



BIOS3300/4300 - MARINE BIOLOGY

Primary Production (Plankton) Long term observations

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Outline

- Introduction to long term observation
 - Why ?
 - Where ?
- Some examples
 - Continuous Plankton Recorder
 - Hawaii
 - Bermuda
- Periodicity in plankton species occurrence
 - English Channel - Roscoff
 - Mediterranean Sea - Blanes
- Plankton species dynamics
 - *Synechococcus* in Woods Hole

Reference material

- Bunse, C. & Pinhassi, J. 2017. Marine Bacterioplankton Seasonal Succession Dynamics. *Trends in Microbiology*. 25:494–505.
- Karl, D.M. & Church, M.J. 2014. Microbial oceanography and the Hawaii Ocean Time-series programme. *Nature Reviews Microbiology*. 12:699–713.
- Hunter-Cevera et al. 2016. Physiological and ecological drivers of early spring blooms of a coastal phytoplankter. *Science* 354:326–329.



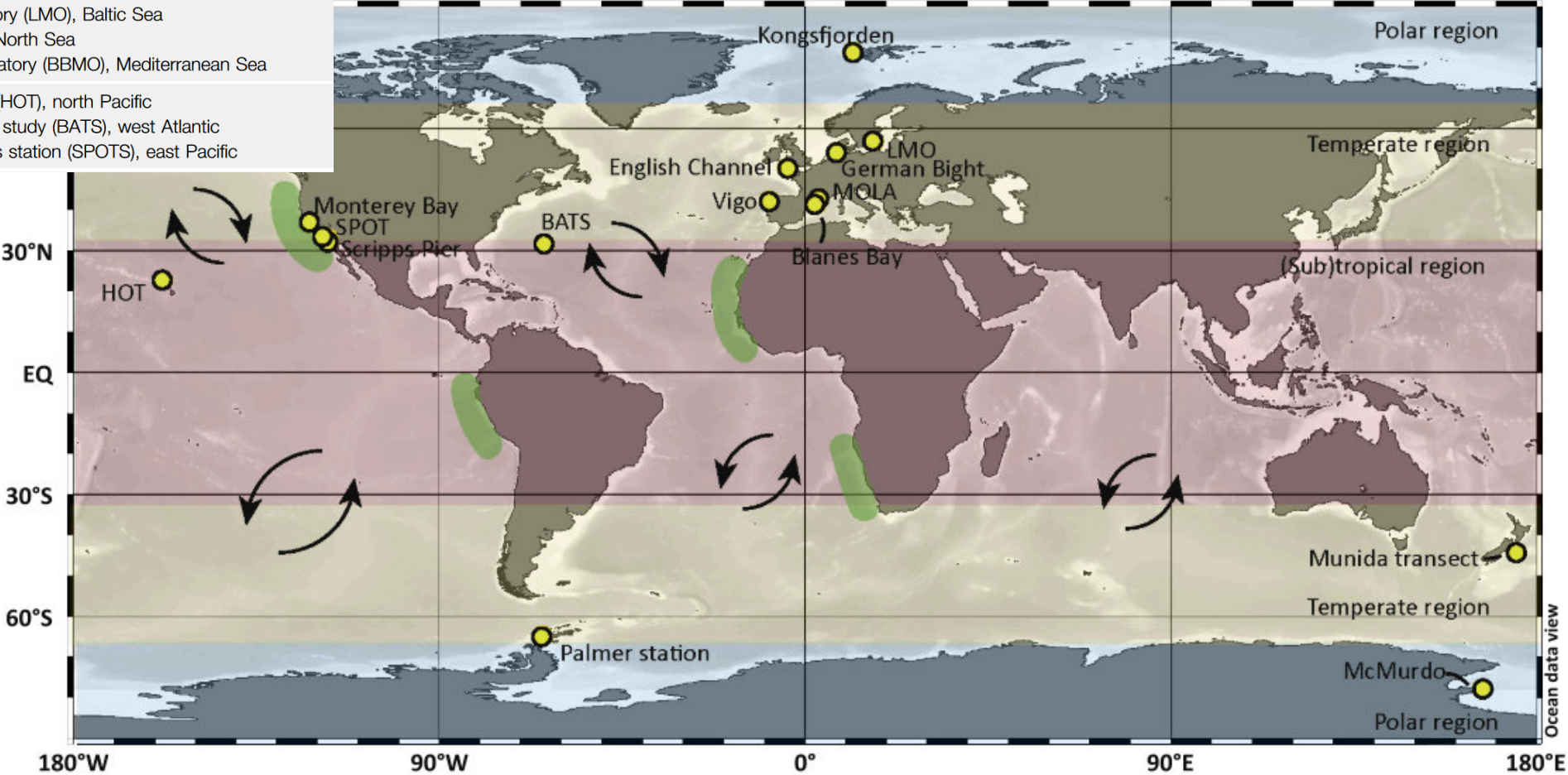
Introduction to time series

Questions to be solved by time series

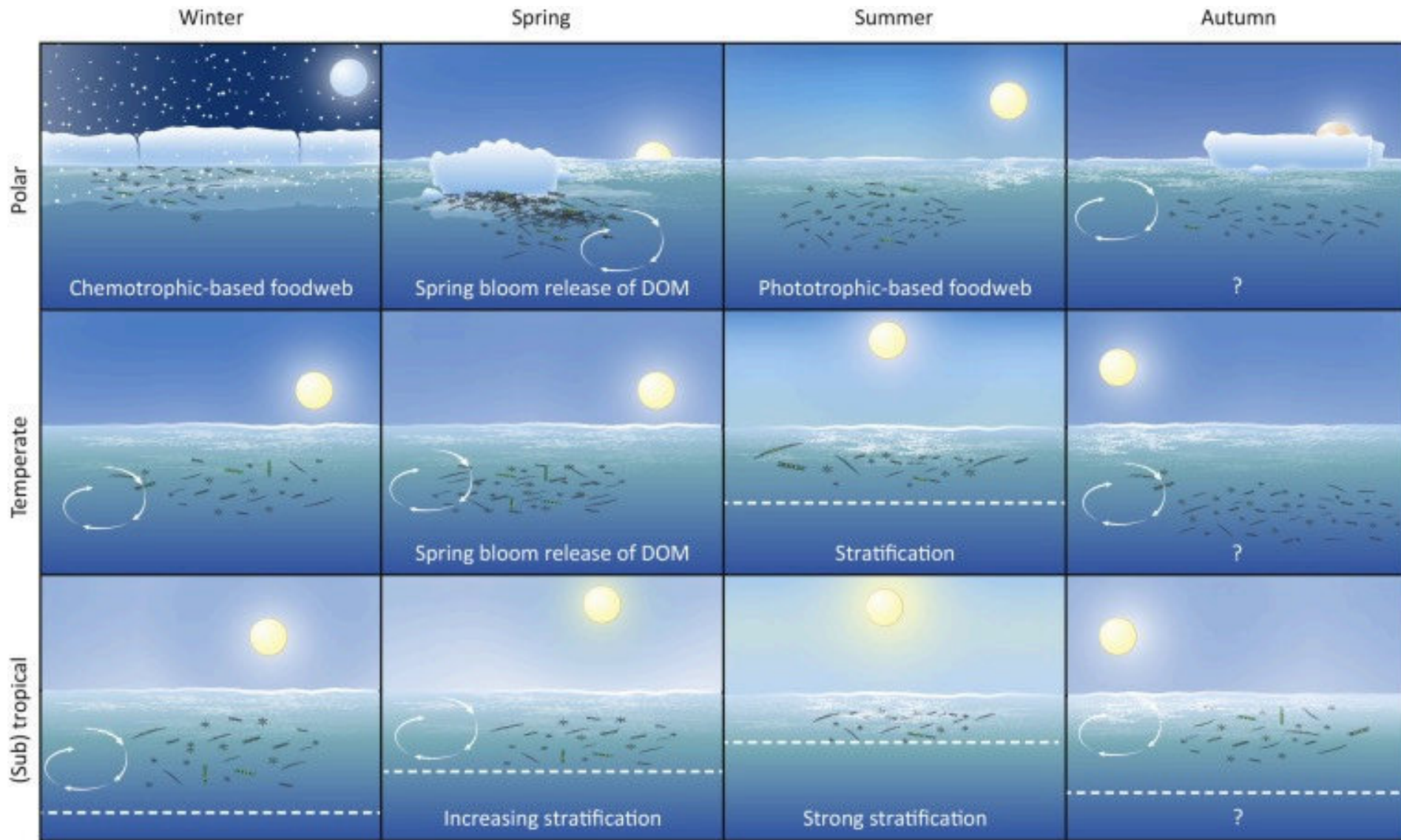
- Monitoring the environment
 - changes in biomass & production
 - changes in species composition
- What are the key periodicities ?
 - annual (what about equator ?)
 - tides (monthly)
 - daily (light-dark cycle)
- Are species recurring from one year to the next ?
- What drives the year to year variability
- Long term climatic trends

Long term pelagic time series

Region	Location
Polar	Antarctic Peninsula Franklin Bay, Western Arctic
Temperate	RADIALES time-series project (E2), east Atlantic Western Channel Observatory (L4), English Channel Linnaeus Microbial Observatory (LMO), Baltic Sea German Bight (Kabeltonne), North Sea Blanes Bay Microbial Observatory (BBMO), Mediterranean Sea
(Sub)- Tropical	Hawaiian Ocean time-series (HOT), north Pacific Bermuda Atlantic time-series study (BATS), west Atlantic San Pedro Ocean time-series station (SPOTS), east Pacific



Long term pelagic time series



Some examples

Continuous Plankton Recorder (CPR)

North Pacific
CPR Survey

NOAA

SAHFOS

MedCPR

JAMSTEC

Brazil Southern-
Ocean CPR

BCLME

SCAR SO CPR
Survey

AusCPR

NIWA

Continuous Plankton Recorder (CPR)

- conceived in 1920's by Sir Alistair Hardy
- started in 1931
- North Atlantic
- 280 000 samples analyzed
- <https://www.cprsurvey.org>

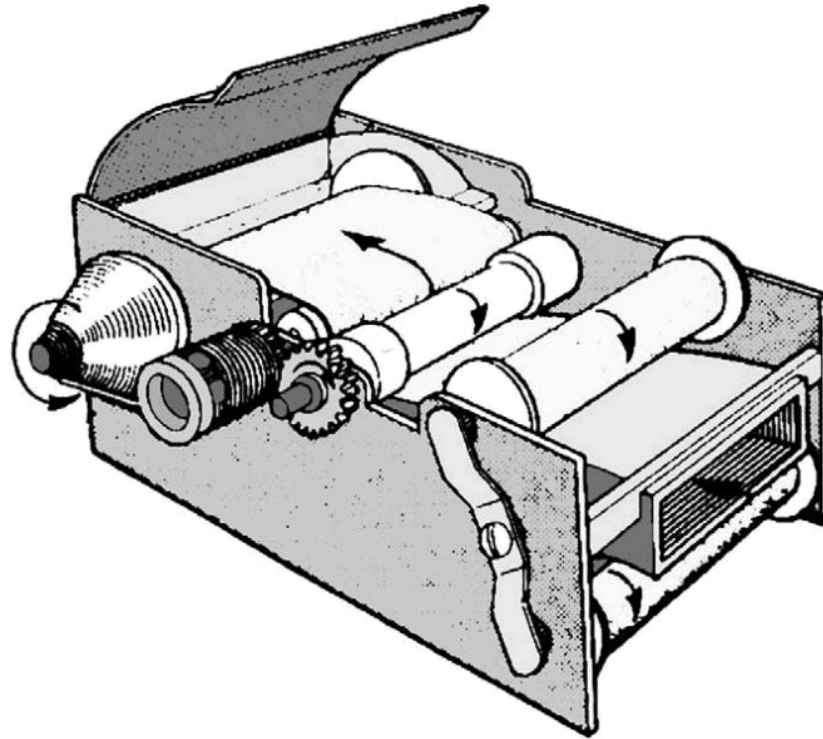


Fig. 5. Drawing of cassette plankton sampling mechanism (PSM) with arrows showing the movement of the silks from the filtering and covering reels to the storage tank. The lid of the tank is open. To the left, indicated by the circular arrow, the fusee mechanism with tightened wire, used to adjust the tension on the rolled up silks.

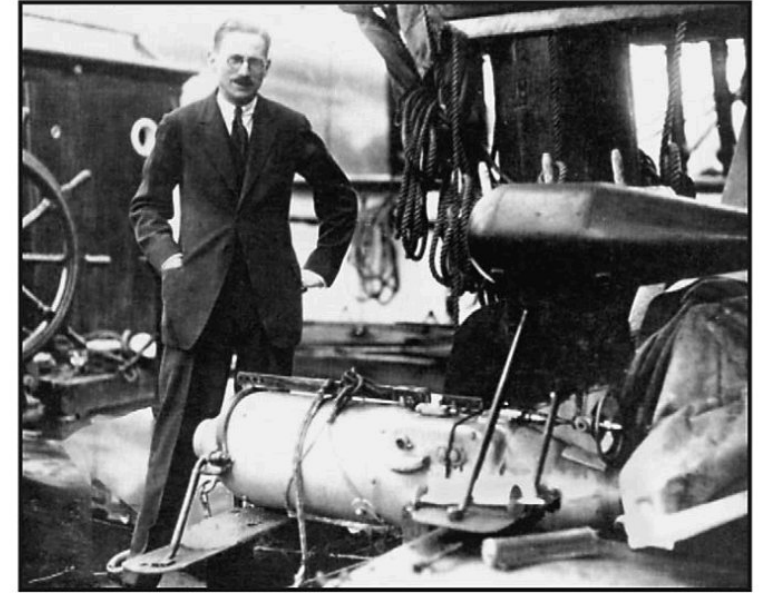


Fig. 2. Alistair Hardy and CPR Type I on RRS *Discovery*.

Instrumentation

- Plankton collection
- Temperature
- Salinity
- Fluorescence



The Continuous Plankton Recorder

A platform for integrated ocean observing

In addition to the traditional biological sampling undertaken by the CPR the towed body can be equipped with a range of sensing capabilities to extend its utility for integrated observing

Seawater enters via the aperture.
Plankton is captured on a filter silk band then covered by a further silk band. The continuously moving band is wound through the CPR on rollers turned by gears, which are powered by a propeller allowing for long distances to be towed

Vemco Minilog :
Temperature sensor

Star Oddi CTD :
Conductivity, Temperature and Pressure (Depth)

SAHFOS CPR Internal :
Phytoplankton, Zooplankton, Planktonic Bacteria and Viruses



Planktag : Conductivity, Temperature, Chlorophyll-a, Fluorescence and ambient Light.
Data telemetry enables observations to be streamed back to within minutes of the CPR surfacing

WAMS : Water and Molecular Sampler

UFE Multispectral Fluorometers : Rapid optical detection of phytoplankton forms, Pressure (Depth) and Temperature

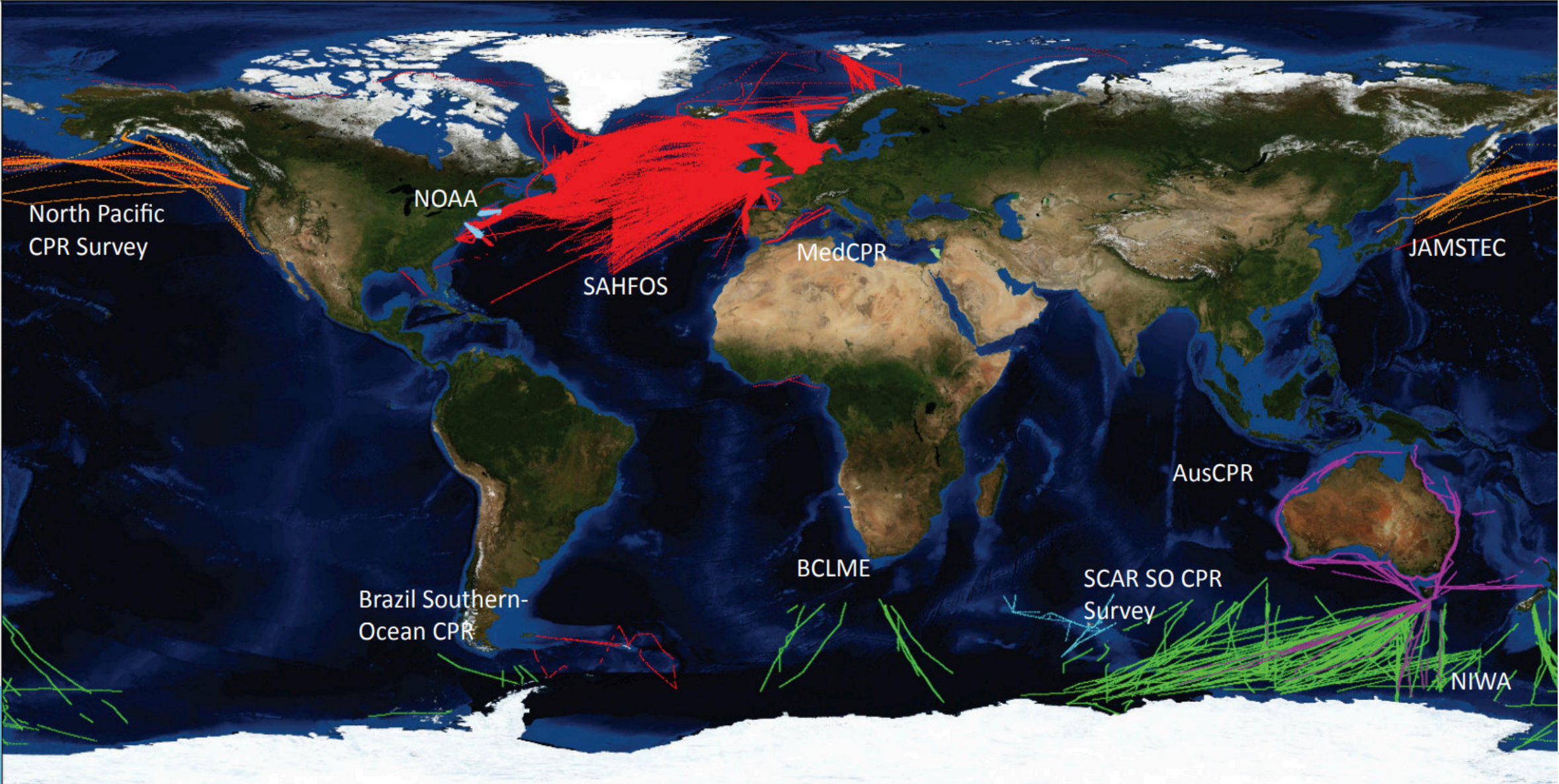
RBR CTD : Conductivity, Temperature, Pressure (Depth) and Fluorescence



