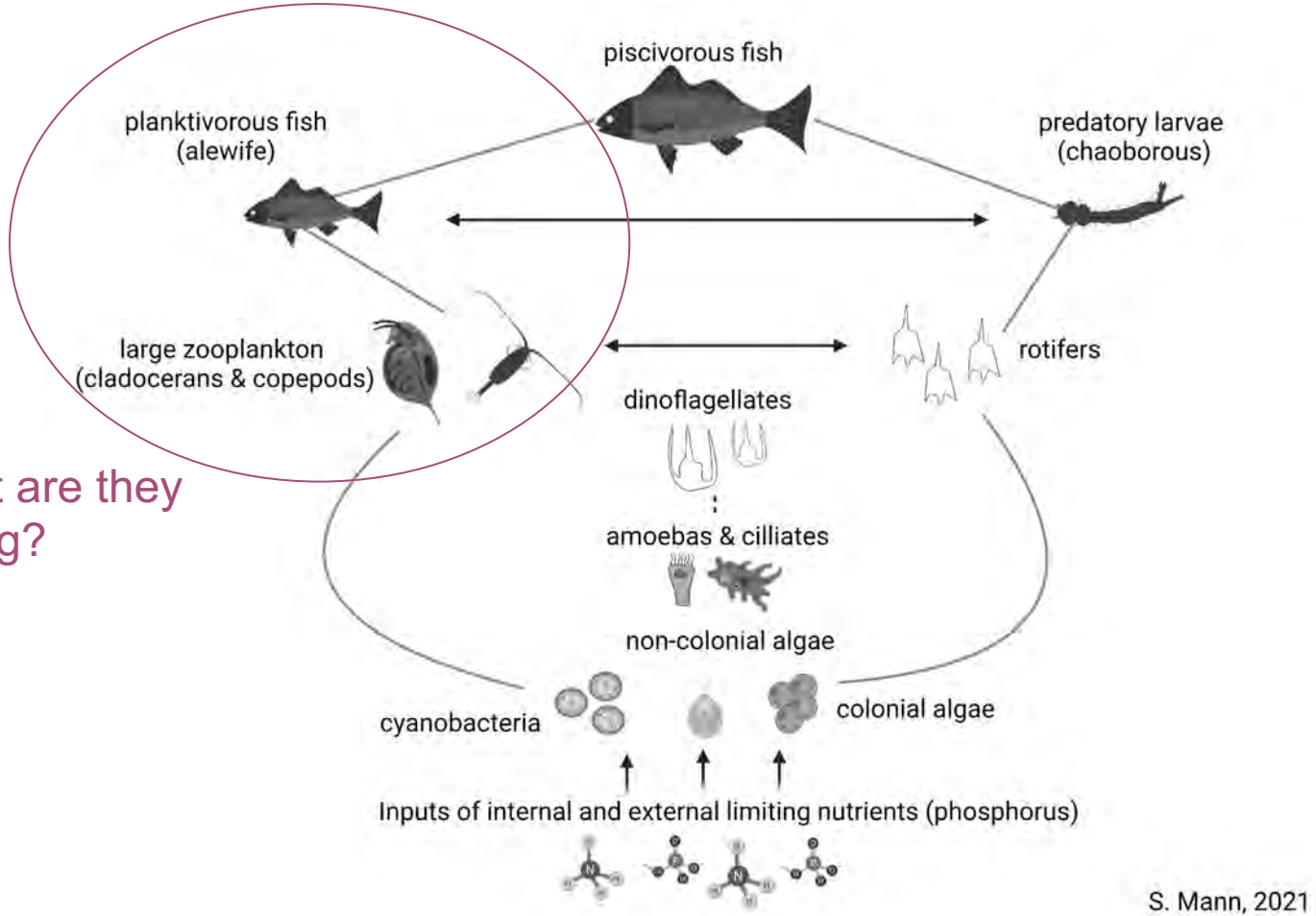


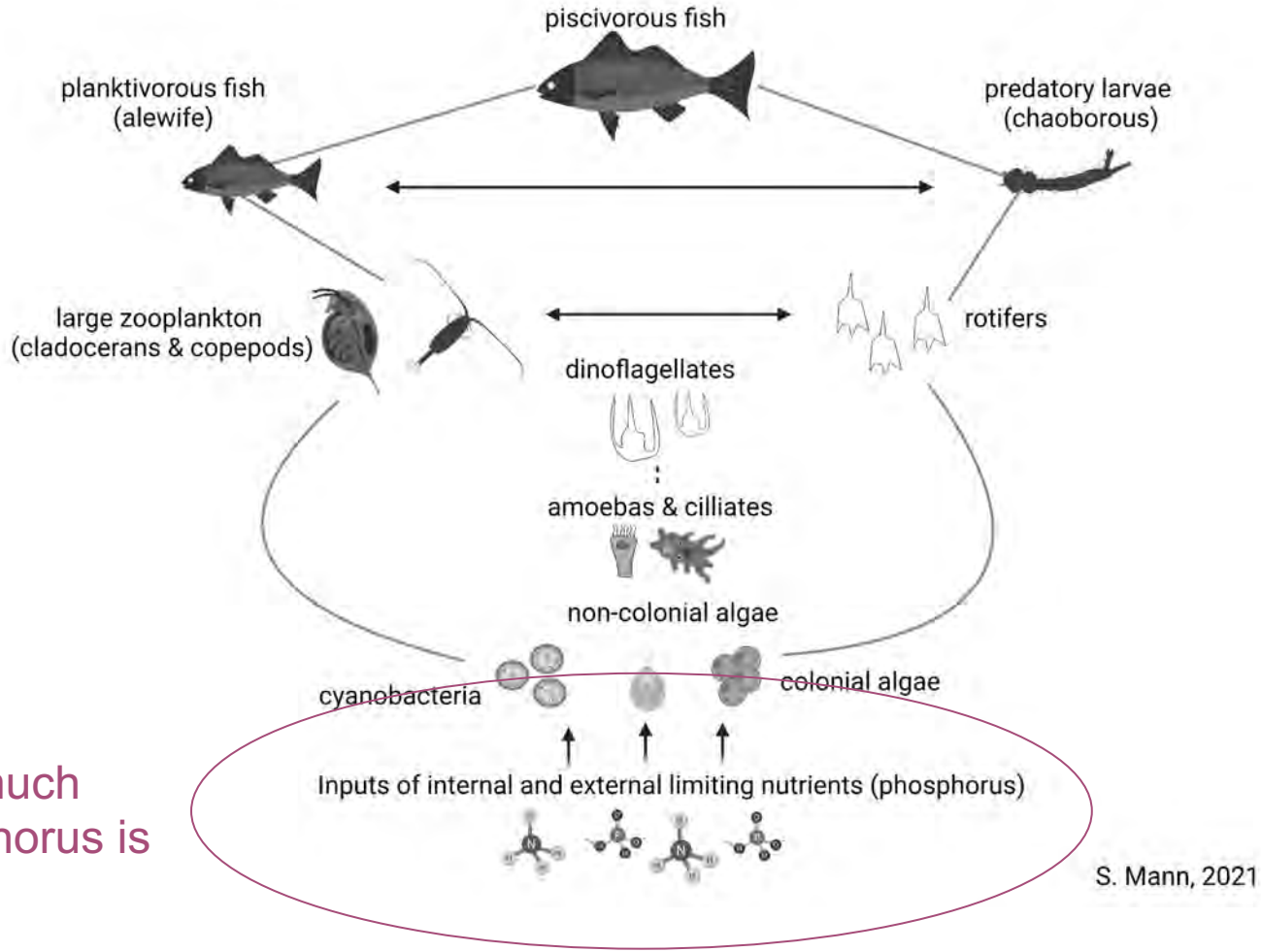
How many juvenile alewife are there?



S. Mann, 2021

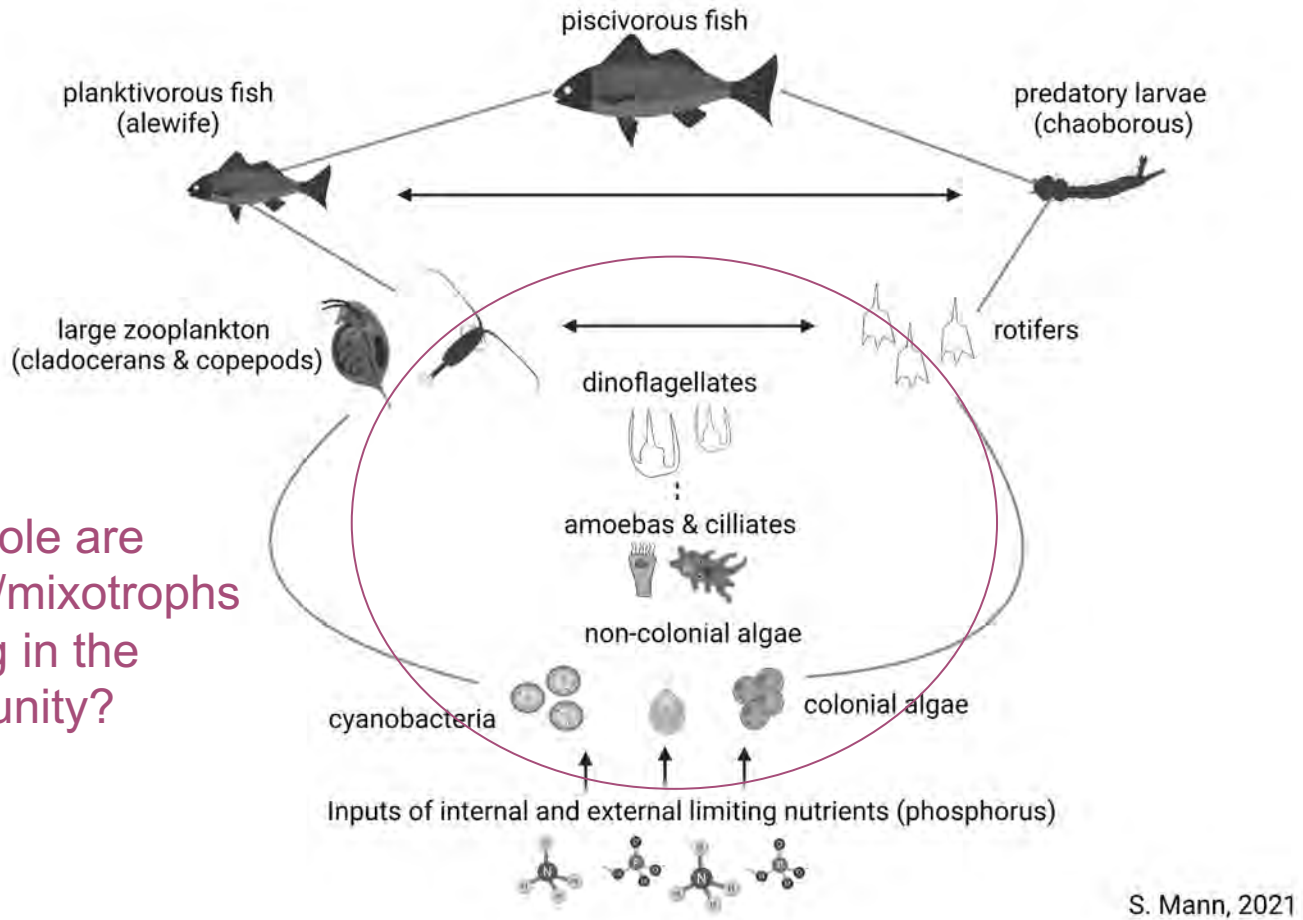
What are they eating?

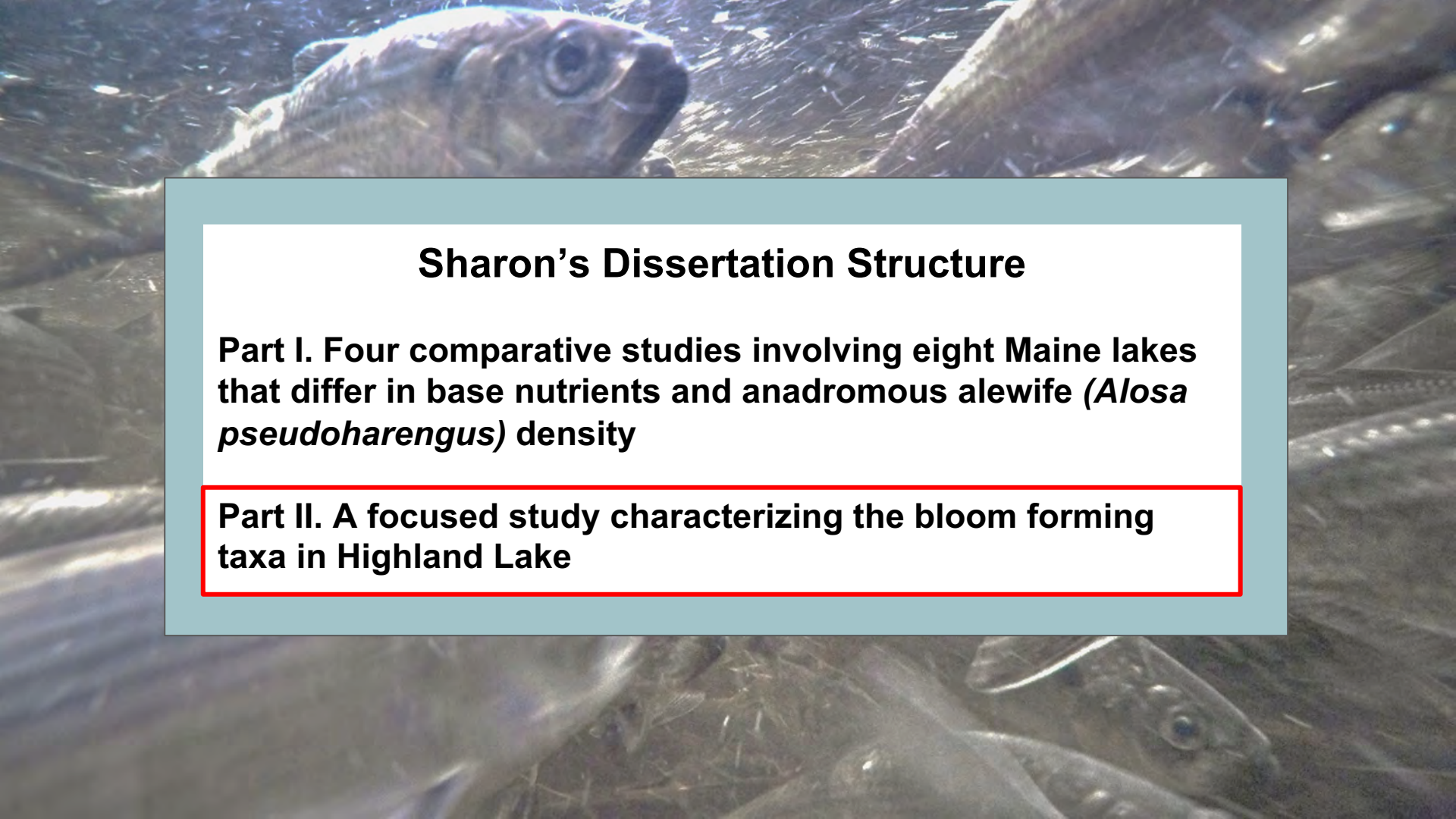




How much phosphorus is there?

What role are hetero/mixotrophs playing in the community?





Sharon's Dissertation Structure

Part I. Four comparative studies involving eight Maine lakes that differ in base nutrients and anadromous alewife *(Alosa pseudoharengus)* density

Part II. A focused study characterizing the bloom forming taxa in Highland Lake

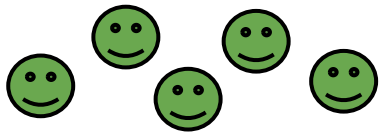
Why is this work needed?

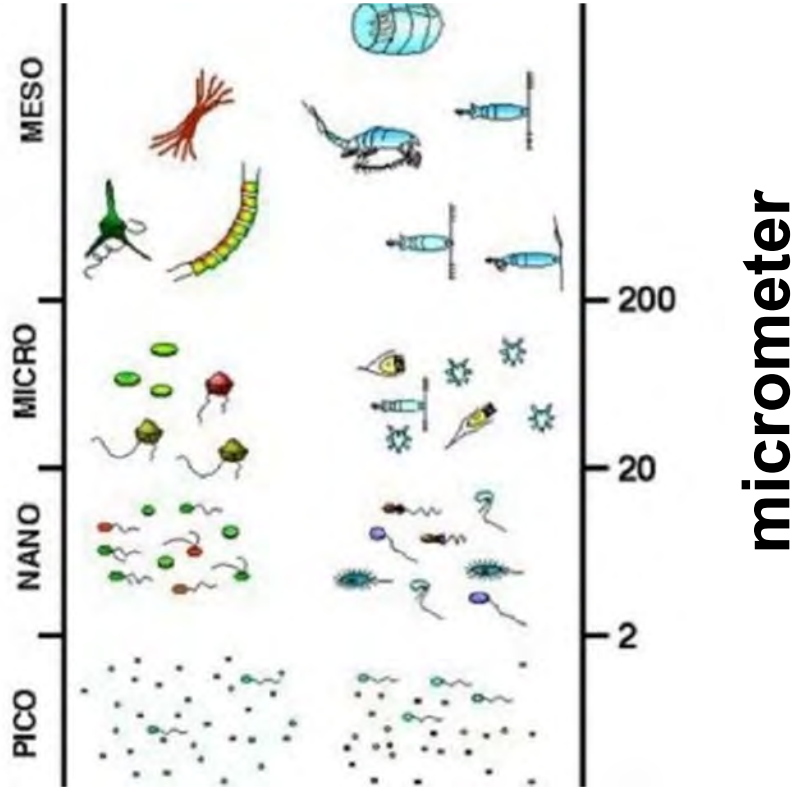
- Pico eukaryotes and pico cyanobacteria have different effects on ecosystems (cyanobacteria such as *Cyanobium spp.* can produce toxins)

Why use eDNA?