Ship lines



Ship lines



Annual variability in the 30's

- *Phaeocystis*

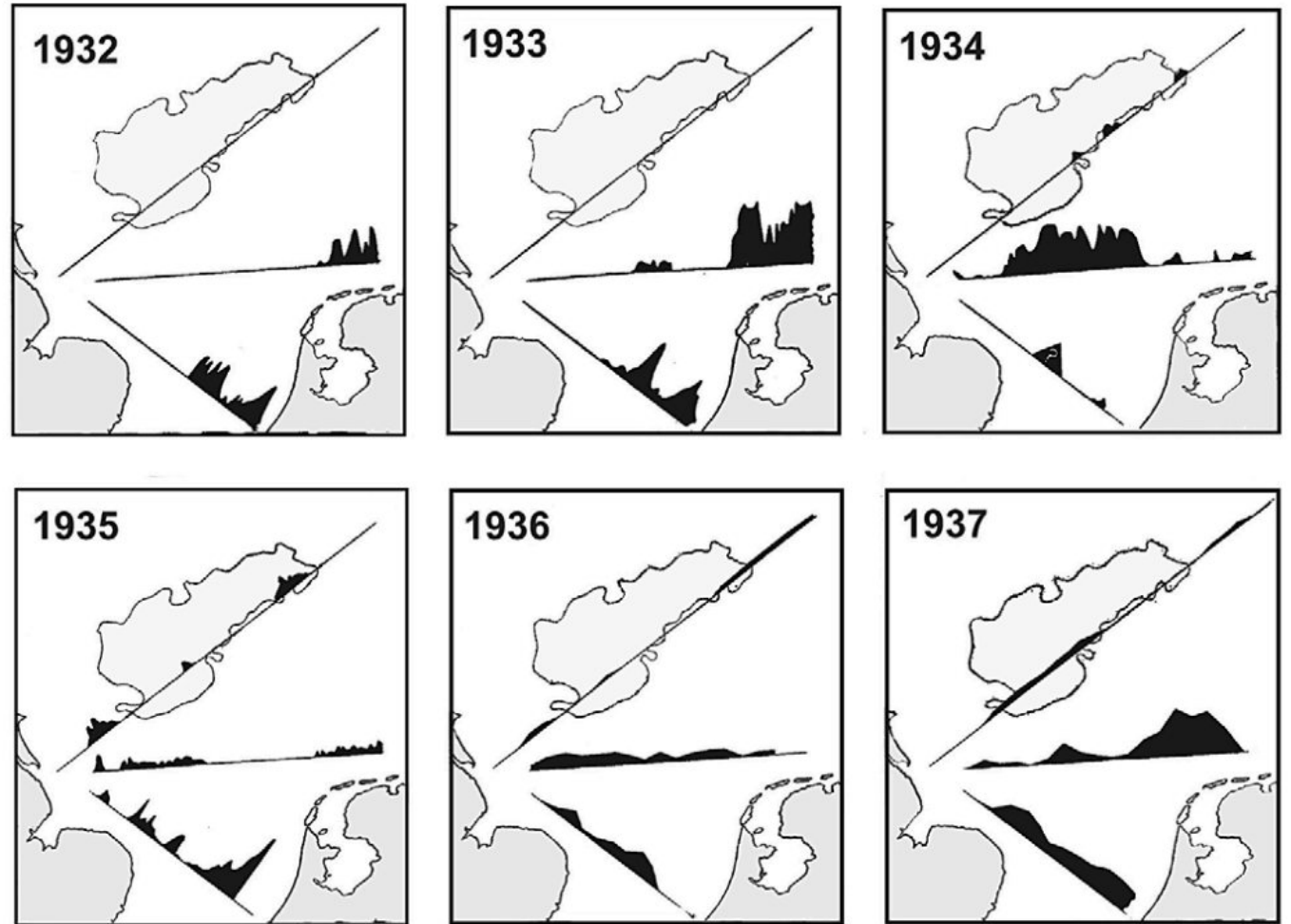
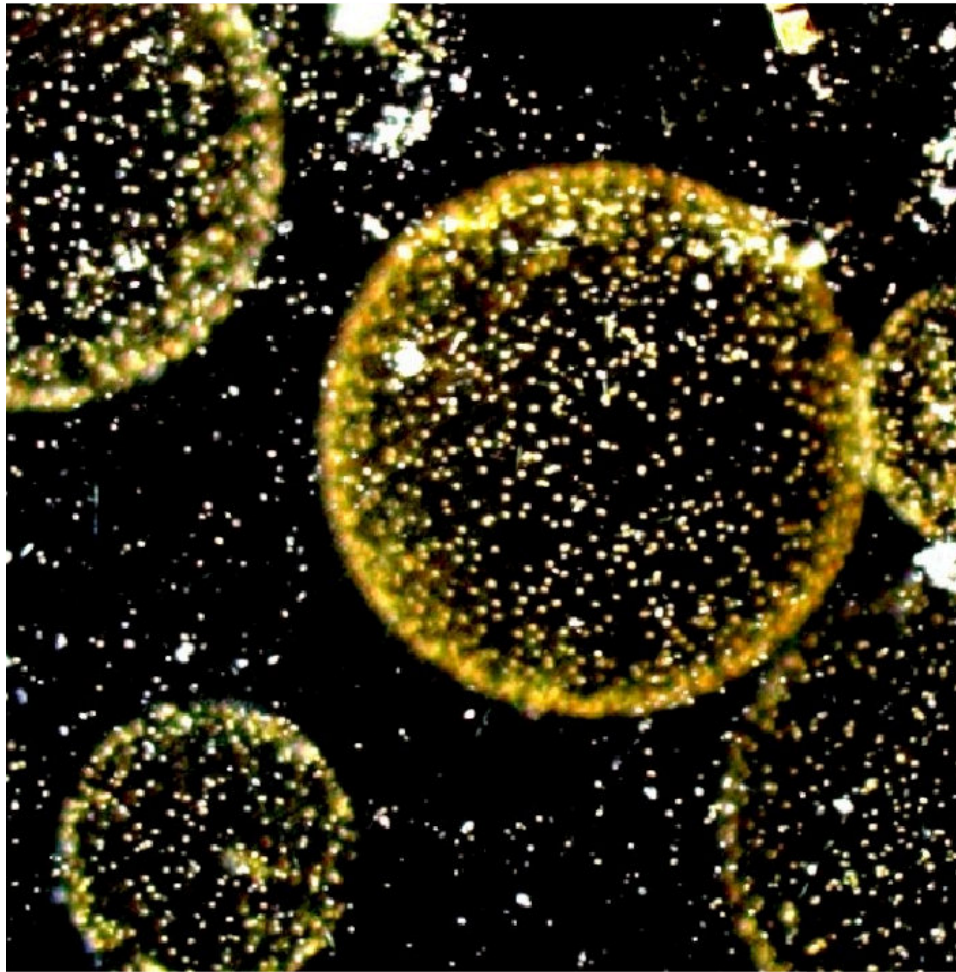
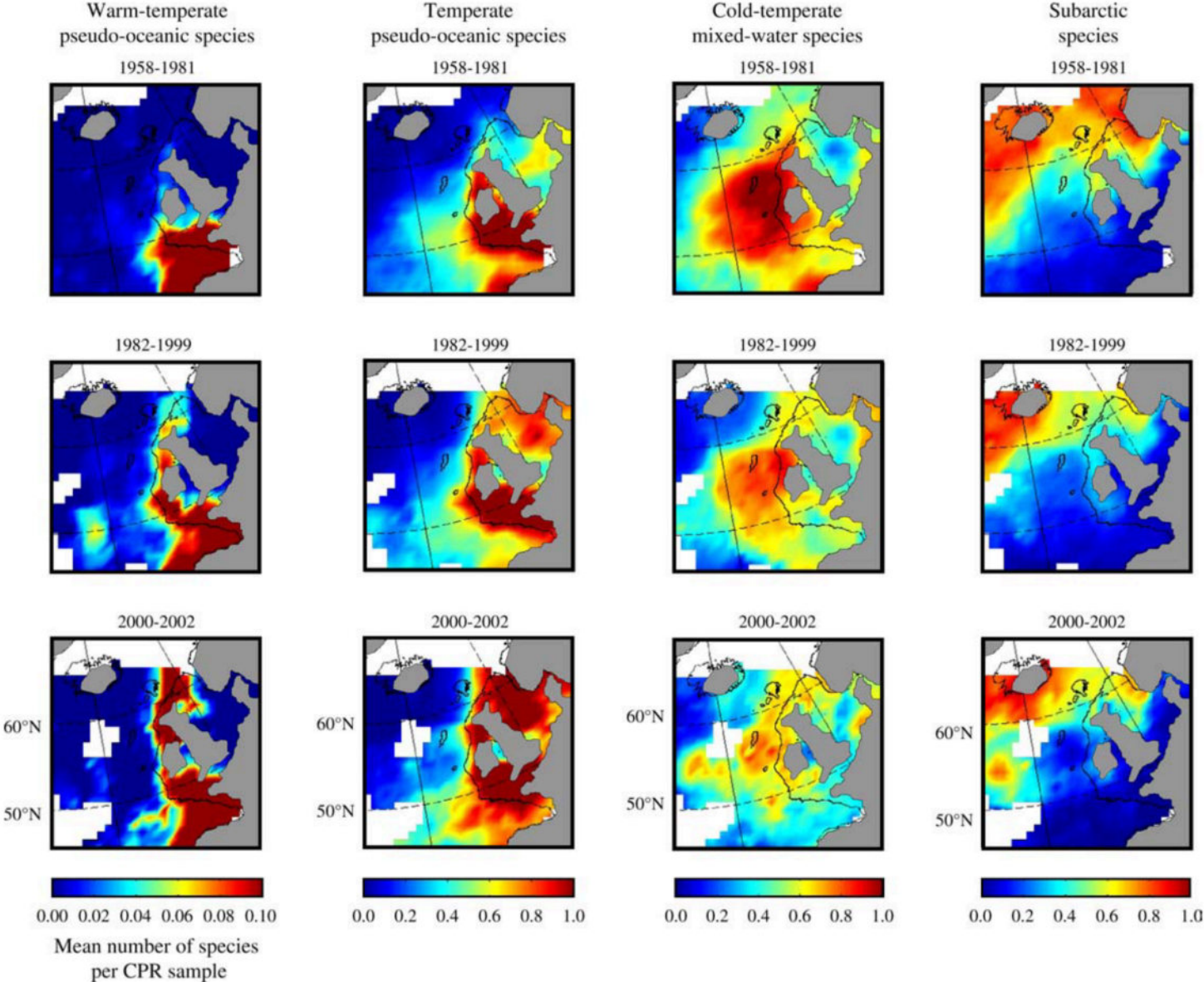


Fig. 7. Abundance of *Phaeocystis* along transects across the southern North Sea between 1932 and 1937 (Redrawn after Lucas, 1940). Dogger Bank outlined in light grey

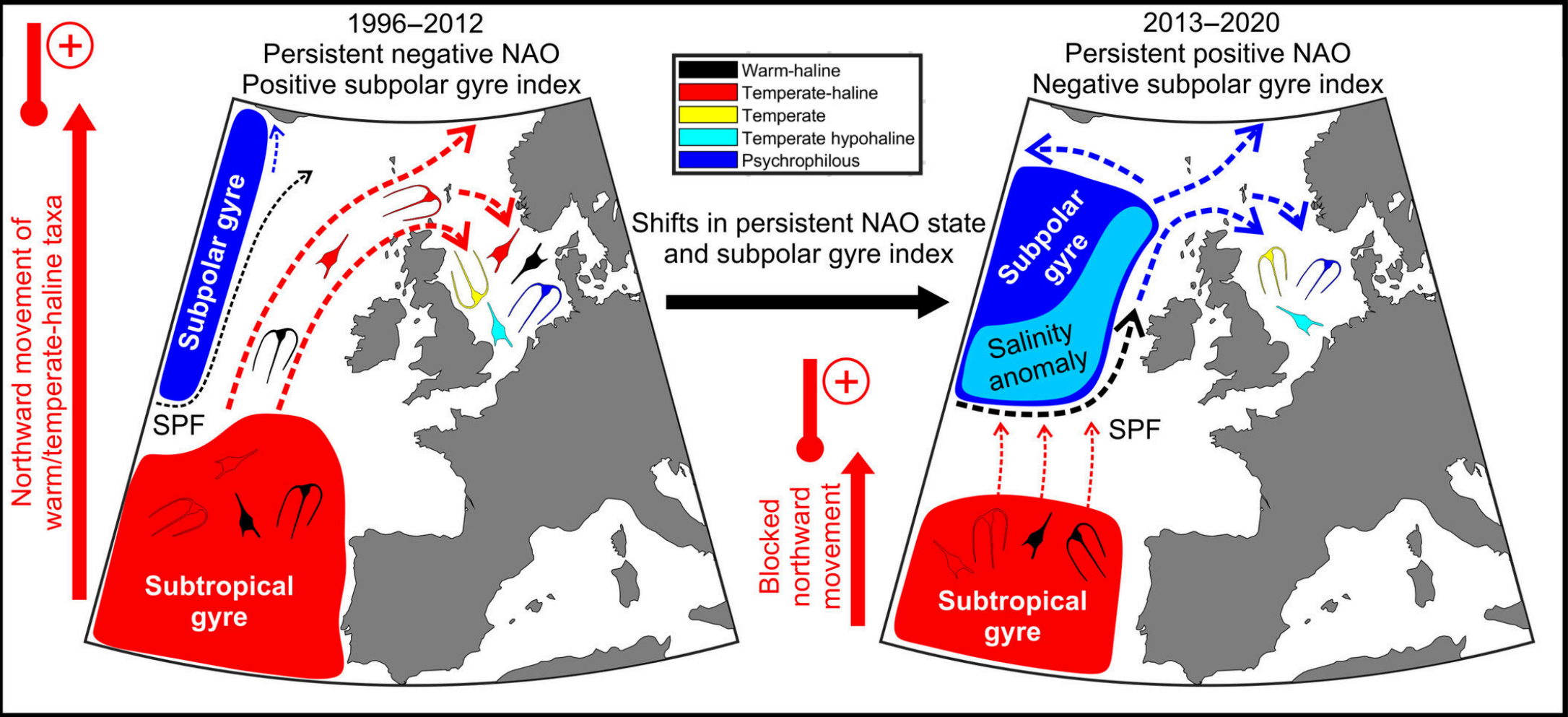
Global change - Species

- Atlantification



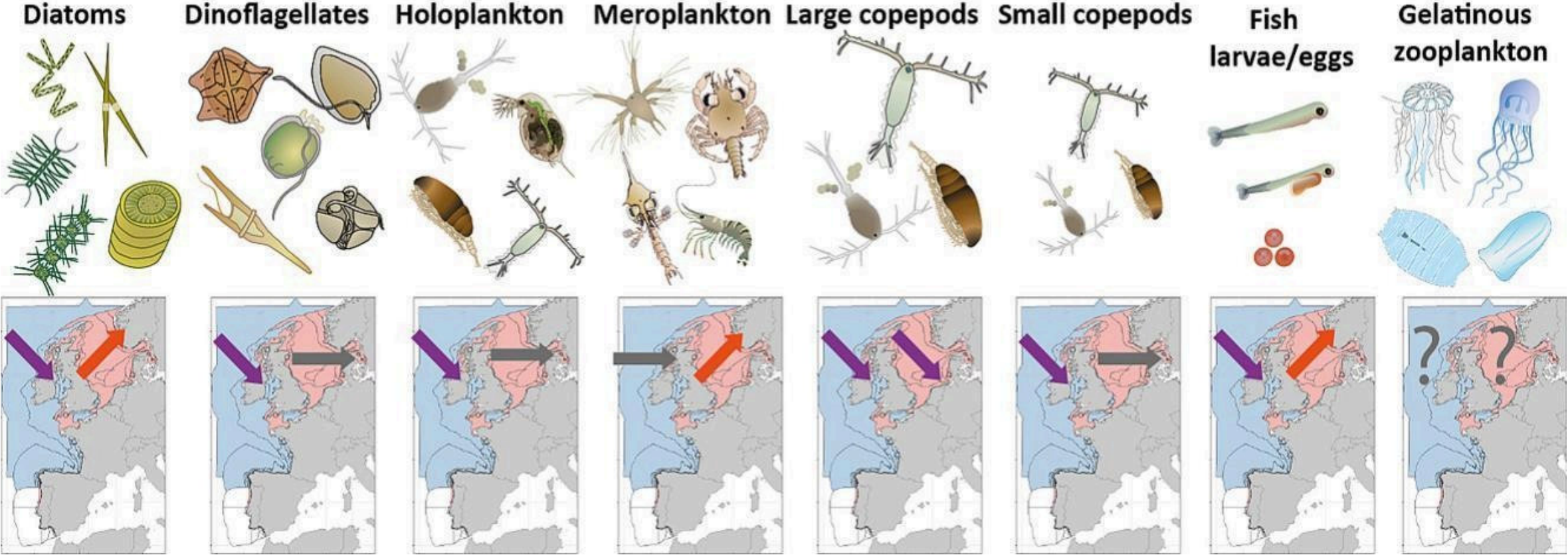
Global change - Species

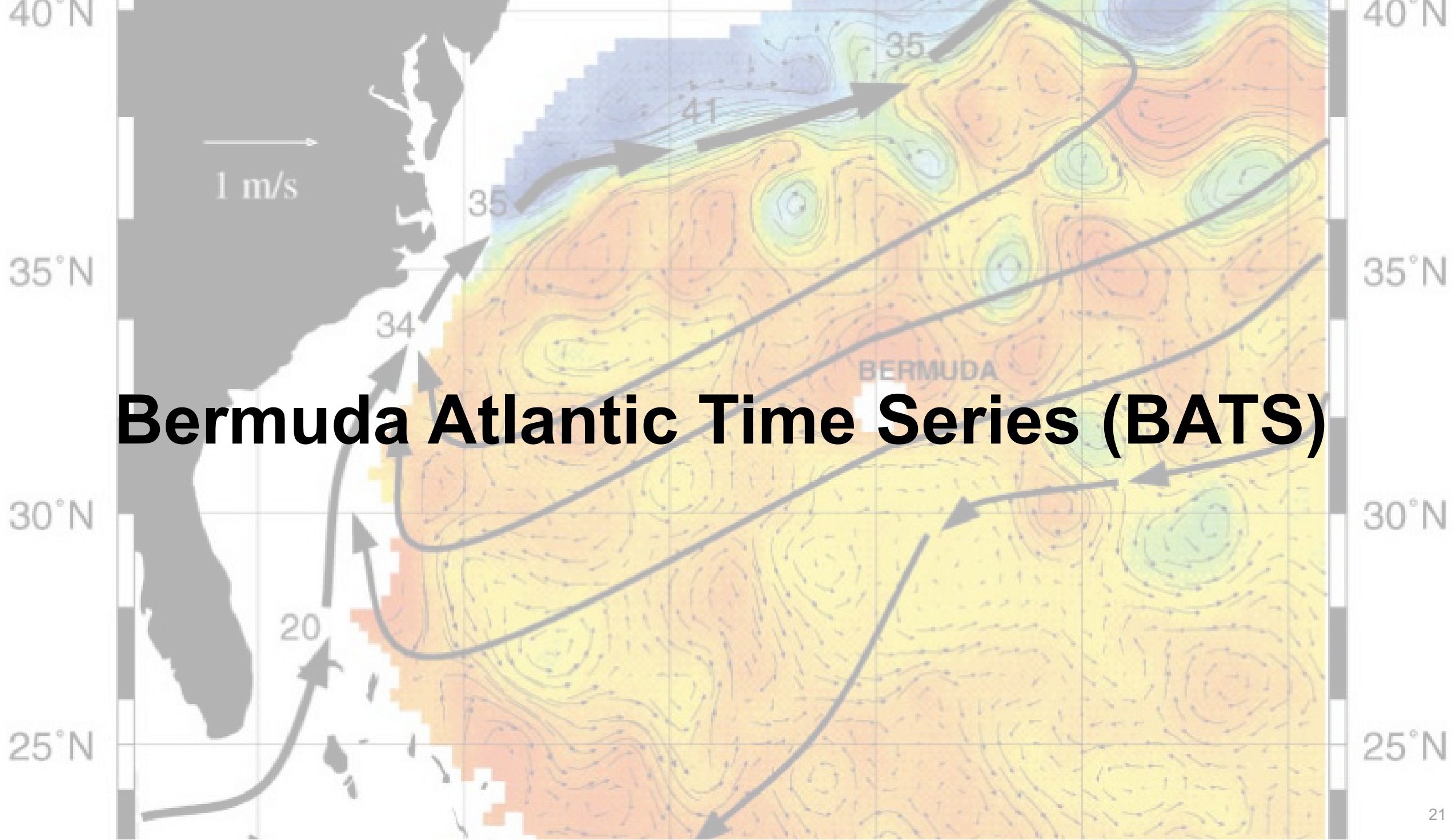
- Dinoflagellates



Global change - Biomass

Long-term abundance trends (1960-2019)