- Unable to identify the taxa responsible for decreasing water transparency using microscopy

L.G.B.





Alcaraz & Calbet (2003)

MAINE eDNA

Pico and nano phytoplankton are difficult to identify

Genetic material obtained directly from environmental samples without any obvious signs of the biological source material (Thomsen and Willerslev, 2015)

Where does eDNA come from?

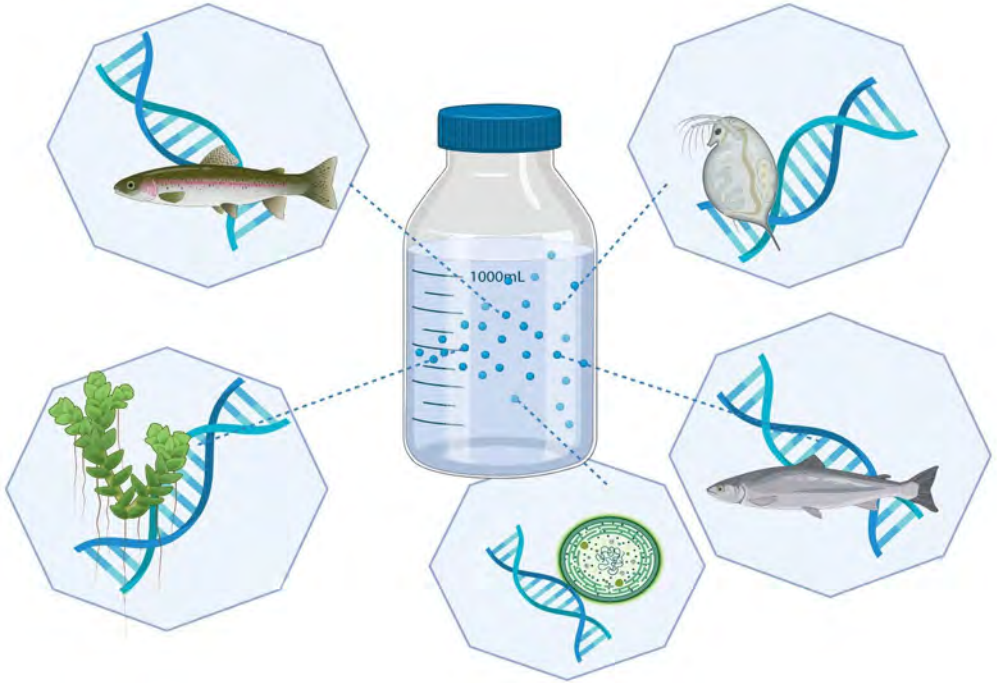
- Cellular decomposition
- Whole shed cells
- Whole microorganisms

Where is eDNA found?

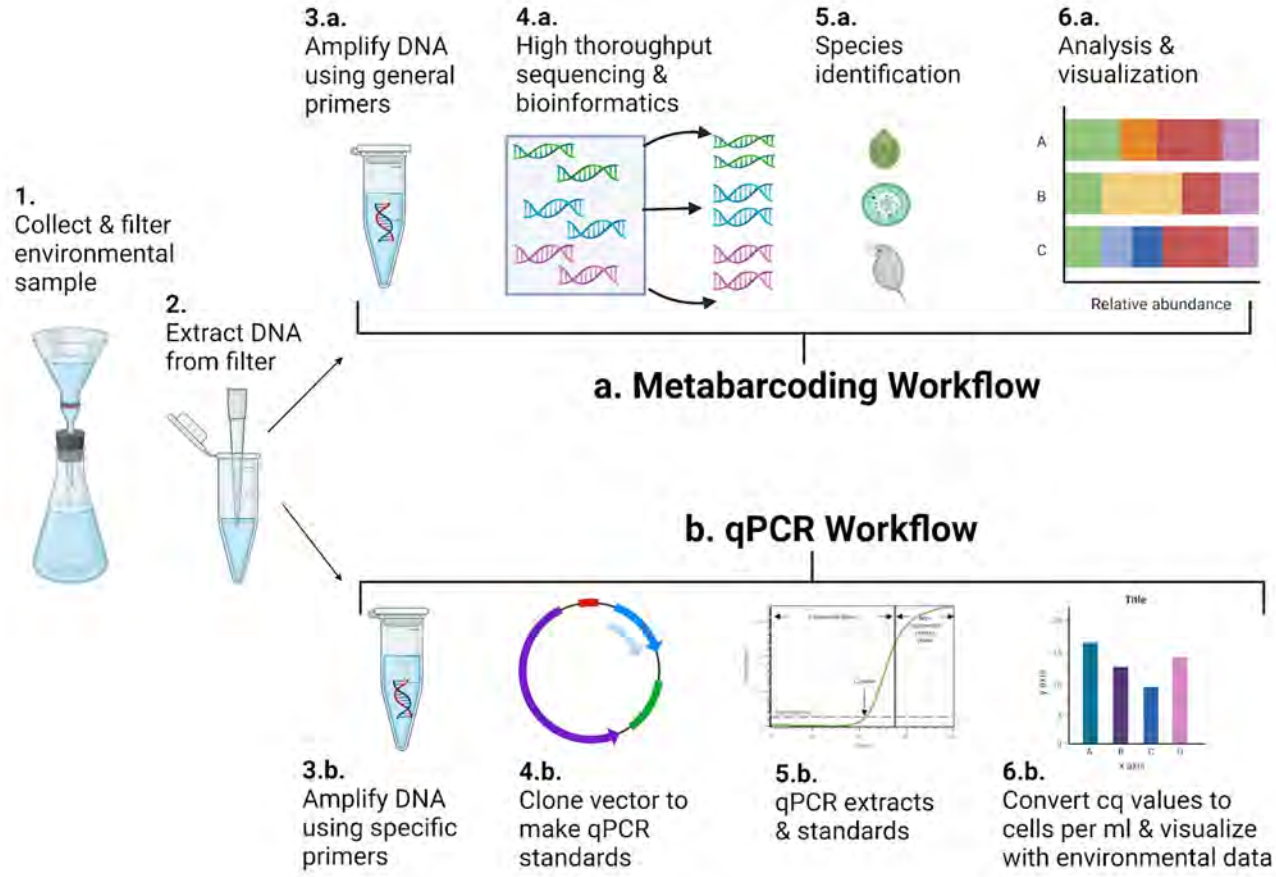
- Water
- Soils
- Air

How is eDNA used?

- Community characterization (metabarcoding)
- Targeted detection & quantification (qPCR)

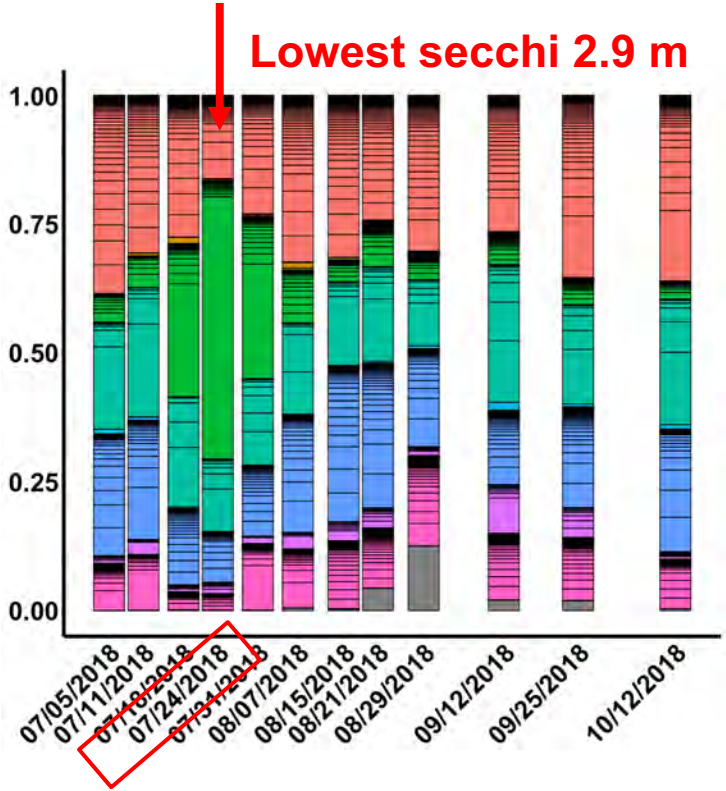


What is environmental DNA?



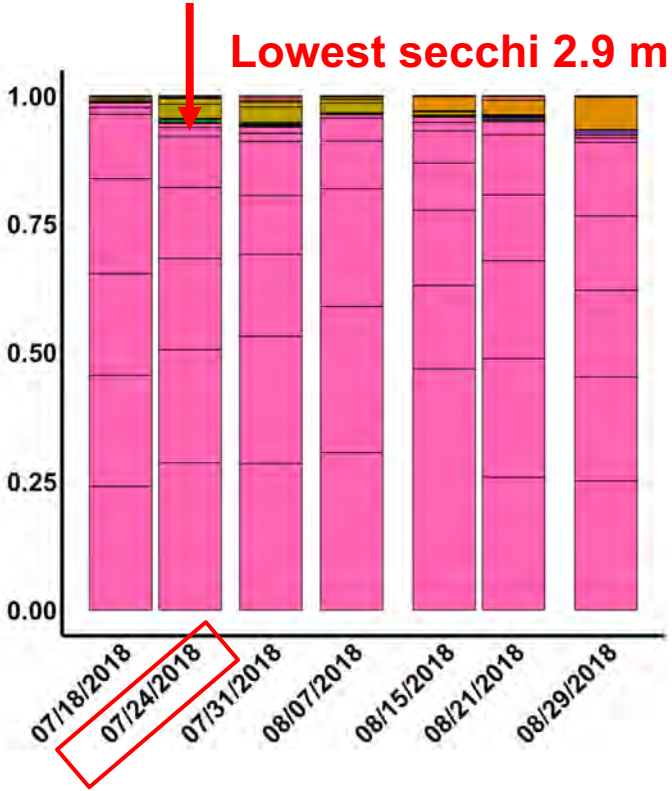
MAINE eDNA

Using a two-tiered approach with eDNA tools

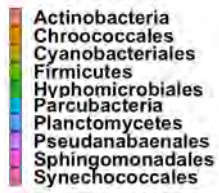


Super Order Eukaryote (18s)

Rhexinema spp.
(formerly known as *Helicodictyon*)

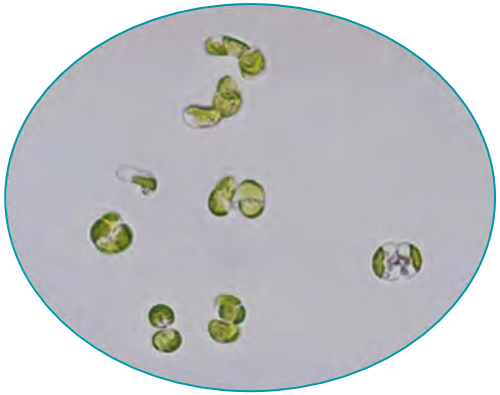


Order Bacteria (16s)

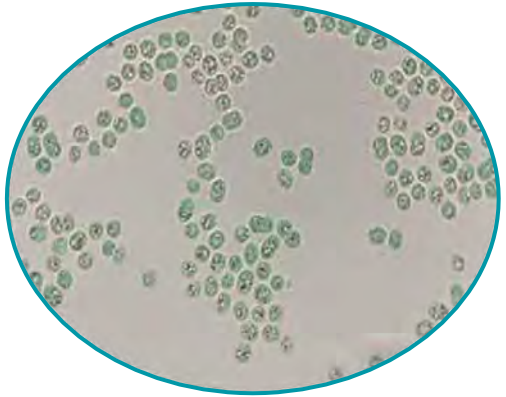


Cyanobium spp.
(pico-cyanobacteria)

Figures provided by Robin Sleith

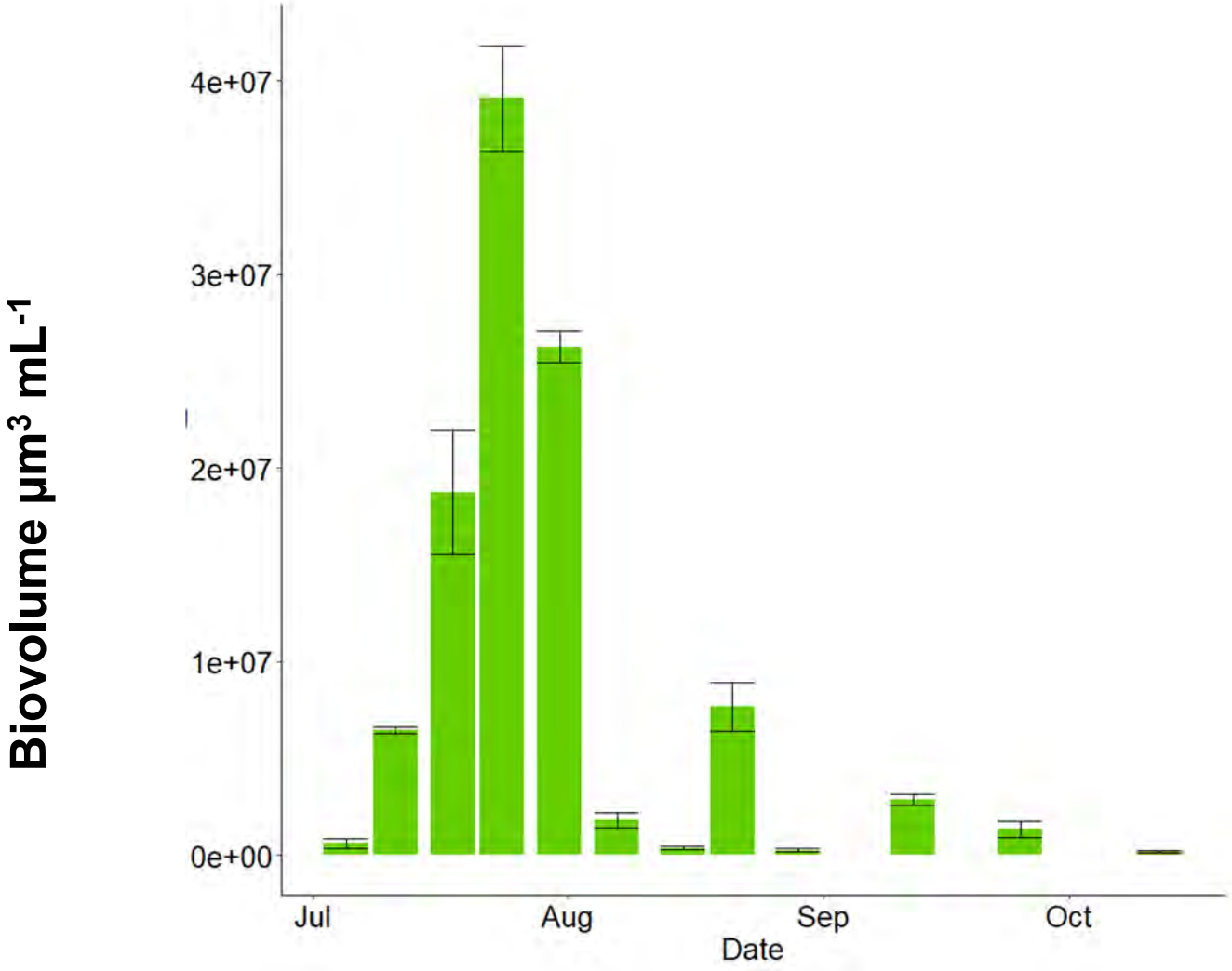


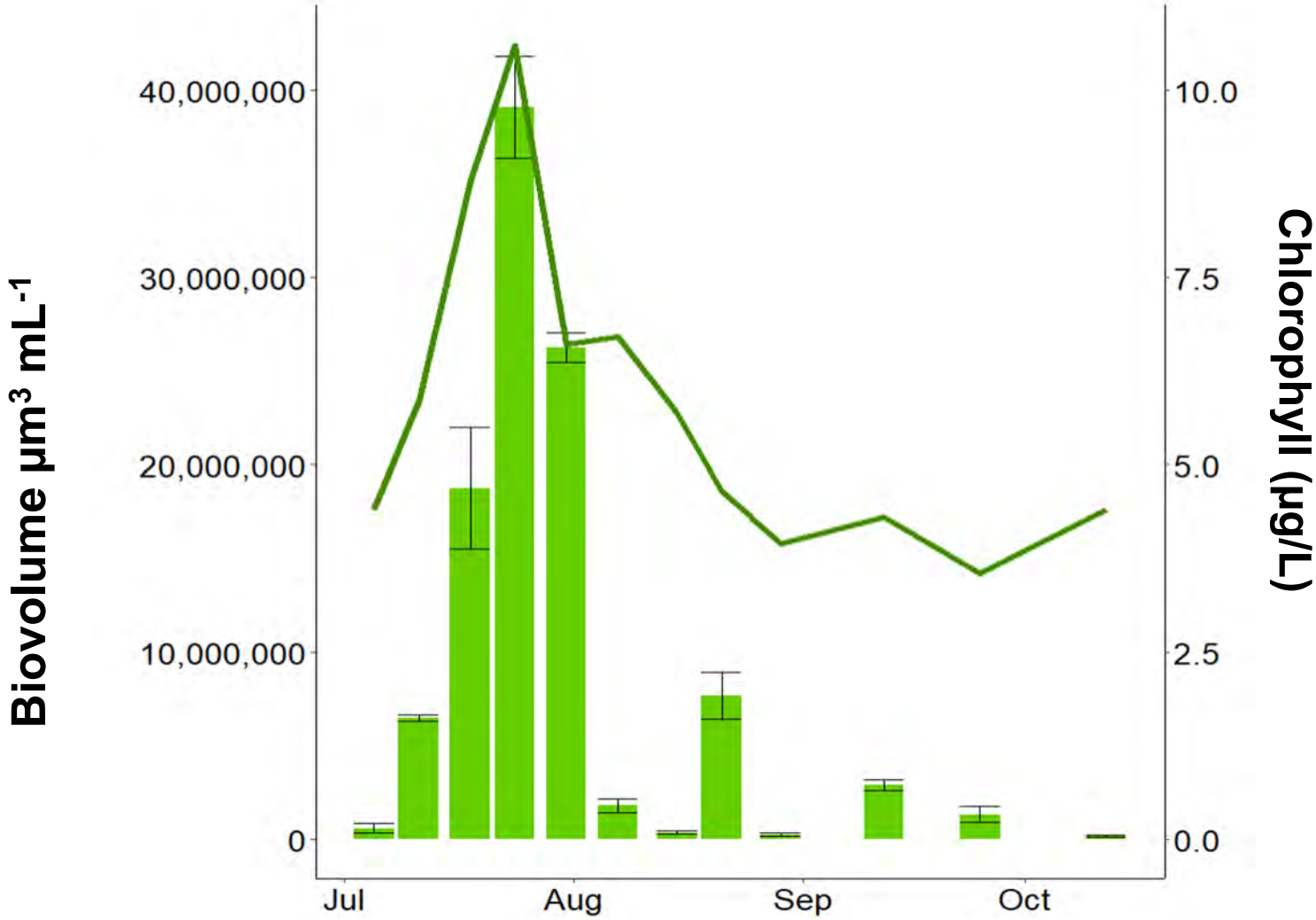
Rhexinema (genus of green algae)
One cell ≤ 10 µ m