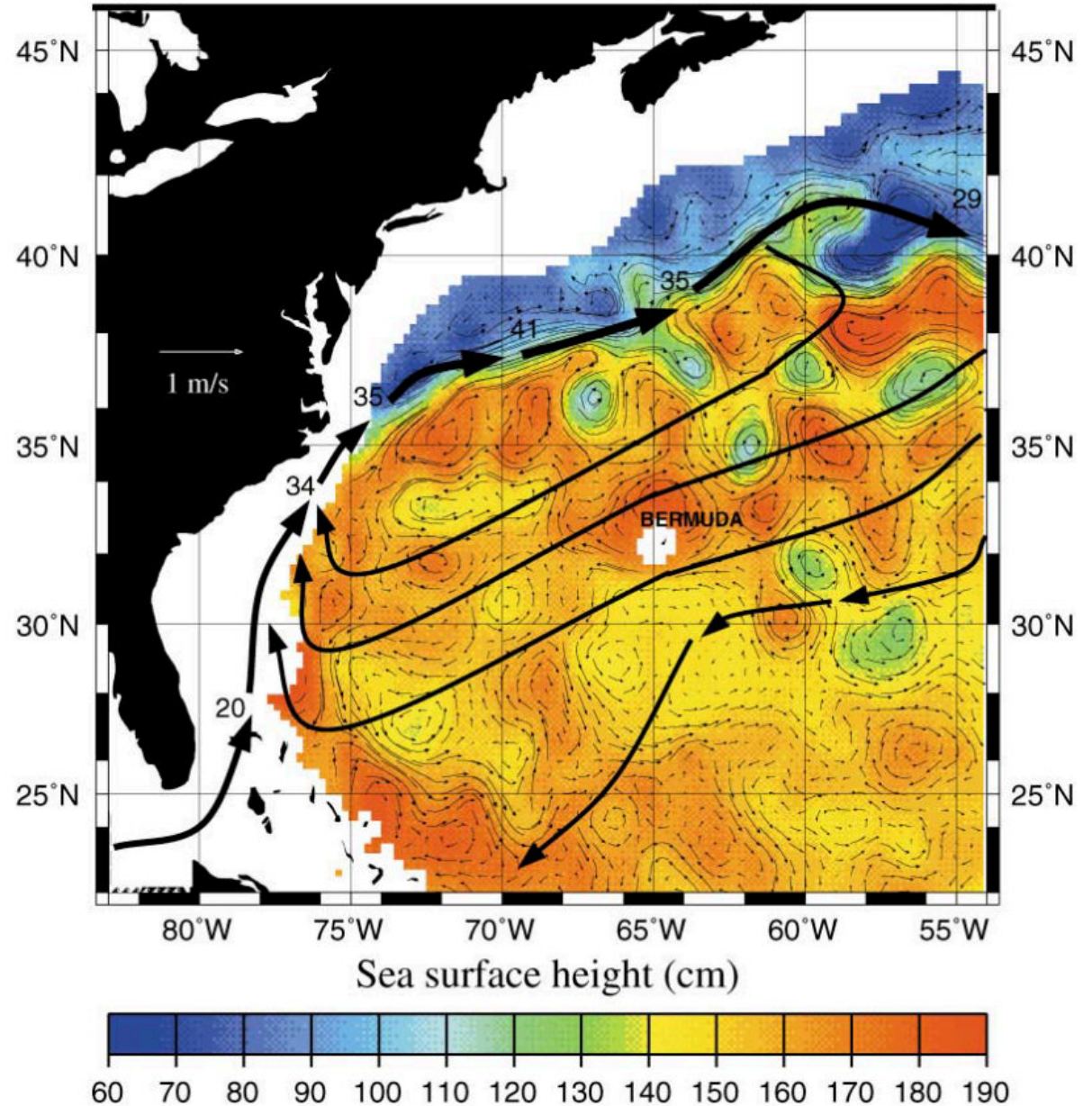




Bermuda Atlantic Time Series (BATS)

Bermuda Atlantic Time Series (BATS)

- located in Sargasso Sea
- started in 1989



Measurements

Parameter	Depth range (m)	Technique/instrument
<i>Continuous electronic measurements</i>		
Temperature	0–4200	Thermistor on SeaBird SBE-911plus CTD
Salinity	0–4200	Conductivity sensor on SeaBird SBE-911plus CTD
Depth	0–4200	Digiquartz pressure sensor on SeaBird SBE-911plus CTD
Dissolved oxygen	0–4200	SeaBird Polarographic Oxygen Electrode
Beam attenuation ^a	0–200	SeaTech, 25 cm Transmissometer
Fluorescence	0–500	Chelsea Instruments Fluorometer
PAR ^a	0–200	Biospherical Scalar Irradiance Sensor, 400–700 nm
<i>Discrete measurements from Niskin bottles on CTD</i>		
Salinity	0–4200	Conductivity on Guildline Autosal 8400A
Oxygen	0–4200	Winkler Titration, automated UV endpoint detection
Total CO ₂	0–500	Automated coulometric analysis
Alkalinity	0–500	High precision titration
Nitrate	0–4200	CFA colorometric with Technicon AA
Nitrite	0–4200	CFA colorometric with Technicon AA
Phosphate	0–4200	CFA colorometric with Technicon AA
Silicate	0–4200	CFA colorometric with Technicon AA
Dissolved organic carbon	0–4200	High-temperature combustion
Dissolved organic nitrogen	0–4200	UV oxidation
Particulate organic carbon	0–4200	High-temperature combustion, CHN analyzer
Particulate organic nitrogen	0–4200	High-temperature combustion, CHN analyzer
Particulate silica	0–4200	Chemical digestion, colorometric analysis
Fluorometric chlorophyll <i>a</i>	0–250	Acetone extraction, Turner fluorometer
Phytoplankton pigments	0–250	HPLC, resolves 19 pigments
Bacteria	0–3000	DAPI stained, fluorescence microscopy
<i>Rate measurements</i>		
Primary production	0–140	Trace-metal clean, in situ incubation, ¹⁴ C uptake
Bacterial activity	0–1000	[³ H-methyl] thymidine incorporation
Particle fluxes	150, 200, 300	Free-drifting cylindrical trap (MultiPITs)
Mass flux		Gravimetric analysis
Total carbon flux		Manual swimmer removal, CHN analysis
Organic carbon flux		Manual swimmer removal, acidification, CHN analysis
Organic nitrogen flux		Manual swimmer removal, CHN analysis

Temperature

- Deep winter mixing

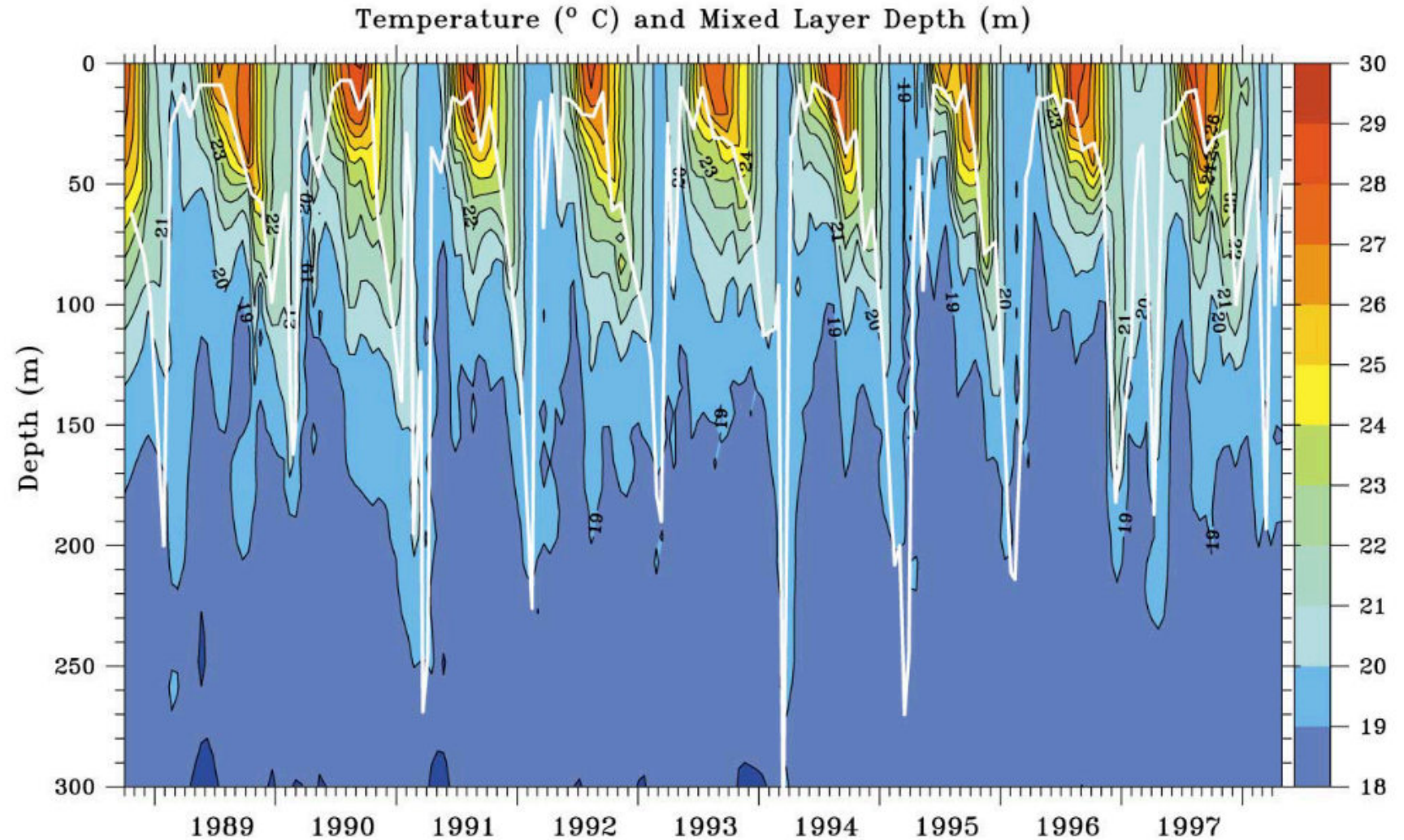


Fig. 3. Time-series contour plot of temperature with a 1° C contour interval. Mixed-layer depth is overlaid as a solid white line. Mixed-layer depth defined by using a variable sigma theta criteria (Sprintall and Tomczak, 1992) and assuming a 0.3° C diurnal temperature change.

Nutrients

- Effect of winter mixing

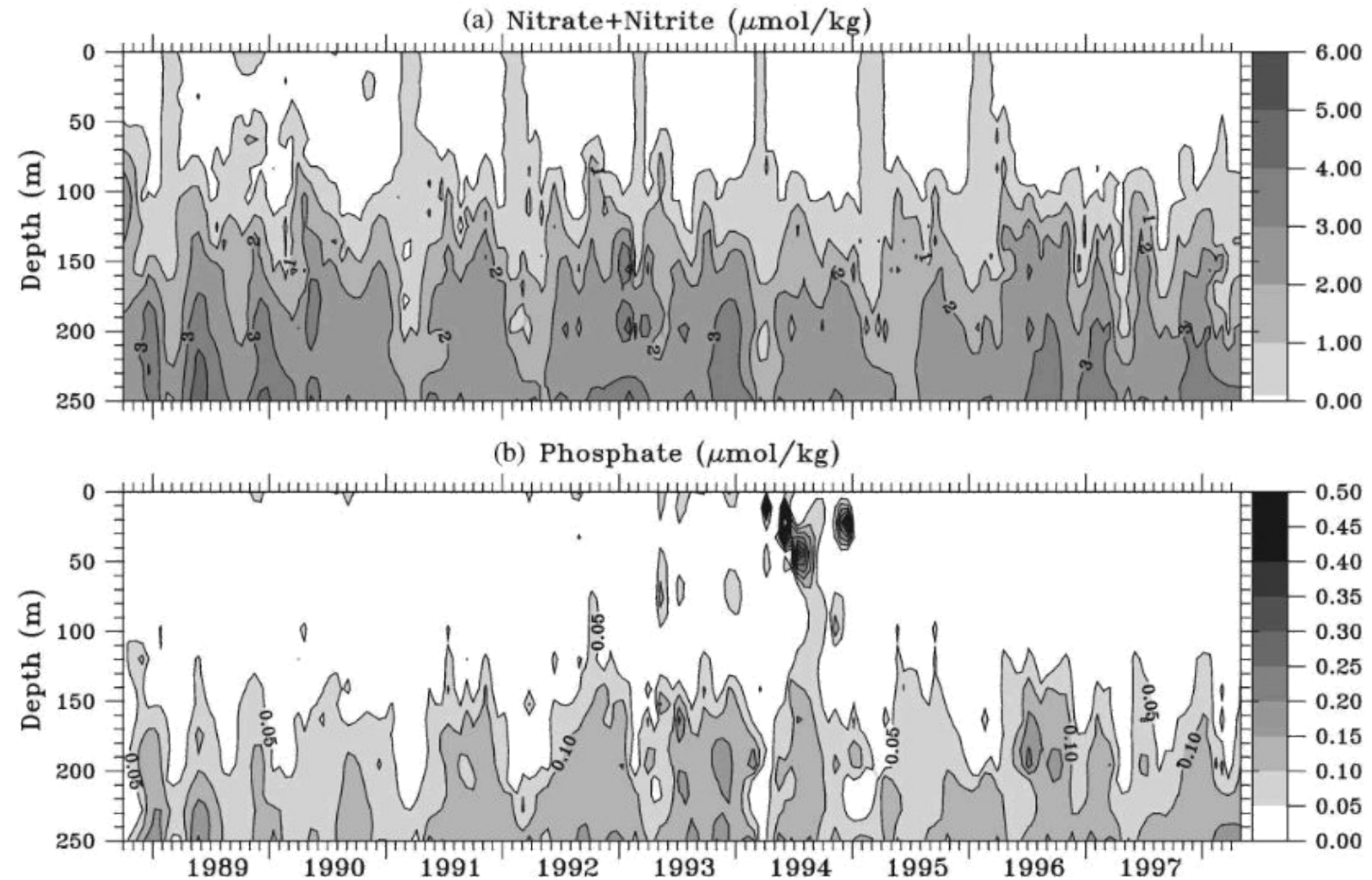


Fig. 6. Time-series contour plots of (a) nitrate + nitrite with a contour interval of $1 \mu\text{mol kg}^{-1}$ (with exception of first interval of $0.1 \mu\text{mol kg}^{-1}$), and (b) soluble reactive phosphate with a contour interval of $0.05 \mu\text{mol kg}^{-1}$.