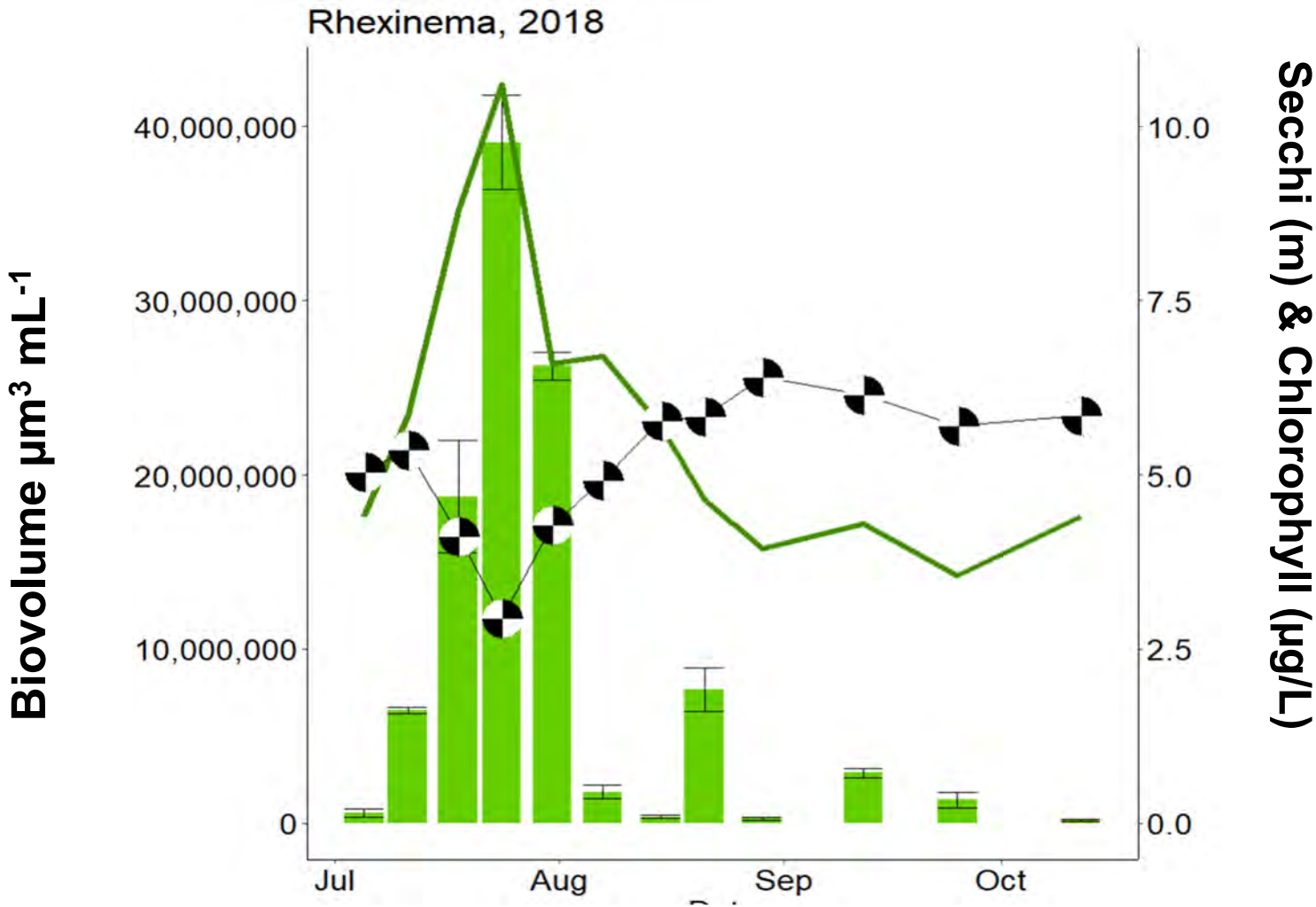


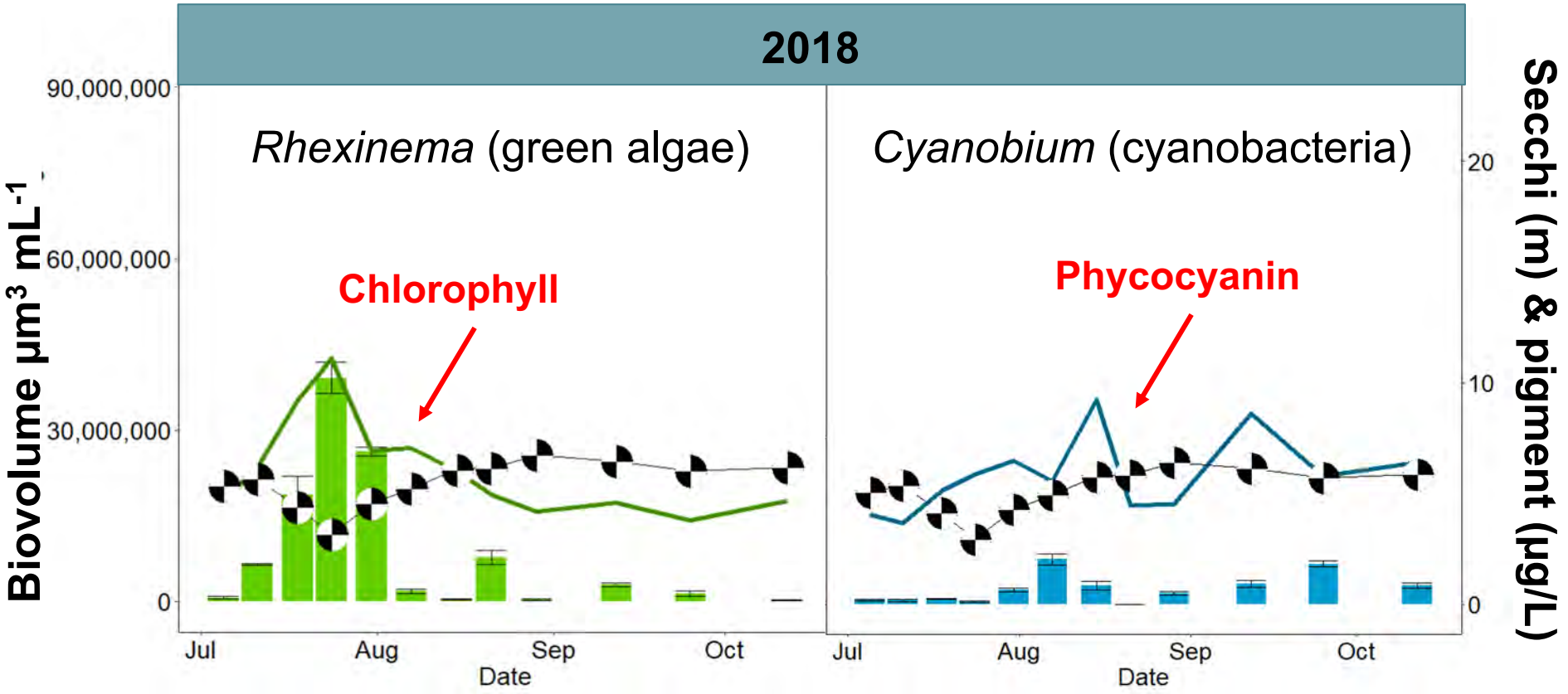
Cyanobium (genus of cyanobacteria)
One cell ≤ 2 µ m