Primary production

- Effect of winter mixing

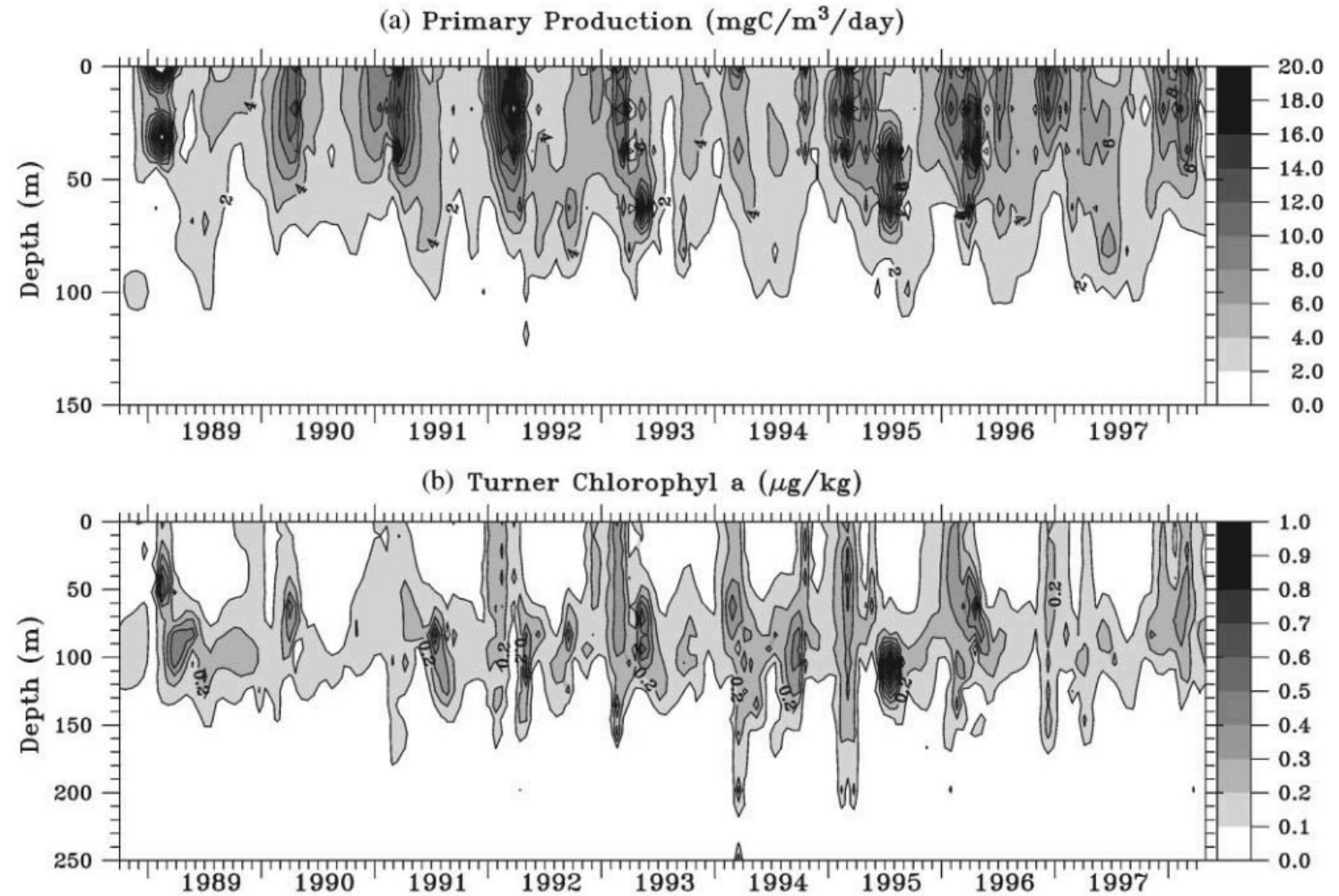
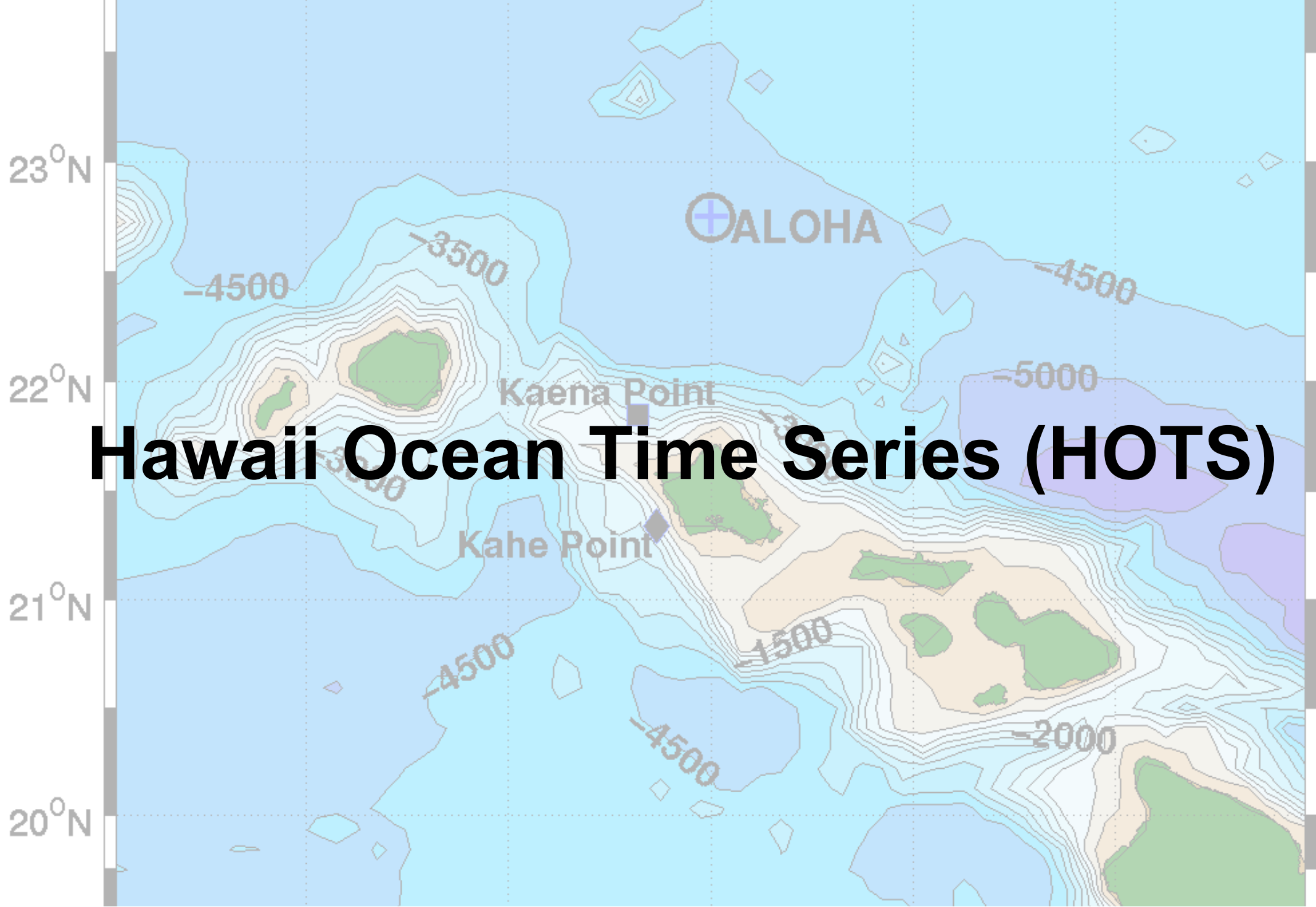


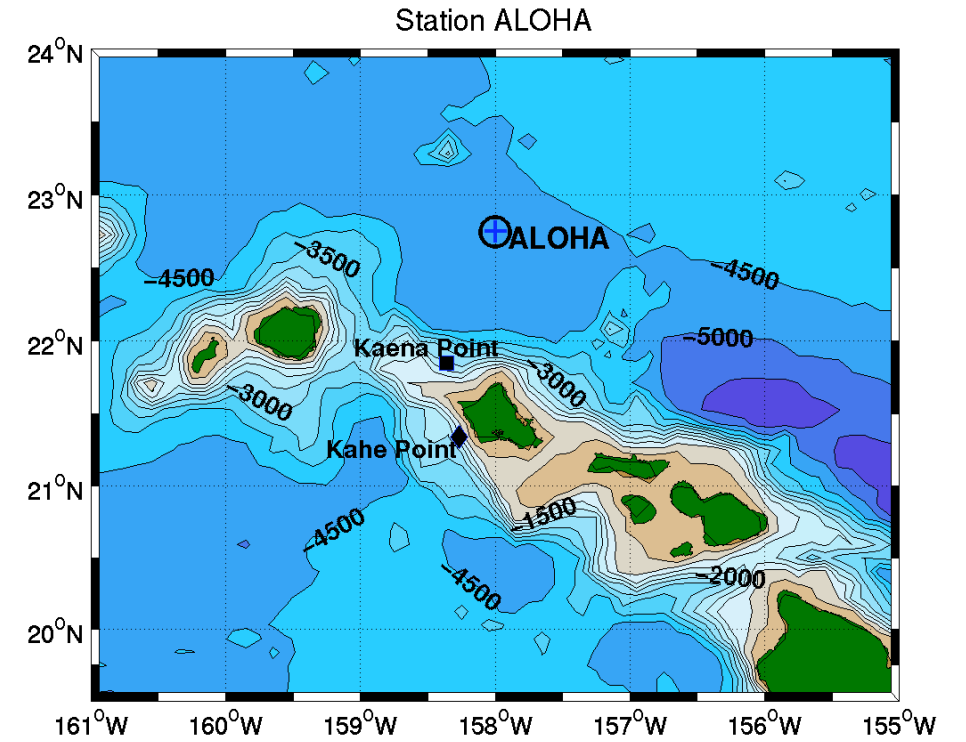
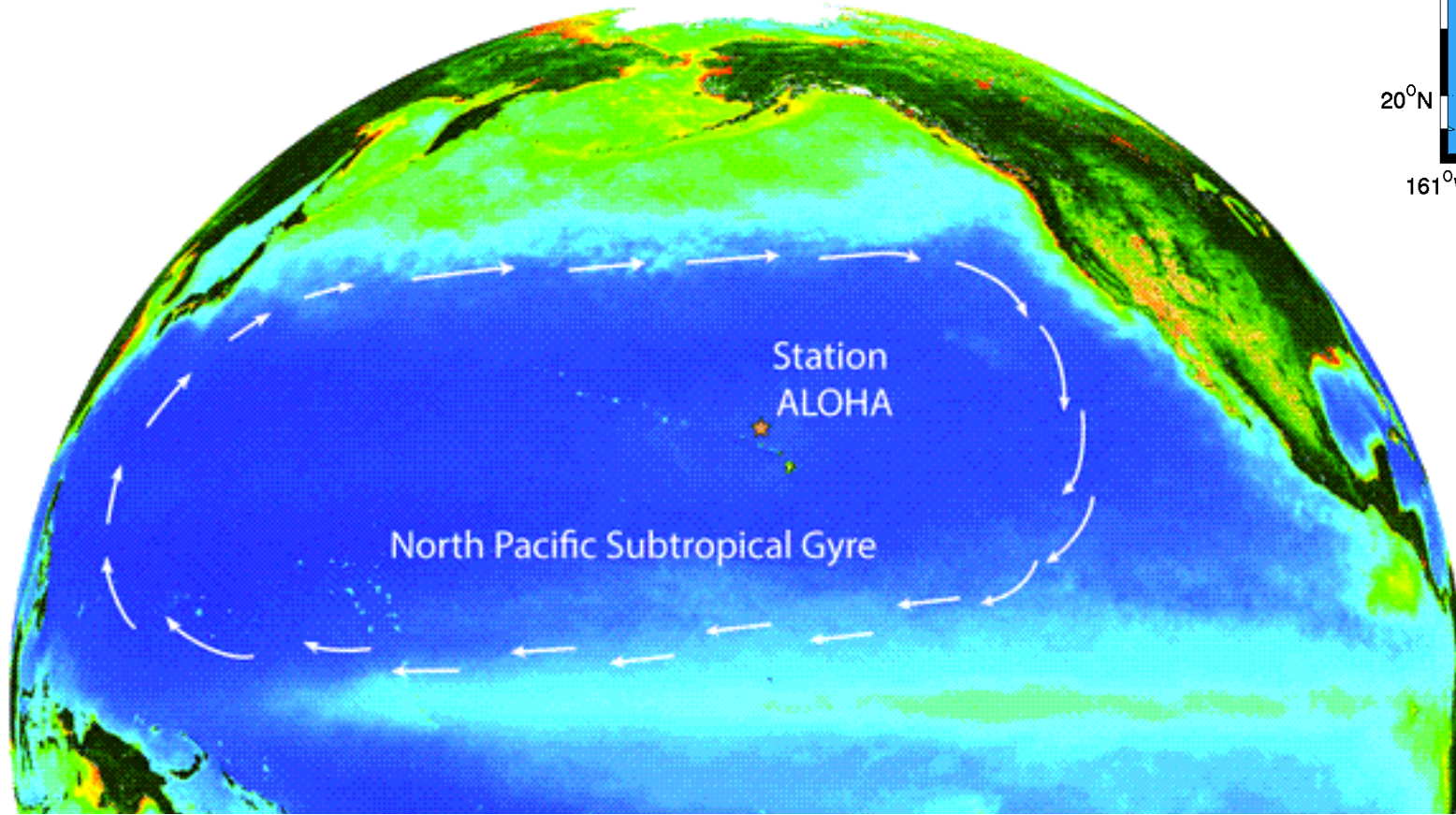
Fig. 7. Time-series contour plots of (a) primary production with a contour interval of $2 \text{ mgC m}^{-3} \text{ d}^{-1}$, and (b) chlorophyll *a* with a contour interval of $0.1 \mu\text{g kg}^{-1}$.



Hawaii Ocean Time Series (HOTS)

Hawaii Ocean Time Series (HOTS)

- Station ALOHA
- started in 1989
- located in oligotrophic area



General characteristics

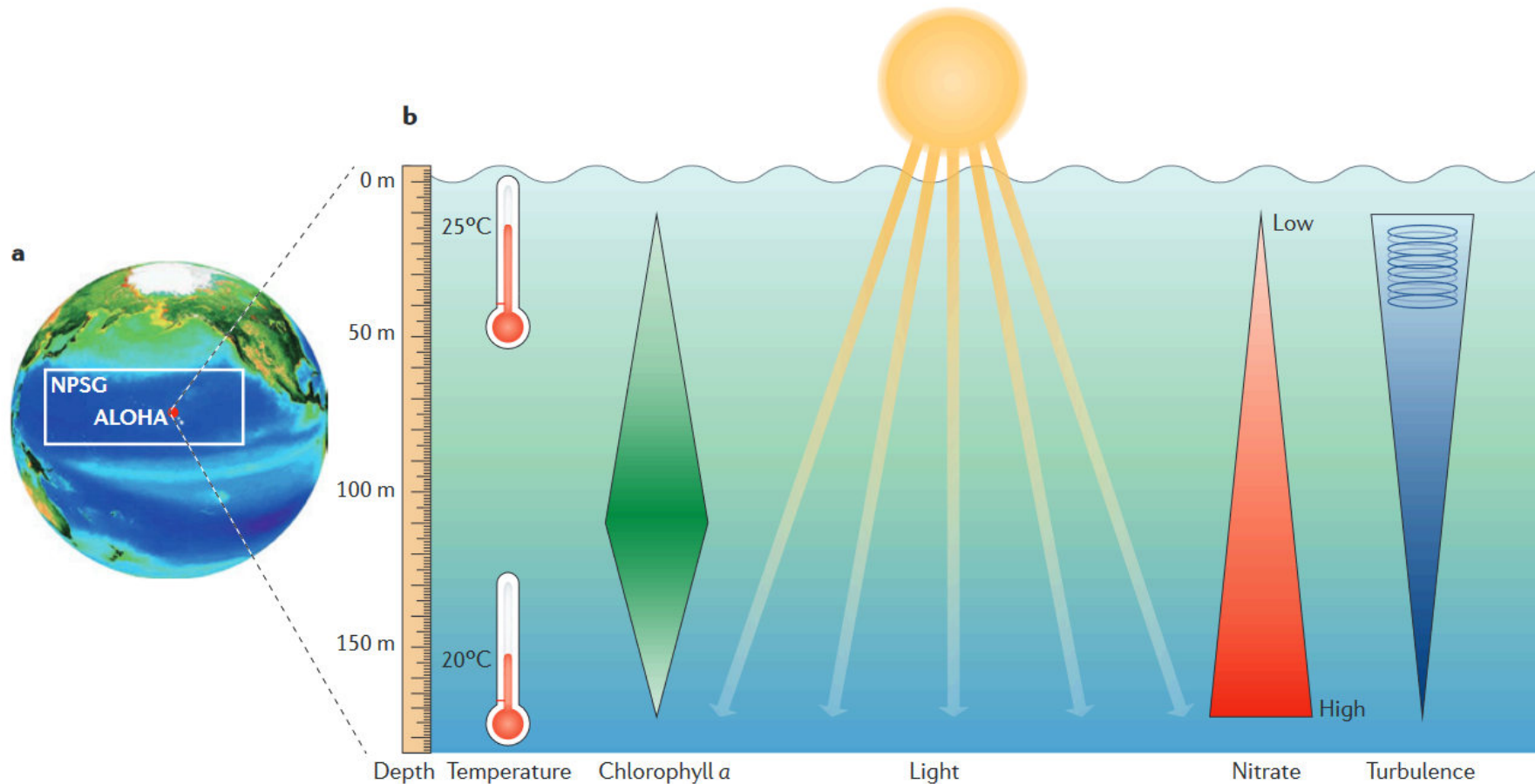
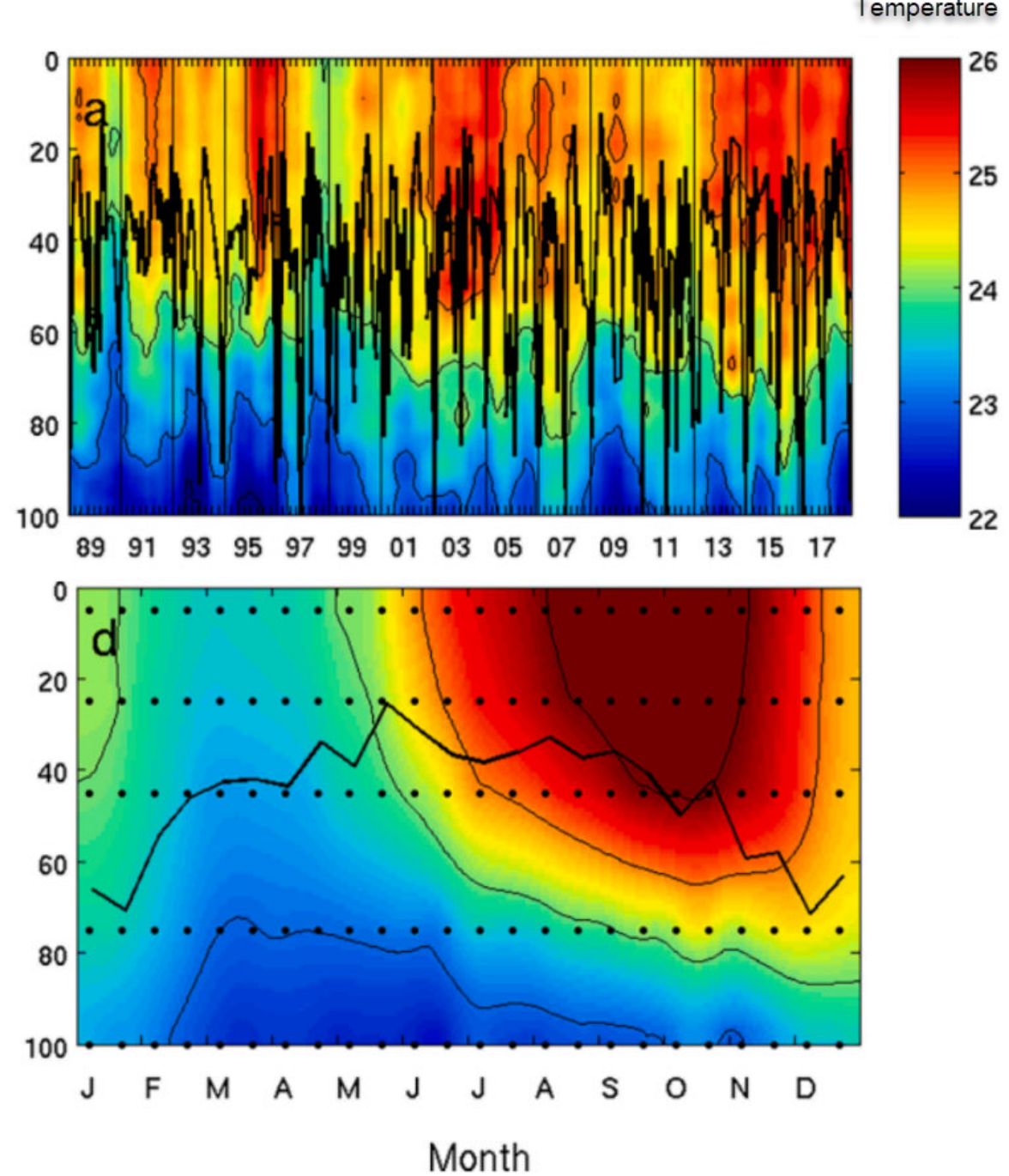


Figure 1 | **Station ALOHA habitat characteristics.** **a** | Location of Station ALOHA (A Long-term Oligotrophic Habitat Assessment) in the North Pacific Subtropical Gyre (NPSG) depicted on a *Sea-viewing Wide Field-of-view Sensor* (SeaWiFS) map of ocean colour (see Further information) showing the low concentrations of chlorophyll *a* (dark blue) that surround the site. **b** | The schematic shows the general habitat characteristics at Station ALOHA based on the 25 year climatology. This is an extremely oligotrophic environment that is characterized by low-standing stocks of chlorophyll (the subsurface chlorophyll peaks at ~105 m) and nitrate concentrations (note that primary production peaks where light is high but nutrients (such as nitrate) are nearly absent). Light that is sufficient for photosynthesis penetrates to at least 175 m. Temperature and the amount of turbulent mixing are also shown.

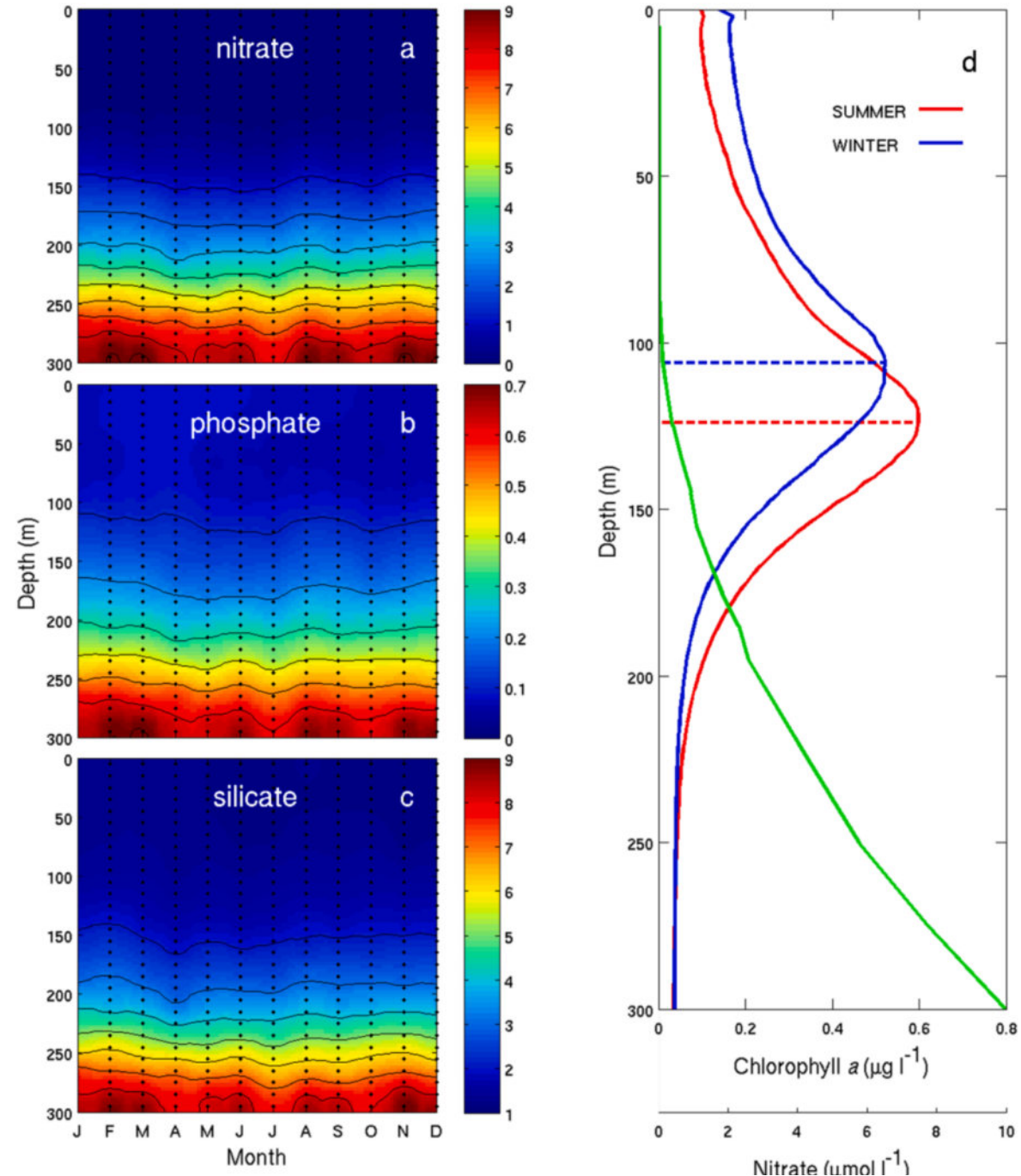
Temperature structure

- Although tropical, temperature change



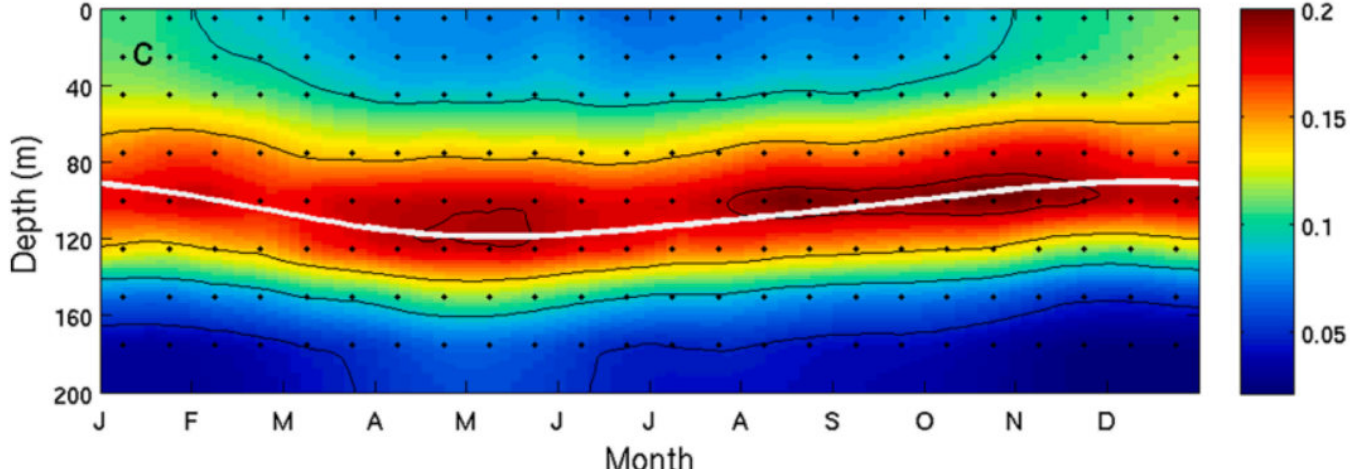
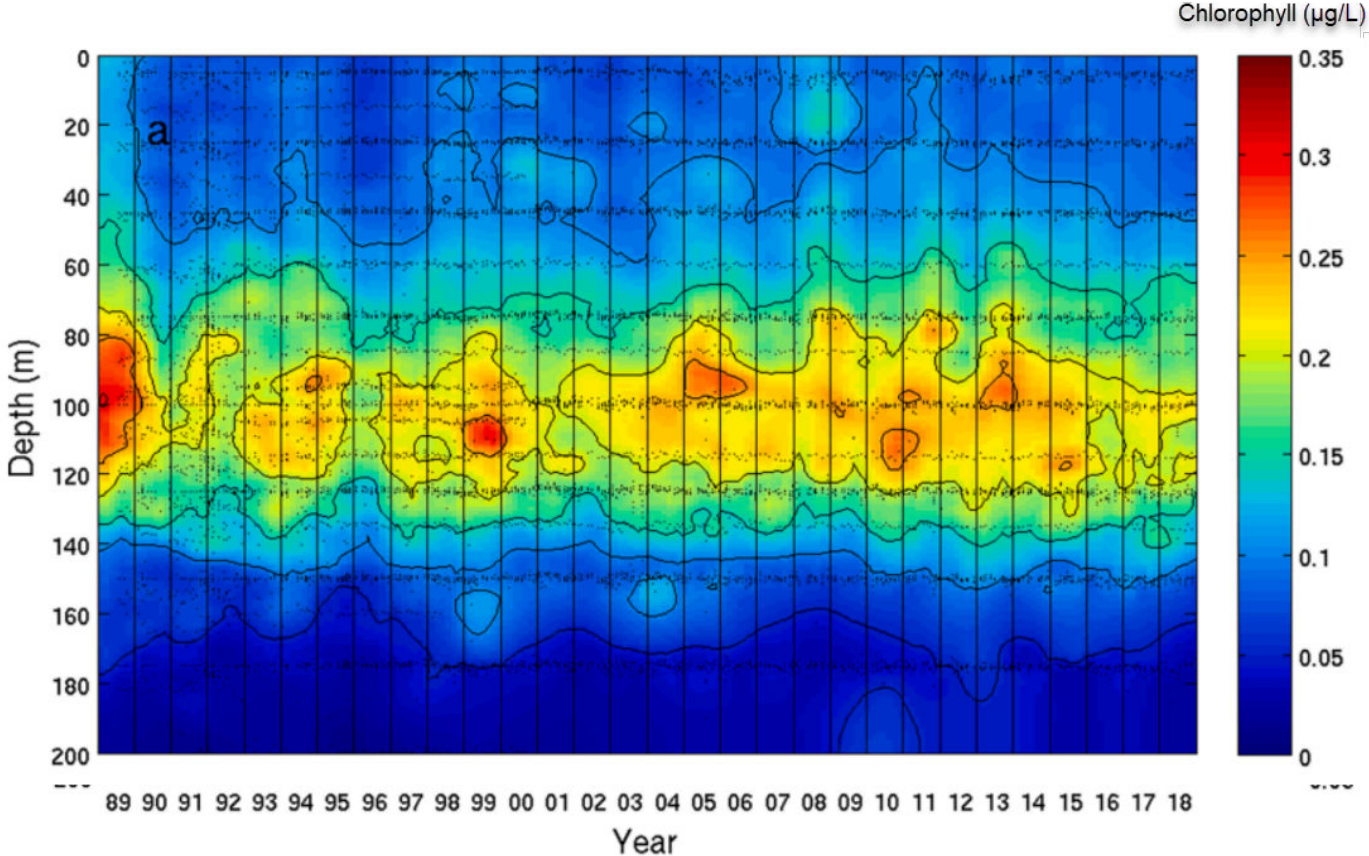
Vertical structure

- Deeper in summer



Chlorophyll

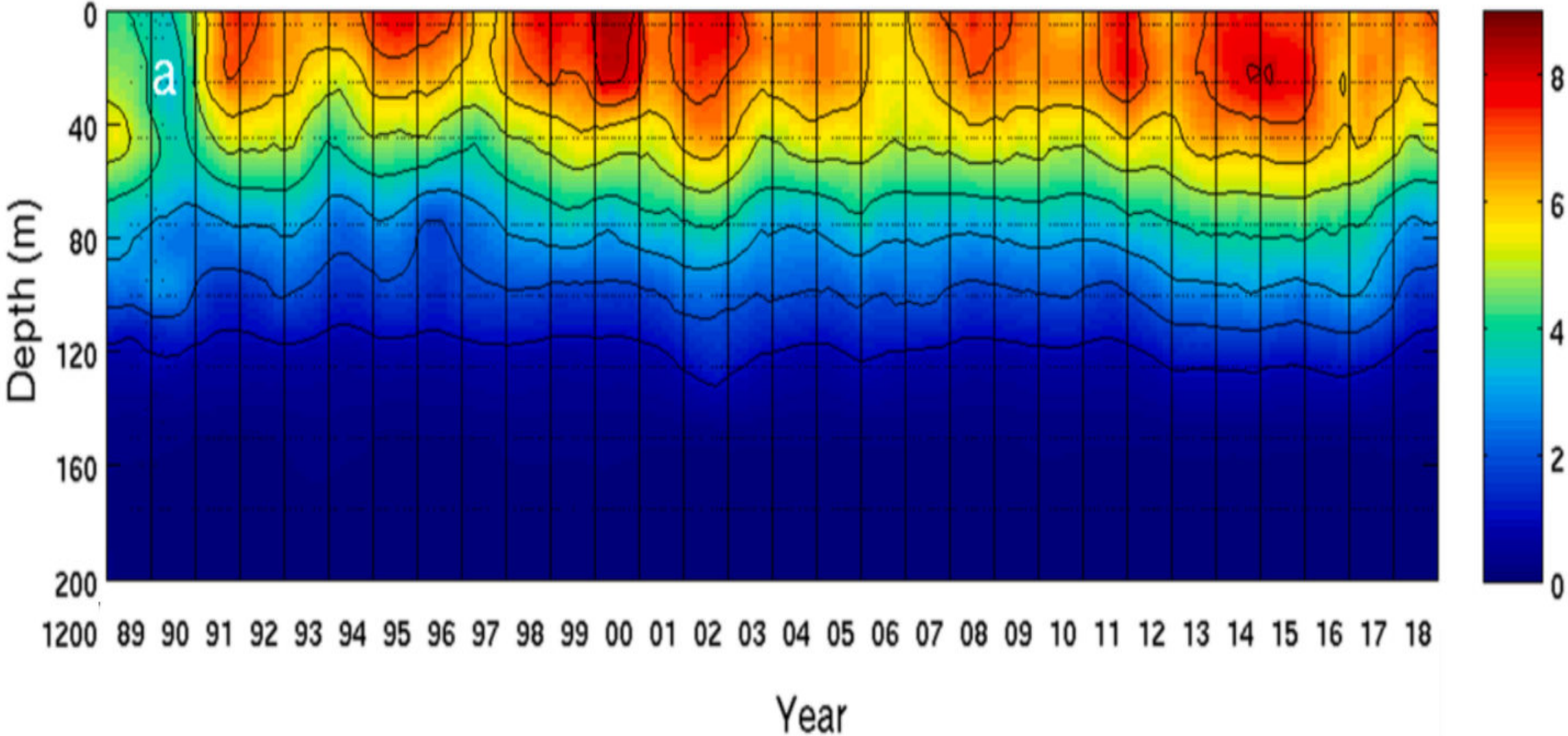
- Deeper in summer



Primary production

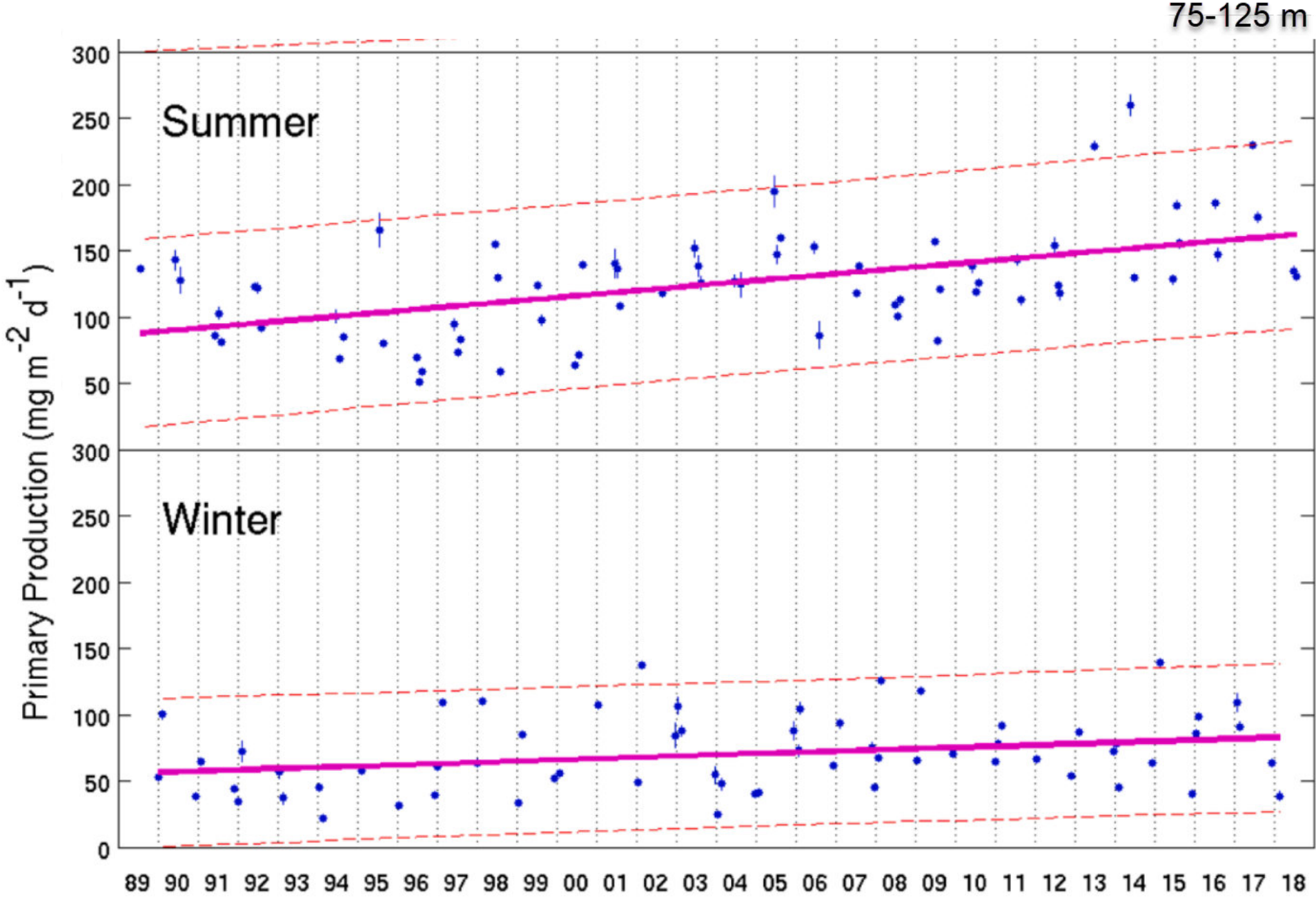
What do you notice ?