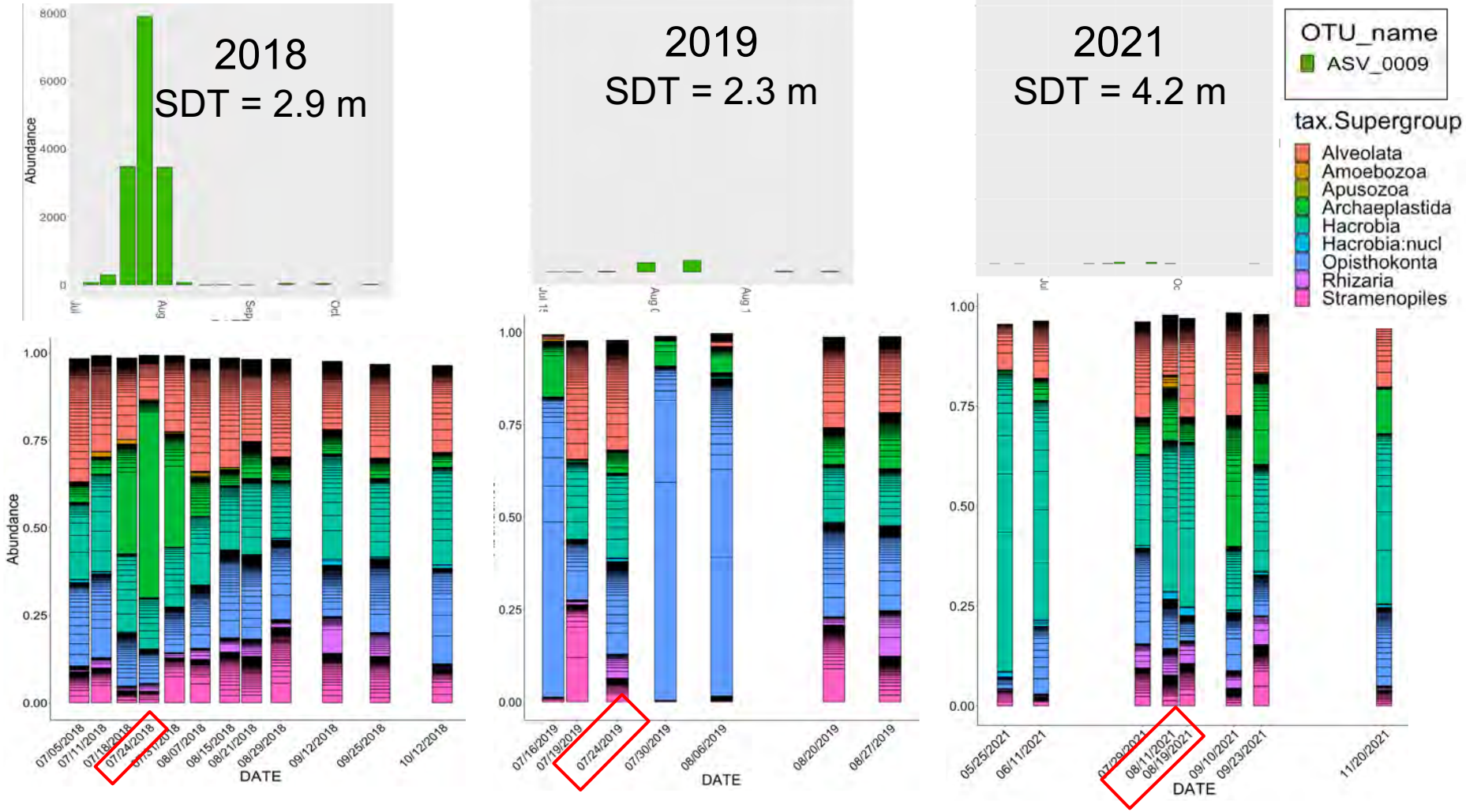
- Design qPCR assays
- Quantify cells volume in epilimnetic water samples

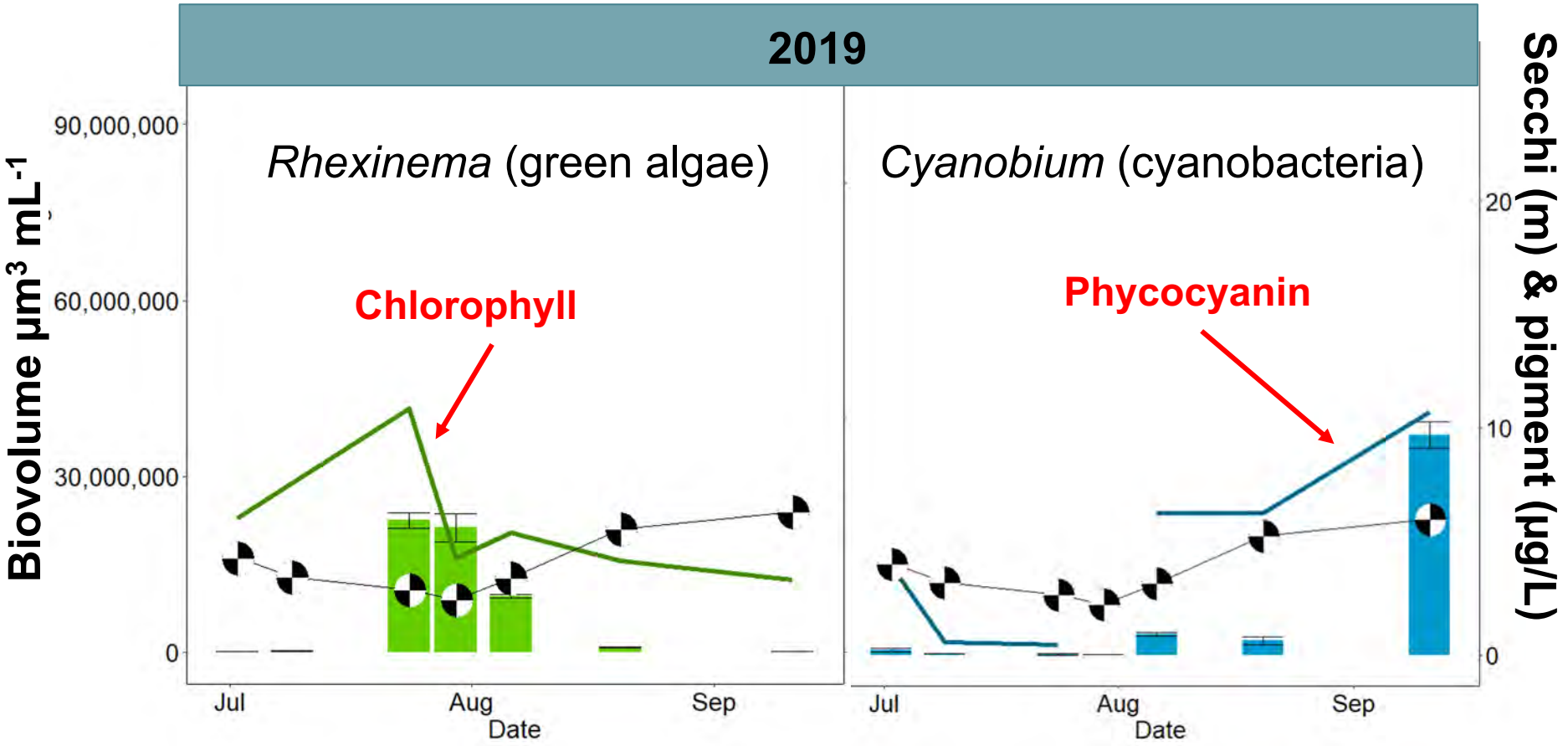

Biovolume µm³ mL⁻¹
4e+07
3e+07
2e+07
1e+07
0e+00
Jul
Aug
Sep
Oct
Date

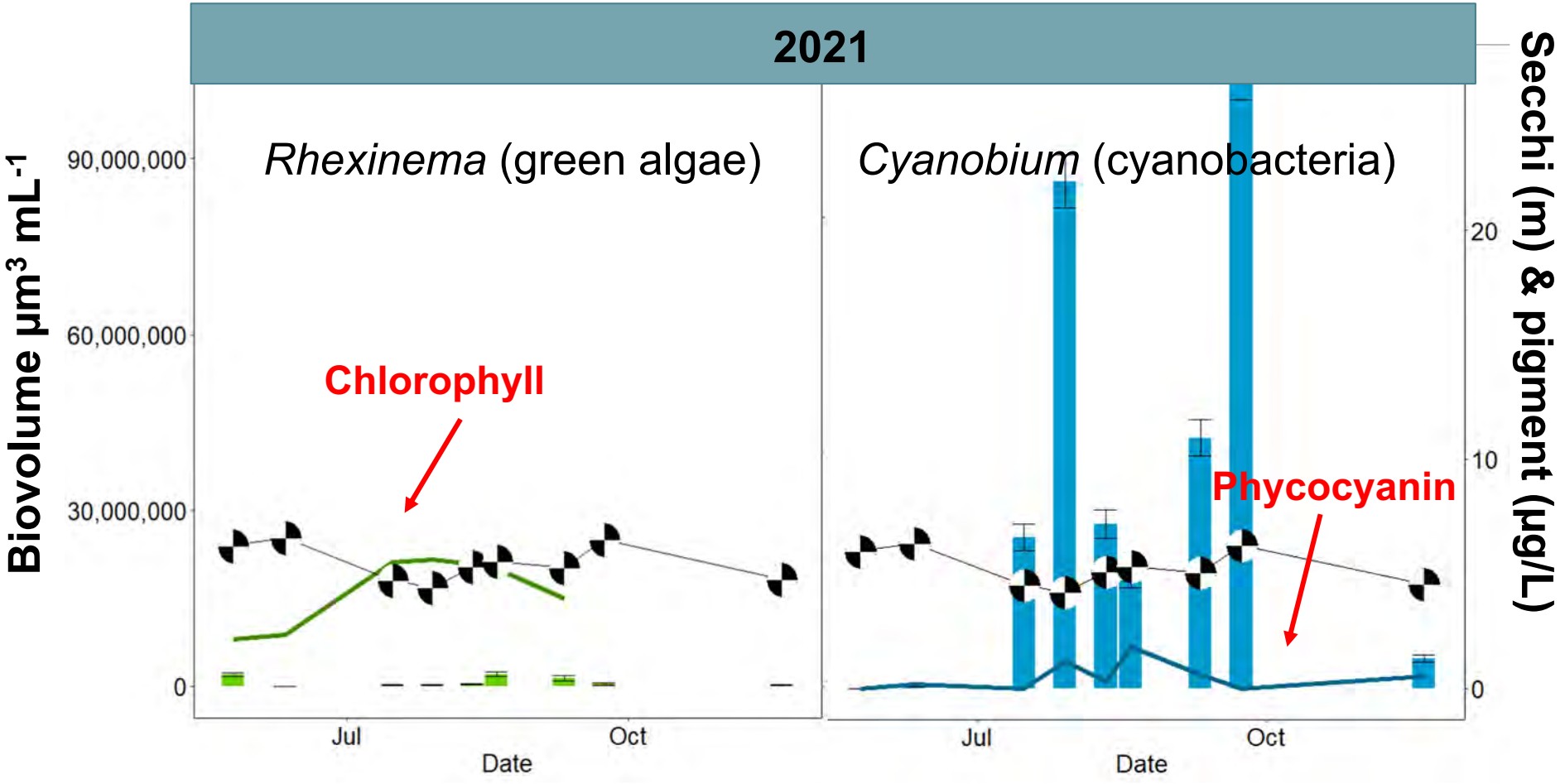

Biovolume µm³ mL⁻¹
40,000,000
30,000,000
20,000,000
10,000,000
0
Jul
Aug
Sep
Oct
10.0
7.5
5.0
2.5
0.0
Chlorophyll (µg/L)

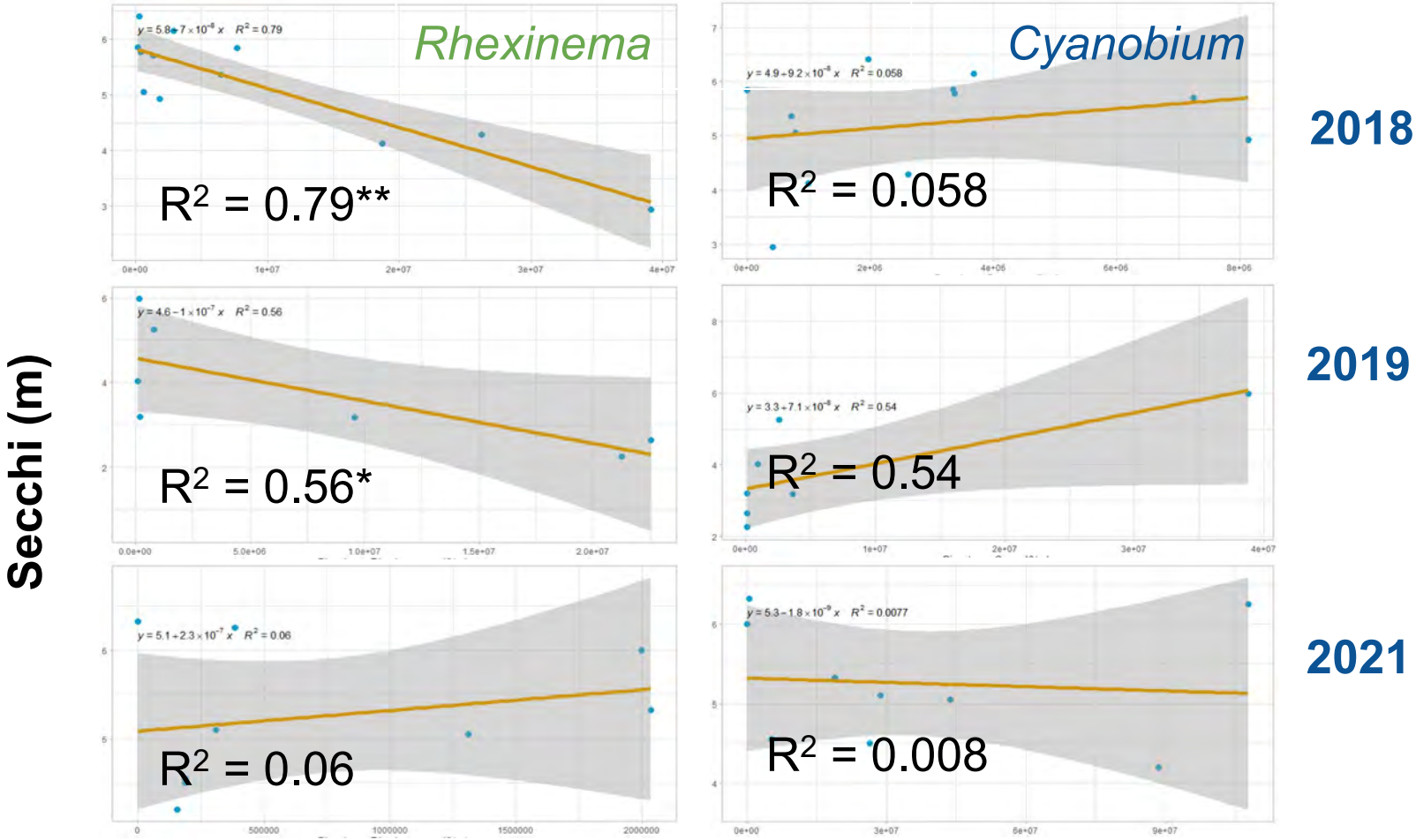

Rhexinema, 2018
Biovolume µm³ mL⁻¹
Secchi (m) & Chlorophyll (µg/L)
40,000,000
30,000,000
20,000,000
10,000,000
0
10.0
7.5
5.0
2.5
0.0
Jul
Aug
Sep
Oct


2018
Rhexinema (green algae)
Cyanobium (cyanobacteria)
Chlorophyll
Phycocyanin
Biovolume µm3 mL-1
90,000,000
60,000,000
30,000,000
0
Secchi (m) & pigment (µg/L)
20
10
0
Jul
Aug
Sep
Oct
Date
Jul
Aug
Sep
Oct
Date

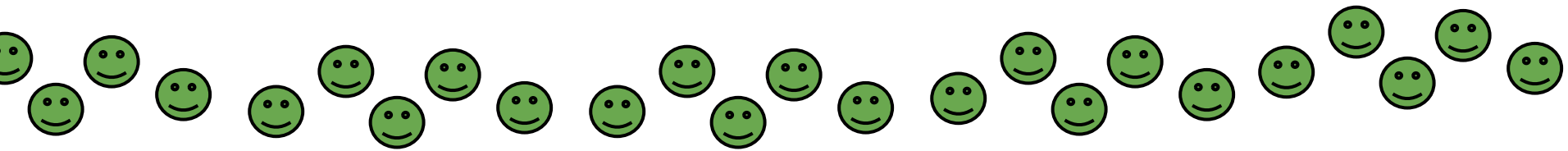

2018
SDT = 2.9 m
2019
SDT = 2.3 m
2021
SDT = 4.2 m
OTU_name
ASV_0009
tax.Supergroup
Alveolata
Amoebozoa
Apusozoa
Archaeplastida
Hacrobia
Hacrobia:nucl
Opisthokonta
Rhizaria
Stramenopiles
Abundance
DATE
07/05/2018
07/11/2018
07/18/2018
07/24/2018
07/31/2018
08/07/2018
08/15/2018
08/21/2018
08/29/2018
09/12/2018
09/25/2018
10/12/2018
07/16/2019
07/19/2019
07/24/2019
07/30/2019
08/06/2019
08/20/2019
08/27/2019
05/25/2021
06/11/2021
07/29/2021
08/11/2021
08/19/2021
09/10/2021
09/23/2021
11/20/2021