^{14}C -based primary production ($\text{mg C m}^{-3} \text{d}^{-1}$)



Long term trends

- CO₂
- Chlorophyll
- Production

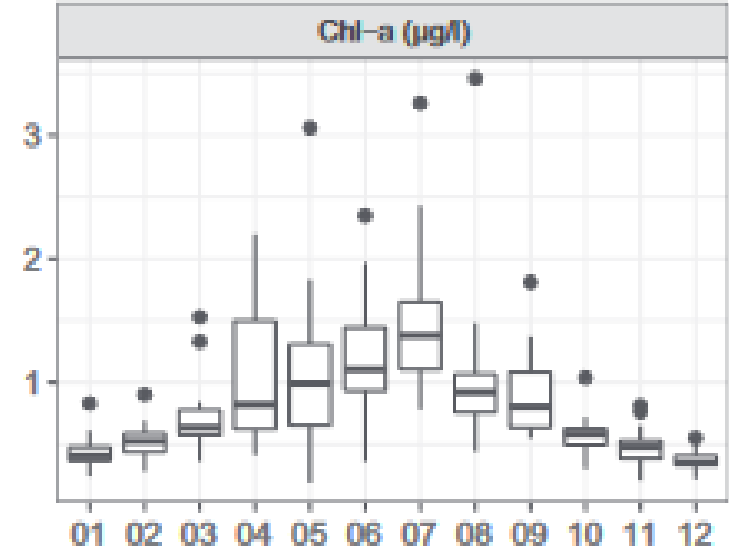
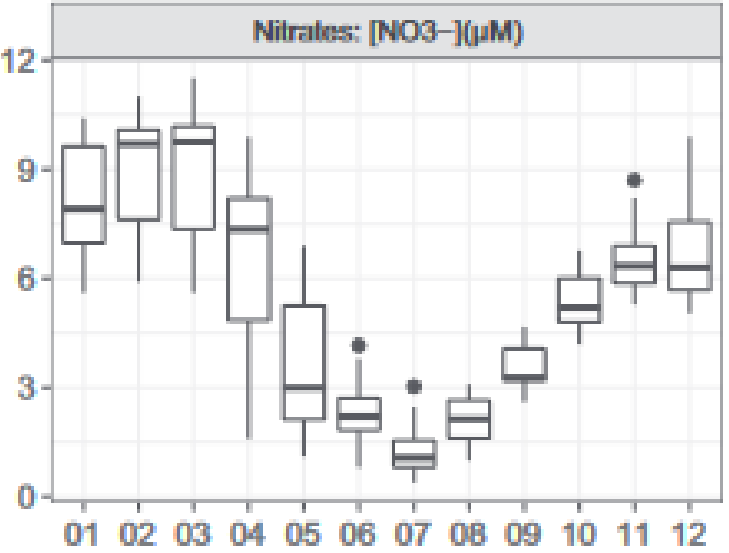
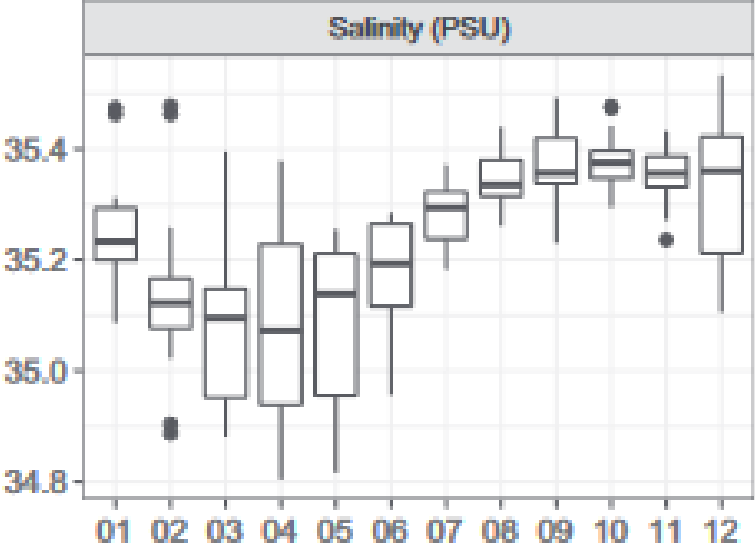
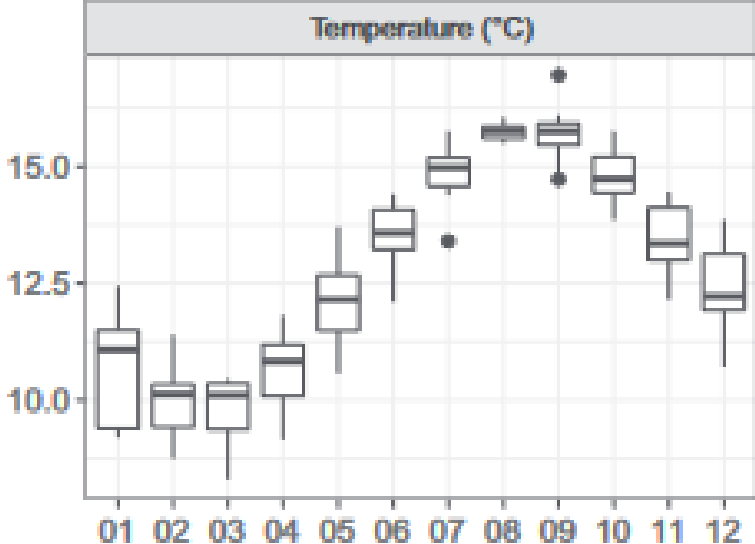
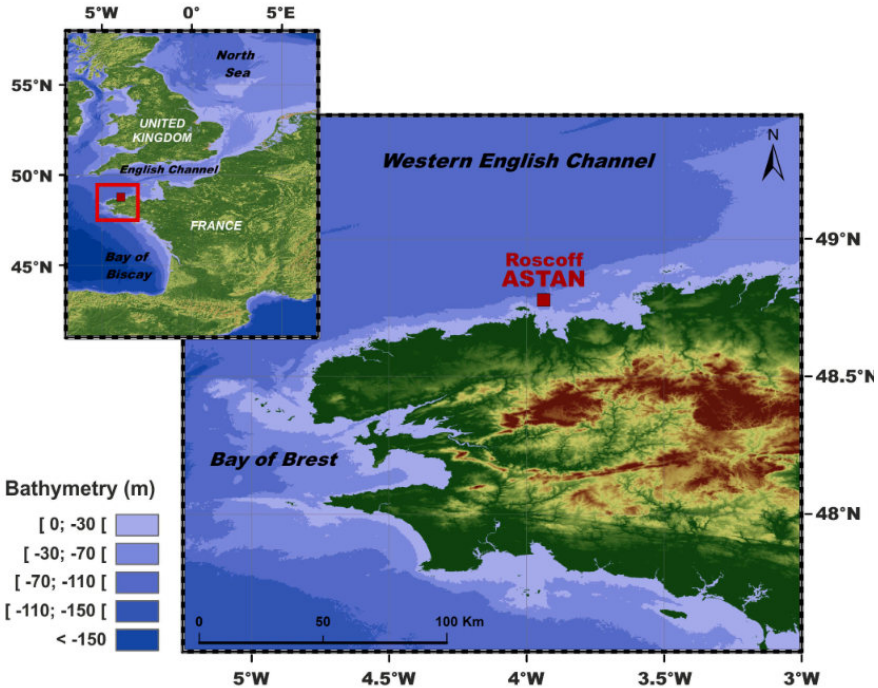