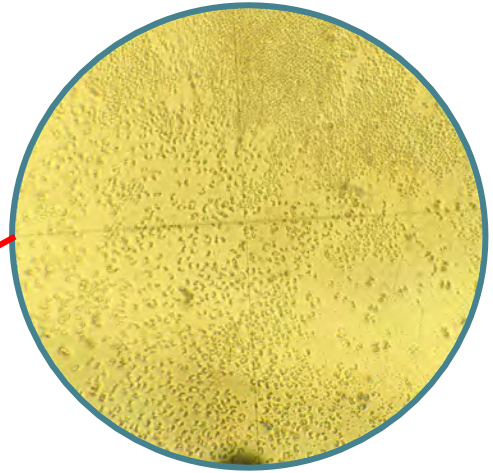

2019
Rhexinema (green algae)
Cyanobium (cyanobacteria)
Chlorophyll
Phycocyanin
Biovolume µm³ mL⁻¹
90,000,000
60,000,000
30,000,000
0
Secchi (m) & pigment (µg/L)
20
10
0
Jul
Aug
Sep
Date


2021
Rhexinema (green algae)
Cyanobium (cyanobacteria)
Chlorophyll
Phycocyanin
Biovolume µm³ mL⁻¹
90,000,000
60,000,000
30,000,000
0
Jul
Oct
Date
Secchi (m) & pigment (µg/L)
20
10
0

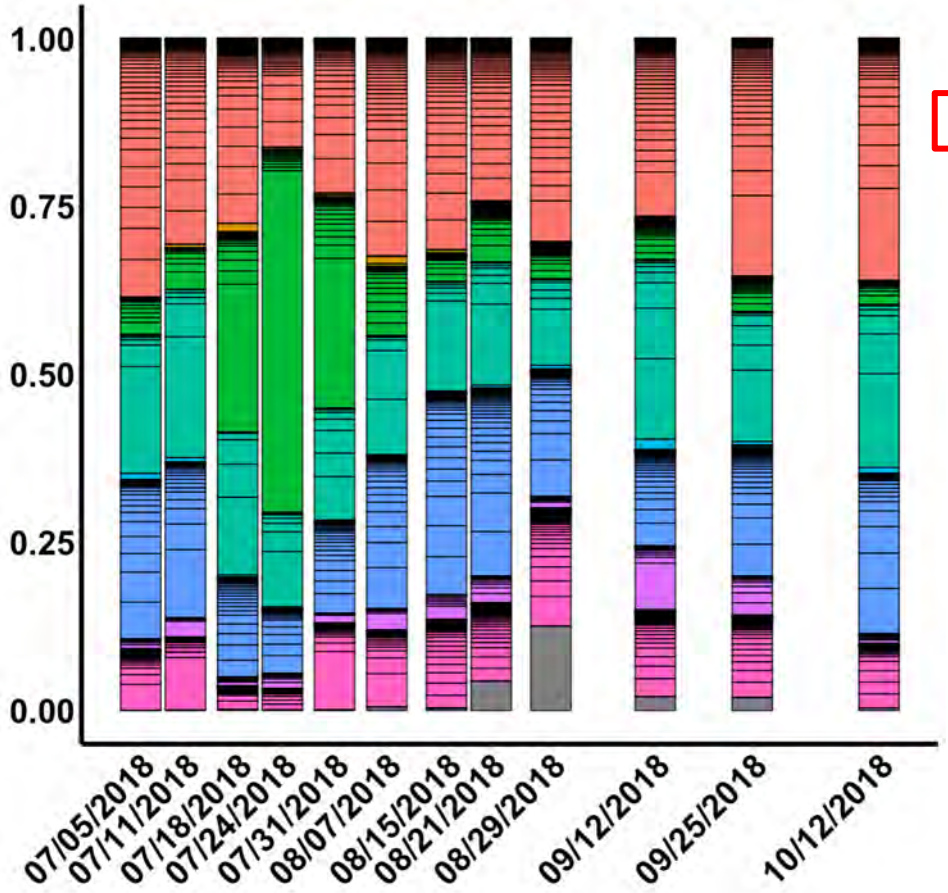

Rhexinema
Cyanobium
2018
2019
2021
R² = 0.79**
R² = 0.058
R² = 0.56*
R² = 0.54
R² = 0.06
R² = 0.008
Secchi (m)
Biovolume µm³ mL⁻¹

Next Steps

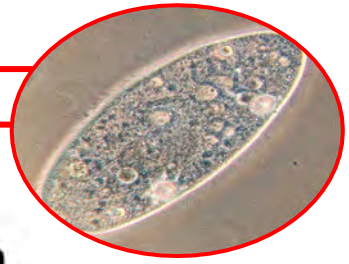




- Culture Rhexinema
- Sequence the culture's DNA
- Send culture isolates to a taxonomist


1.00
0.75
0.50
0.25
0.00
07/05/2018
07/11/2018
07/18/2018
07/24/2018
07/31/2018
08/07/2018
08/15/2018
08/21/2018
08/29/2018
09/12/2018
09/25/2018
10/12/2018


Alveolata
Amoebozoa
Apusozoa
Archaeplastida
Hacrobia
Hacrobia:nucl
Opisthokonta
Rhizaria
Stramenopiles
NA



Thank you,

Please send additional questions to:

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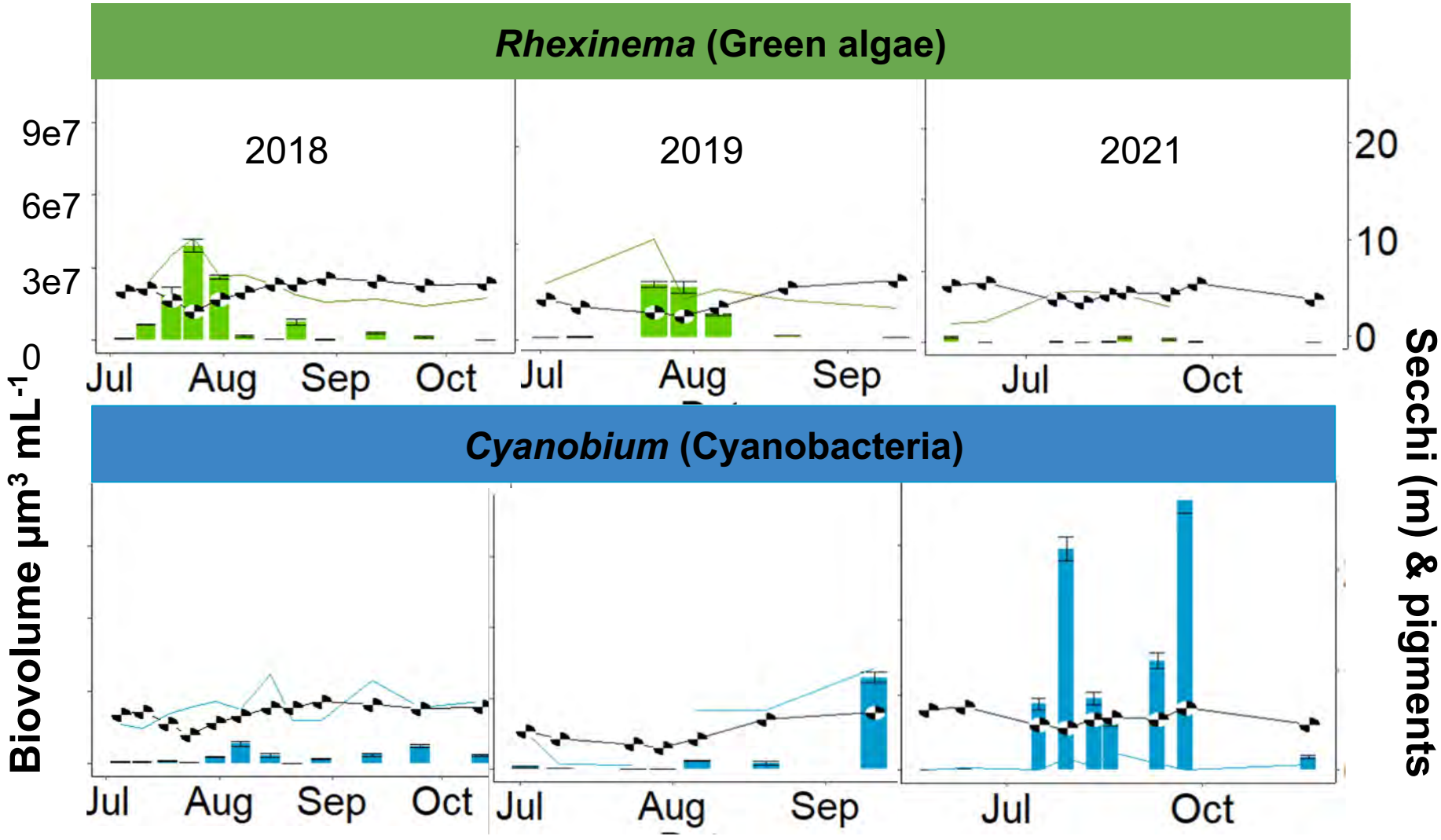
karen.wilson@maine.edu

rsleith@bigelow.org


Rhexinema (Green algae)
2018
2019
2021
9e7
6e7
3e7
0
Jul
Aug
Sep
Oct
20
10
0
Cyanobium (Cyanobacteria)
Biovolume µm³ mL⁻¹
Secchi (m) & pigments