Determining species periodicity

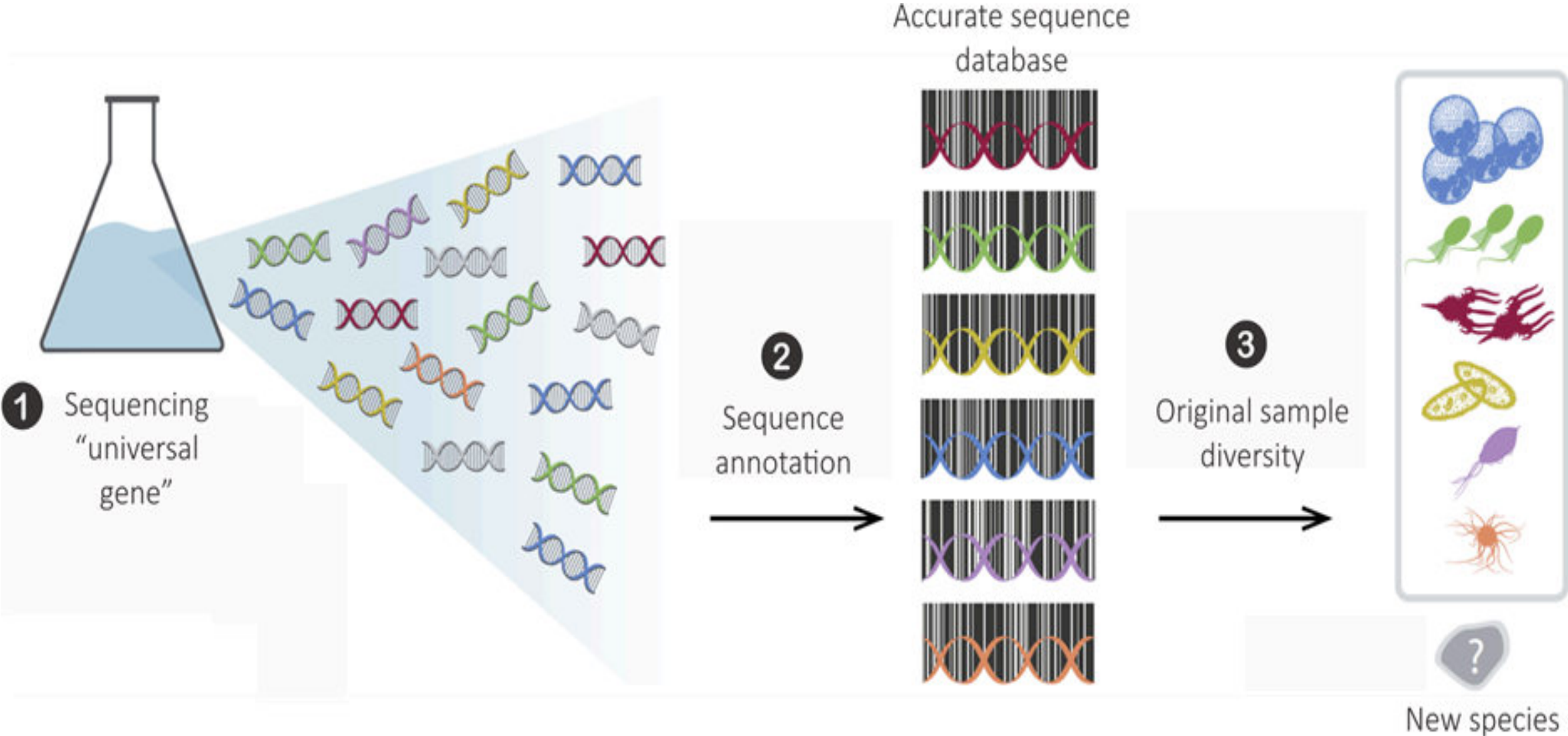


English Channel

- Roscoff
- 2009-2016
- Very strong tide
- Always mixed

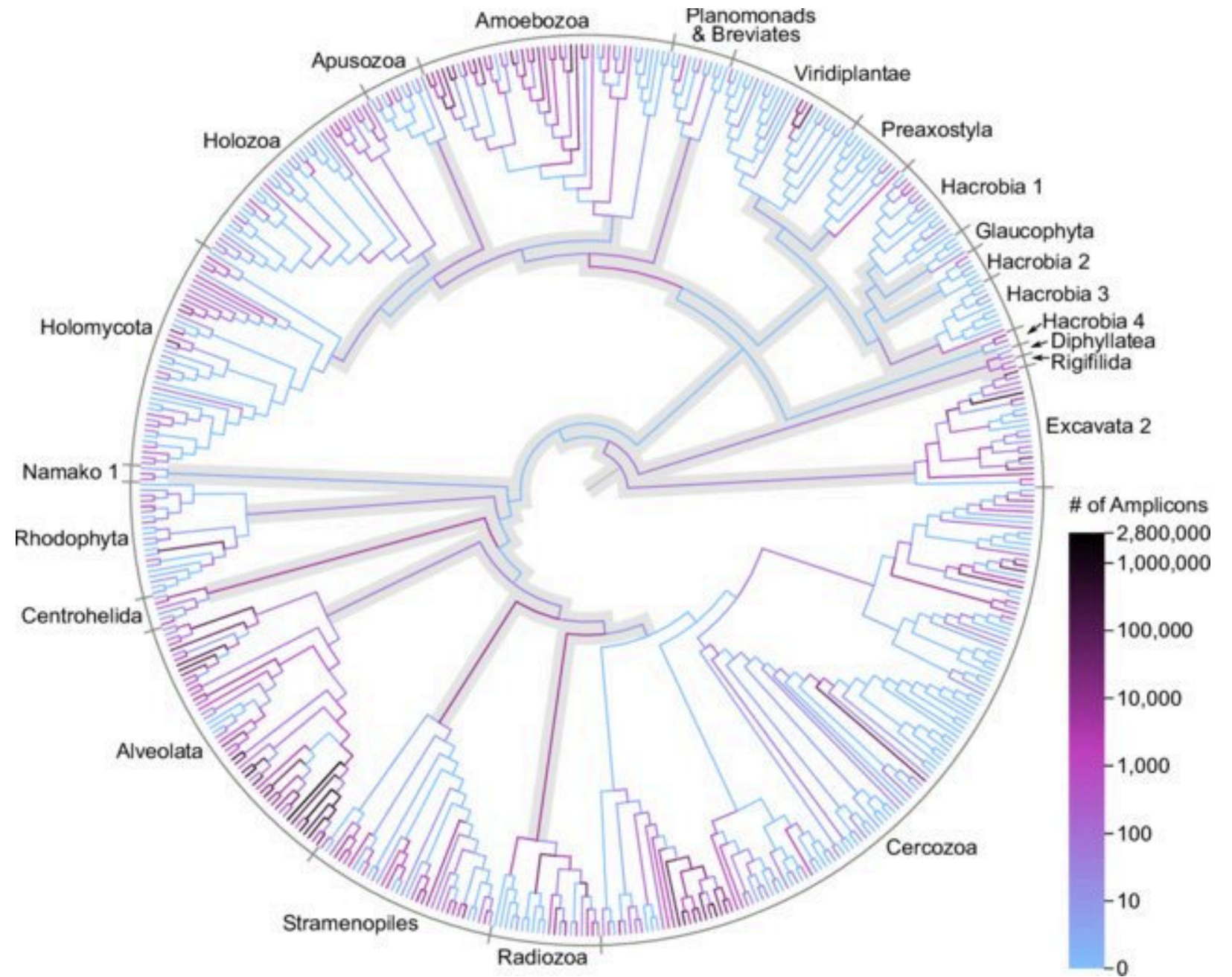


Metabarcoding



Metabarcoding

- Taxonomy of sequences



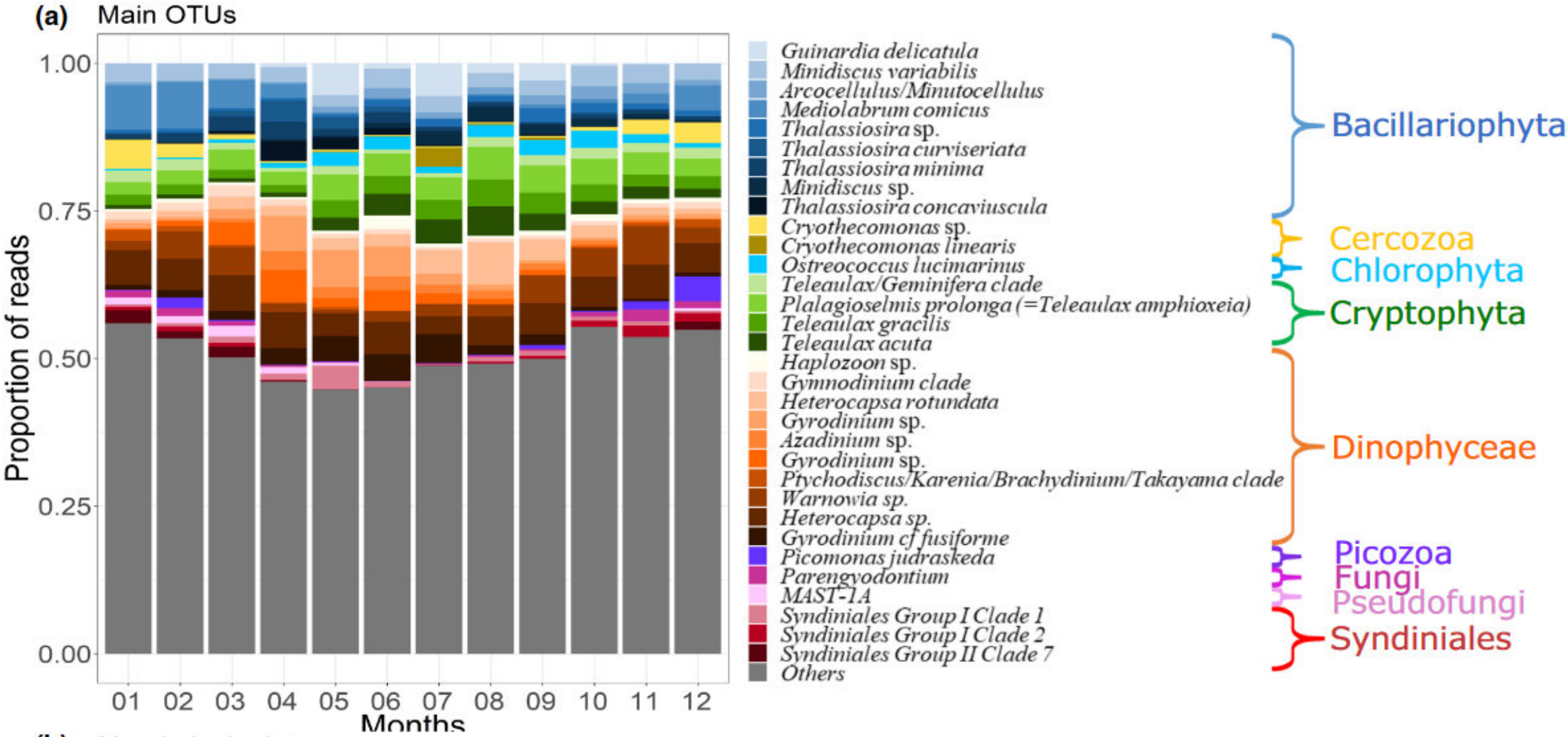
Metabarcoding

- Output of metabarcoding

Taxonomic units (OTUs, ASVs)							Samples														
otu_id	kingdom	supergroup	division	class	genus	species	EC04XS	EC06XS	EC07XS1	EC08XS1	EC11XS2	EC13XS3	EC14XS3	EC15XS4	EC17XS5	EC18XS6	EC19XS6	EC20X16XS10	EC21X16XS11	EC22X16XS11	
12	Eukaryota	Alveolata	Dinoflagellata	Dinophyceae	Gyrodinium	Gyrodinium fusiforme	0	0	0	0	0	0	256	239	0	0	0	0	474	0	11
44	Eukaryota	Alveolata	Dinoflagellata	Dinophyceae	Gonyaulax	Gonyaulax spinifera	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
48	Eukaryota	Opisthokonta	Metazoa	Arthropoda	Temora	Temora turbinata	0	0	0	0	0	414	0	0	0	0	102	88	0	0	0
52	Eukaryota	Opisthokonta	Metazoa	Arthropoda	Bestiolina	Bestiolina similis	0	0	697	0	0	0	478	1452	0	0	0	1748	0	5	0
61	Eukaryota	Stramenopiles	Ochrophyta	Bacillariophyta	Chaetoceros	Chaetoceros sp. P_quina	26	0	0	0	44	40	0	26	0	0	116	37	0	0	0
66	Eukaryota	Opisthokonta	Metazoa	Arthropoda	Paracalanus	Paracalanus aculeatus	0	0	0	0	1831	973	455	186	1644	0	0	0	0	16	0
72	Eukaryota	Stramenopiles	Ochrophyta	Bacillariophyta	Thalassiosira	Thalassiosira sp.	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
78	Eukaryota	Opisthokonta	Metazoa	Urochordata	Oikopleura	Oikopleura dioica	324	238	575	1421	0	0	241	2208	97	246	0	590	0	32	0
79	Eukaryota	Opisthokonta	Metazoa	Cnidaria	Calcigorgia	Calcigorgia beringi	52	0	0	0	0	0	0	0	0	0	0	0	0	0	0
80	Eukaryota	Archaeplastida	Chlorophyta	Mamiellophyceae	Micromonas	Micromonas commoda AB	483	0	0	183	135	96	453	158	719	1006	388	0	1446	4	0
84	Eukaryota	Opisthokonta	Metazoa	Arthropoda	Acrocalanus	Acrocalanus gracilis	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
88	Eukaryota	Opisthokonta	Metazoa	Mollusca	Bathymodiolinae	Bathymodiolinae gen.	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
95	Eukaryota	Opisthokonta	Metazoa	Arthropoda	Parvocalanus	Parvocalanus crassirostris	0	0	0	0	0	0	0	161	0	0	0	0	0	0	0
108	Eukaryota	Opisthokonta	Metazoa	Urochordata	Oikopleura	Oikopleura dioica	315	0	400	540	108	0	0	0	0	784	64	339	0	32	0
115	Eukaryota	Alveolata	Dinoflagellata	Dinophyceae	Dinophyceae_XXX	Dinophyceae_XXX_sp.	151	0	0	0	0	0	1056	488	0	269	0	315	2079	4	0
119	Eukaryota	Opisthokonta	Metazoa	Arthropoda	Paracalanus	Paracalanus sp.	81	0	1925	855	0	0	0	371	0	113	179	0	11	0	0
127	Eukaryota	Archaeplastida	Chlorophyta	Mamiellophyceae	Micromonas	Micromonas clade B war	246	0	0	0	0	109	251	178	153	226	152	233	0	0	0
136	Eukaryota	Hacrobia	Cryptophyta	Cryptophyceae	Geminigera	Geminigera cryophila	347	299	0	289	135	52	247	146	194	430	201	109	341	2	0
141	Eukaryota	Archaeplastida	Chlorophyta	Trebouxiophyceae	Nannochloris	Nannochloris sp.	0	0	0	0	0	44	0	0	0	0	0	0	0	0	0
146	Eukaryota	Opisthokonta	Metazoa	Arthropoda	Bestiolina	Bestiolina sp.	0	0	706	83	558	0	0	0	0	0	0	0	0	51	0
148	Eukaryota	Archaeplastida	Chlorophyta	Trebouxiophyceae	Nannochloris	Nannochloris sp.	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
151	Eukaryota	Opisthokonta	Metazoa	Arthropoda	Oithona	Oithona davisae	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
171	Eukaryota	Archaeplastida	Chlorophyta	Mamiellophyceae	Ostreococcus	Ostreococcus sp.	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
173	Eukaryota	Alveolata	Dinoflagellata	Dinophyceae	Dinophyceae_XXX	Dinophyceae_XXX_sp.	0	54	551	0	0	0	0	0	0	0	0	0	0	14	0
175	Eukaryota	Stramenopiles	Ochrophyta	Bacillariophyta	Cerataulina	Cerataulina pelagica	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
177	Eukaryota	Stramenopiles	Ochrophyta	Bacillariophyta	Cyclotella	Cyclotella choctawhatchee	0	0	0	0	0	47	67	0	0	0	0	0	0	0	0
190	Eukaryota	Alveolata	Dinoflagellata	Dinophyceae	Gyrodinium	Gyrodinium gutrula	0	131	176	0	0	0	0	0	0	0	118	0	0	8	0
191	Eukaryota	Rhizaria	Radioliana	RAD-B	RAD-B-Group-IV_X	RAD-B-Group-IV_X_sp.	0	20	0	51	0	0	0	0	656	68	0	0	0	0	0
193	Eukaryota	Opisthokonta	Metazoa	Arthropoda	Acrocalanus	Acrocalanus gracilis	0	0	0	0	0	0	0	0	0	0	1252	0	0	0	0
194	Eukaryota	Opisthokonta	Metazoa	Porifera	Unclassified_Halichondrida	Halichondrida sp.	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
198	Eukaryota	Opisthokonta	Metazoa	Arthropoda	Oithona	Oithona similis	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
199	Eukaryota	Alveolata	Dinoflagellata	Dinophyceae	Woloszynskia	Woloszynskia halophila	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
205	Eukaryota	Archaeplastida	Chlorophyta	Mamiellophyceae	Ostreococcus	Ostreococcus sp.	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
208	Eukaryota	Rhizaria	Cercozoa	Filosa-Imbricate	Novel-clade-2_X	Novel-clade-2_X_sp.	329	40	0	0	0	58	0	18	0	123	123	0	0	2	0
209	Eukaryota	Opisthokonta	Metazoa	Cnidaria	Forskalia	Forskalia edwardsi	0	0	0	0	0	0	0	0	0	209	0	0	0	0	0
217	Eukaryota	Rhizaria	Cercozoa	Filosa-Thecofilo	TAGIR1-lineage_X	TAGIR1-lineage_X_sp.	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
219	Eukaryota	Stramenopiles	Ochrophyta	Bacillariophyta	Thalassiosira	Thalassiosira hispida	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
224	Eukaryota	Stramenopiles	Ochrophyta	Bacillariophyta	Cyclotella	Cyclotella choctawhatchee	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
226	Eukaryota	Opisthokonta	Metazoa	Arthropoda	Oithona	Oithona davisae	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
227	Eukaryota	Opisthokonta	Metazoa	Arthropoda	Artemia	Artemia salina	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
229	Eukaryota	Archaeplastida	Chlorophyta	Mamiellophyceae	Ostreococcus	Ostreococcus clade B	0	0	0	57	0	0	0	0	0	0	0	129	0	0	0

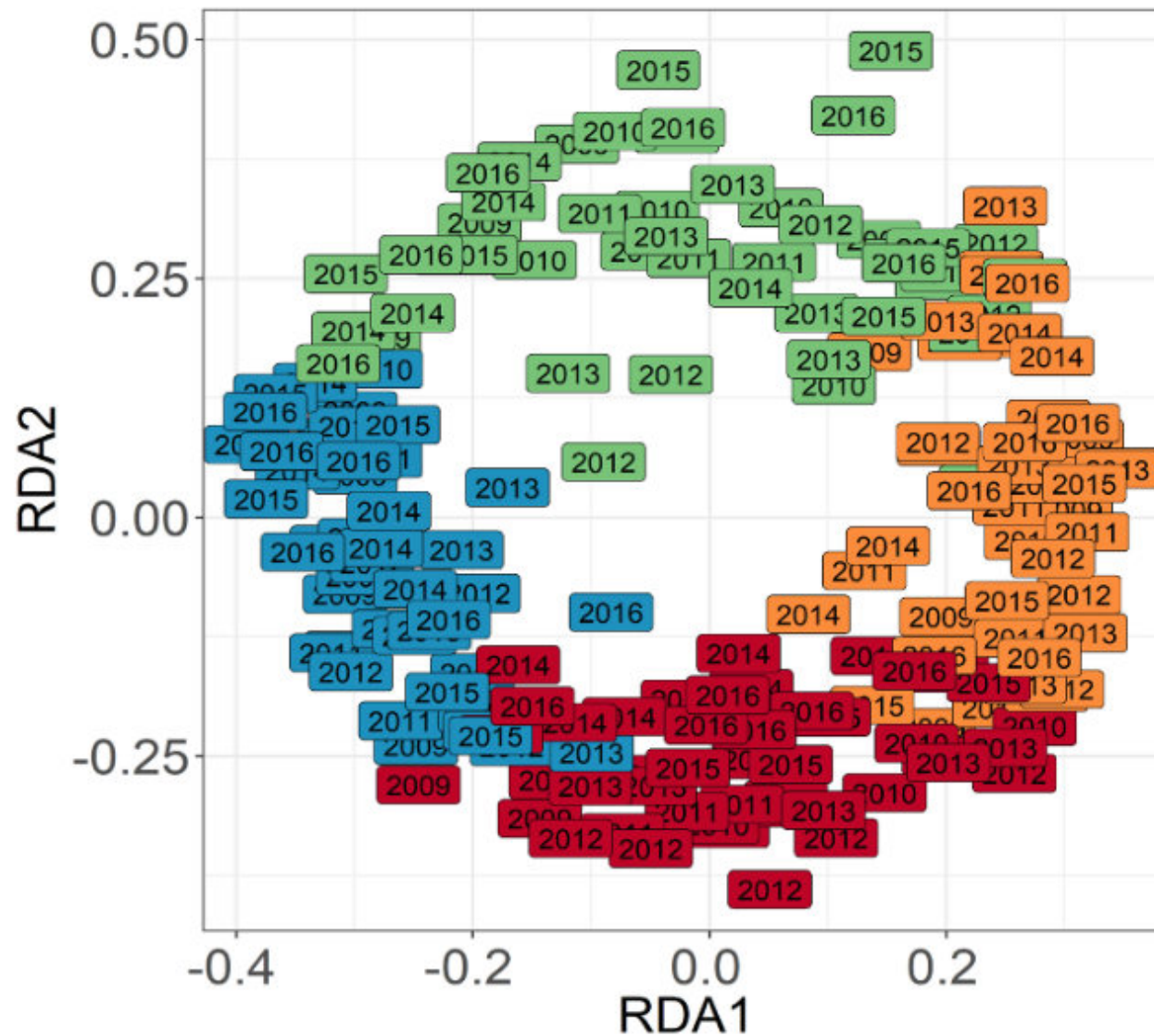
Number of sequences

Mean community composition



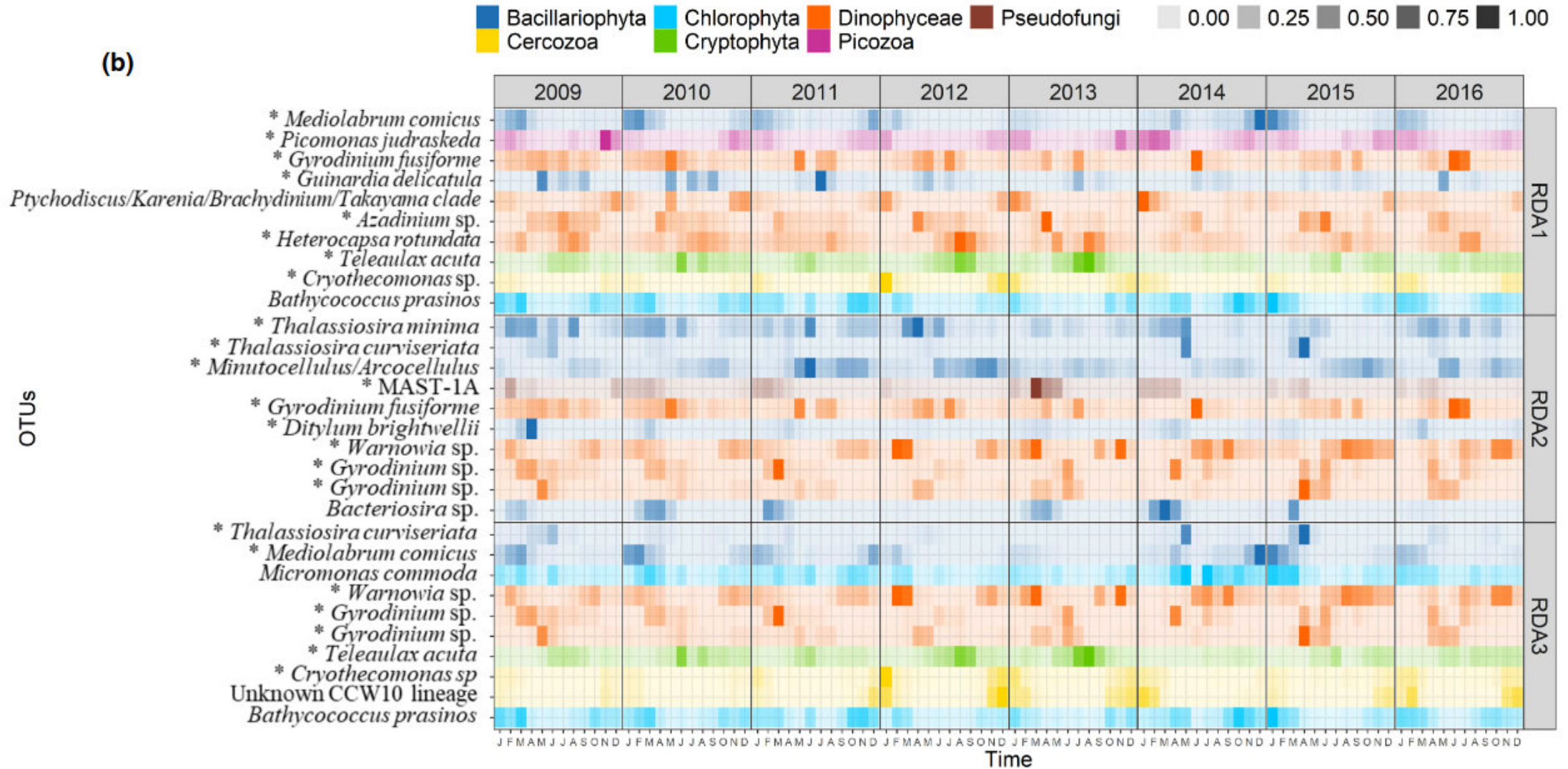
Yearly cycle

- Redundancy analysis (RDA)

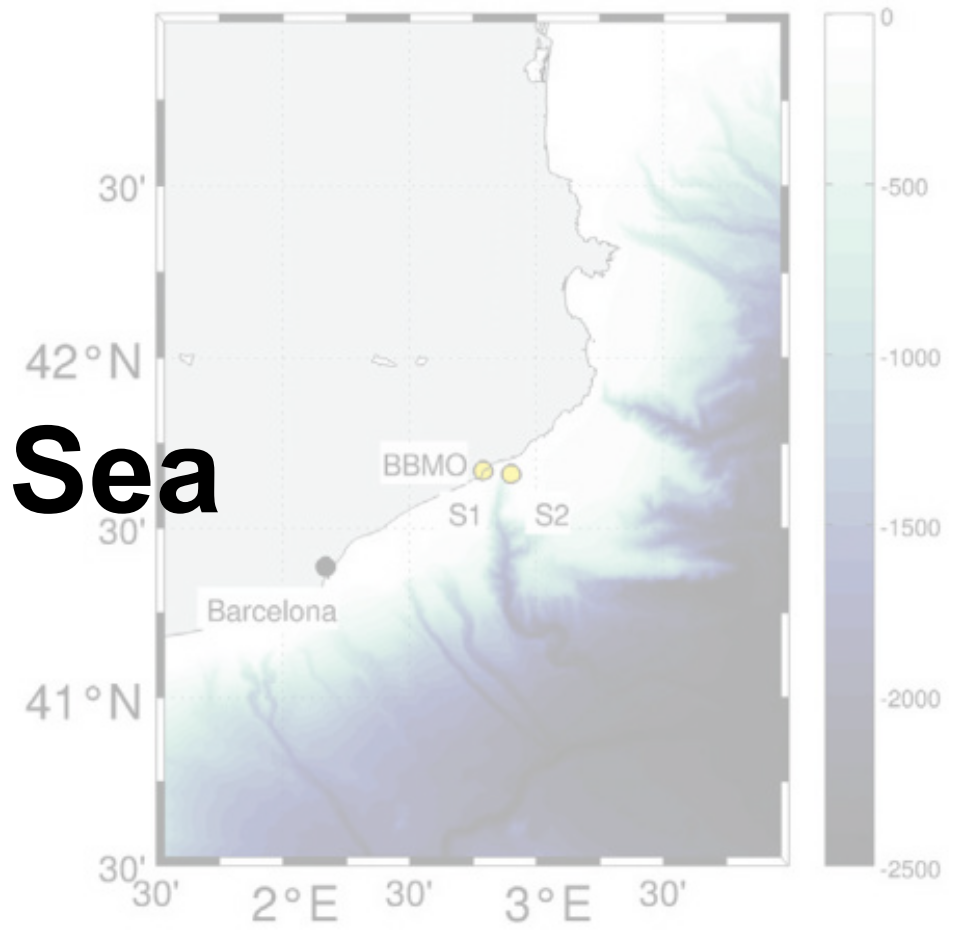
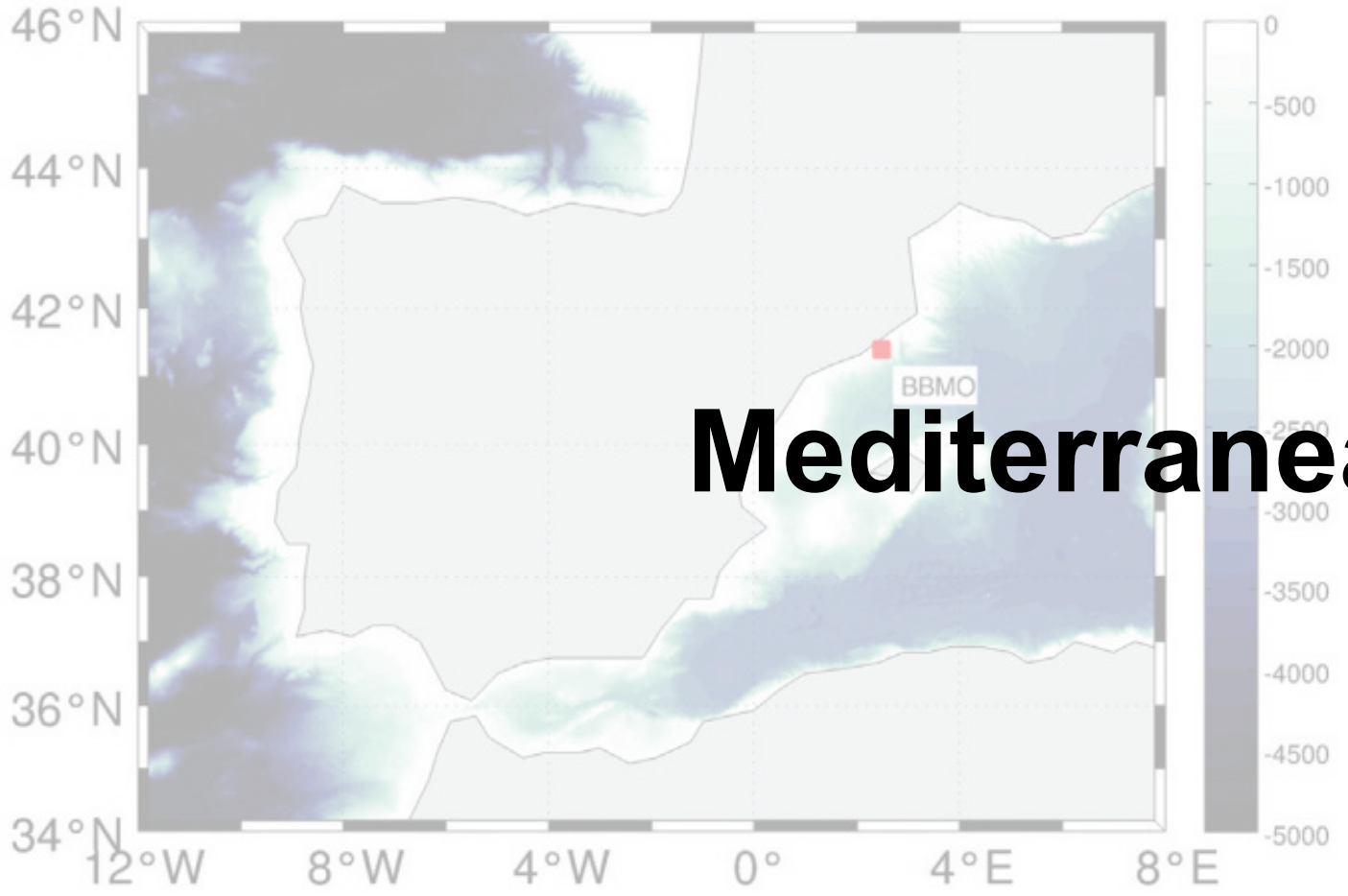


Yearly cycle

- Redundancy analysis (RDA)

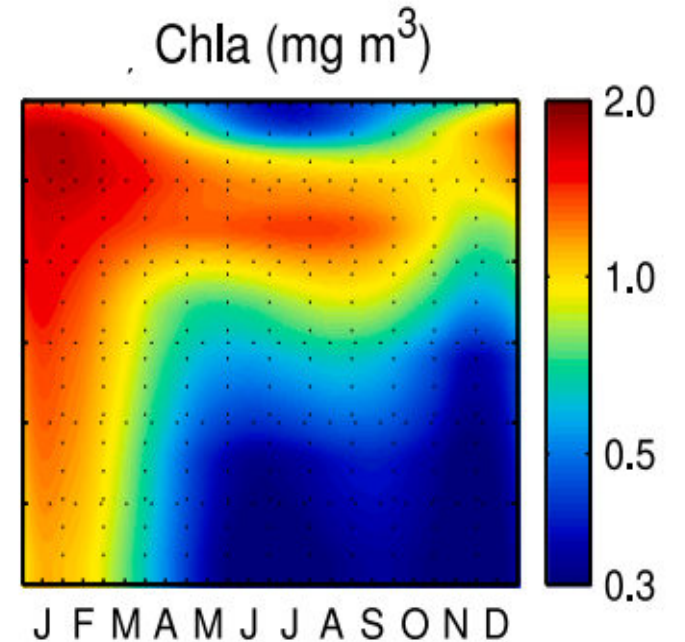
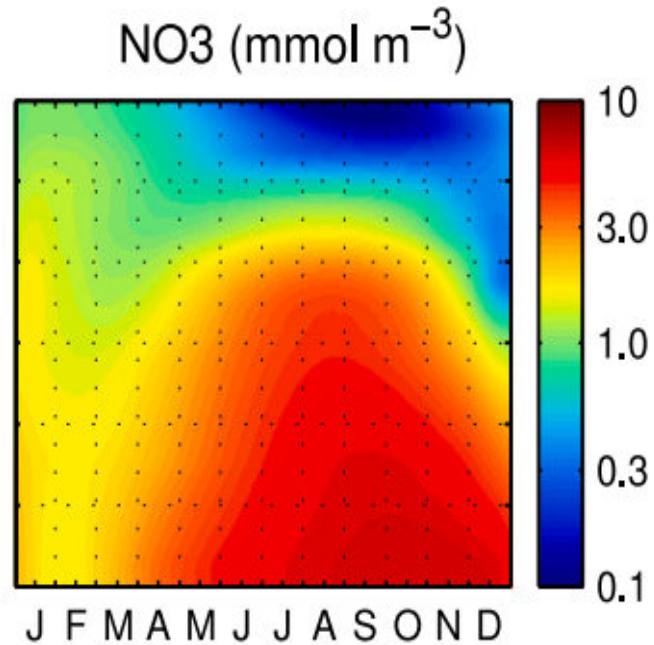
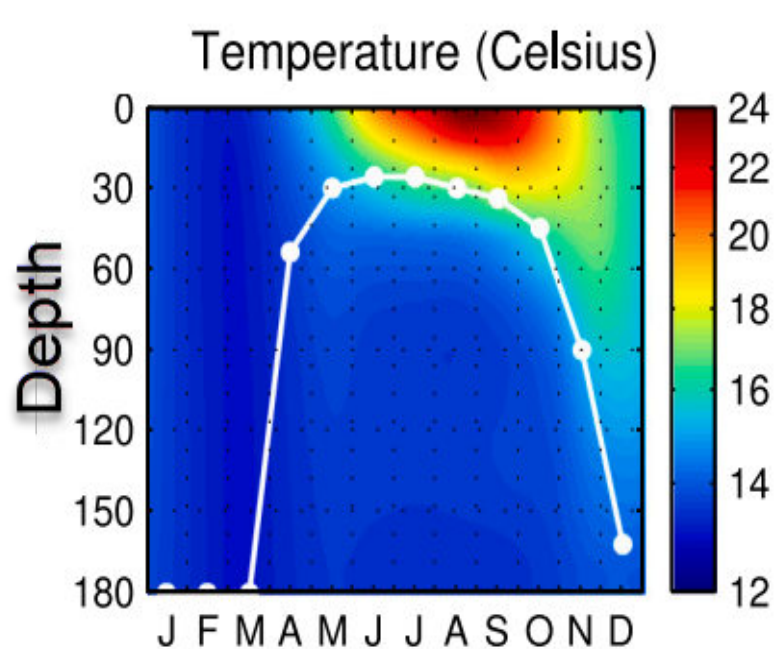
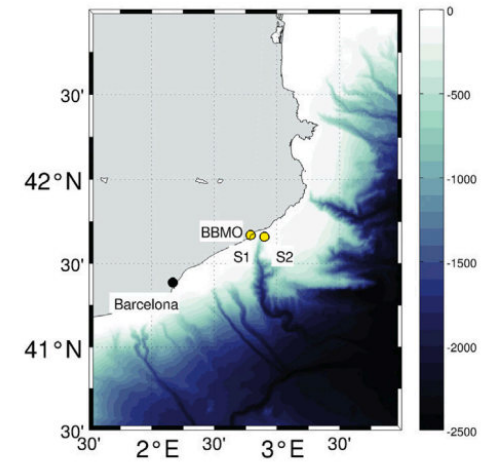
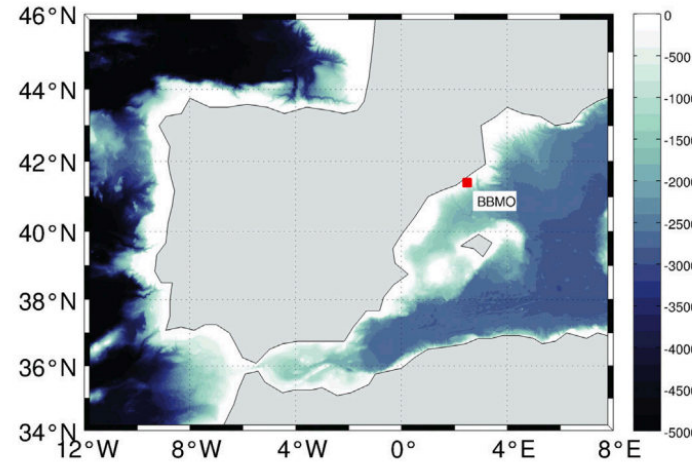


Mediterranean Sea

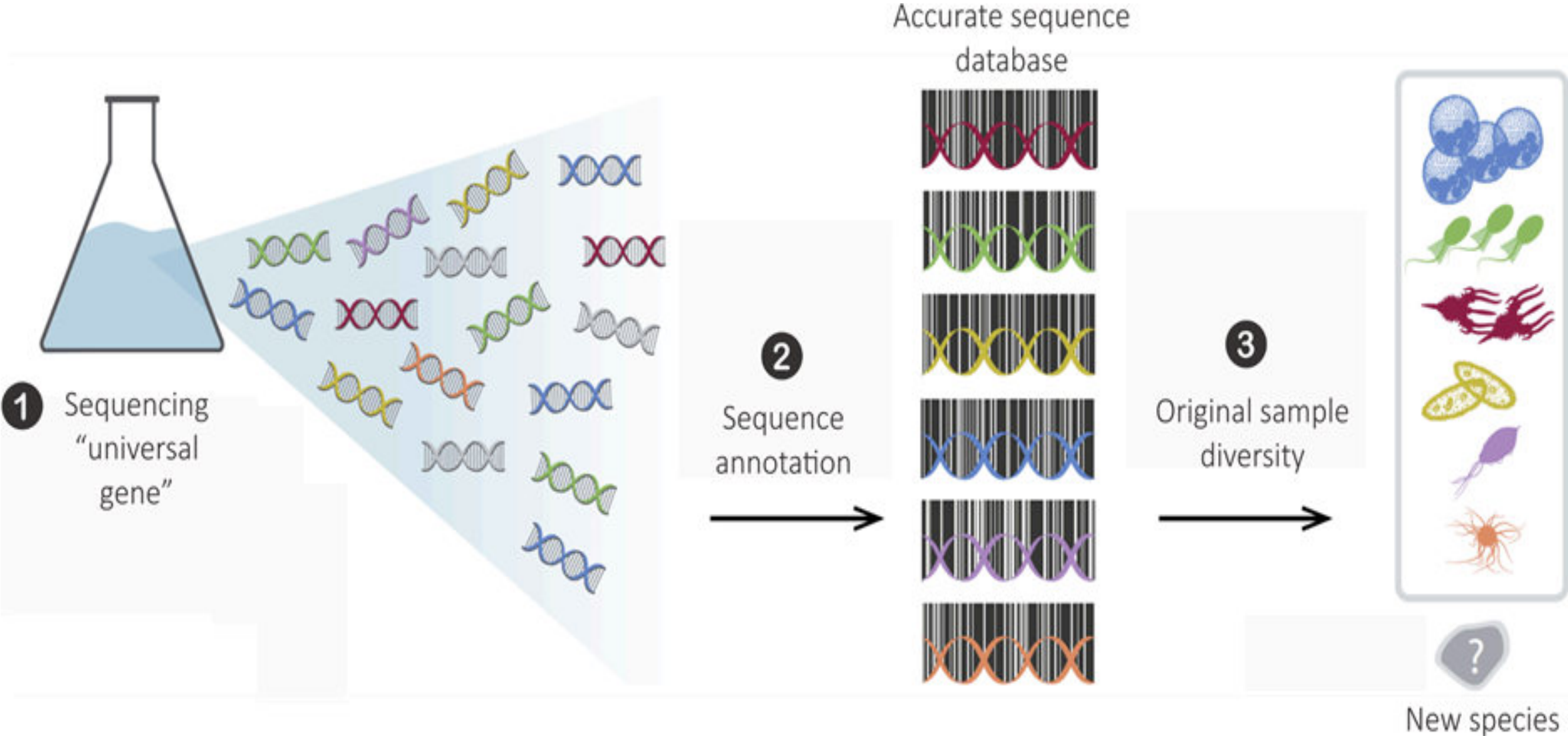


Mediterranean Sea

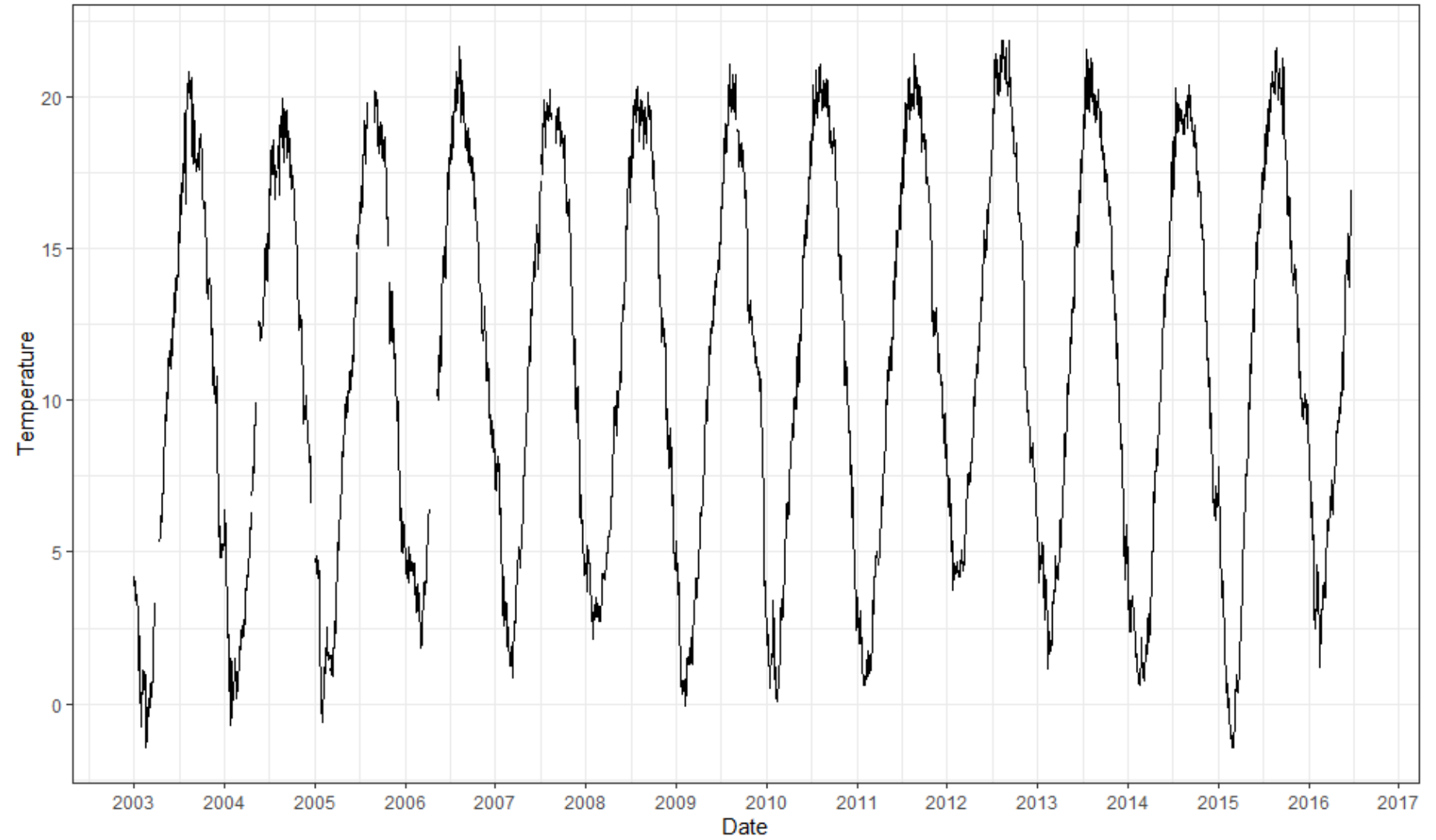
- Blanes Bay (near Barcelona)
- 2004-2013
- Winter mixing
- January bloom
- Which groups/species exhibit periodic recurrence ?



Metabarcoding

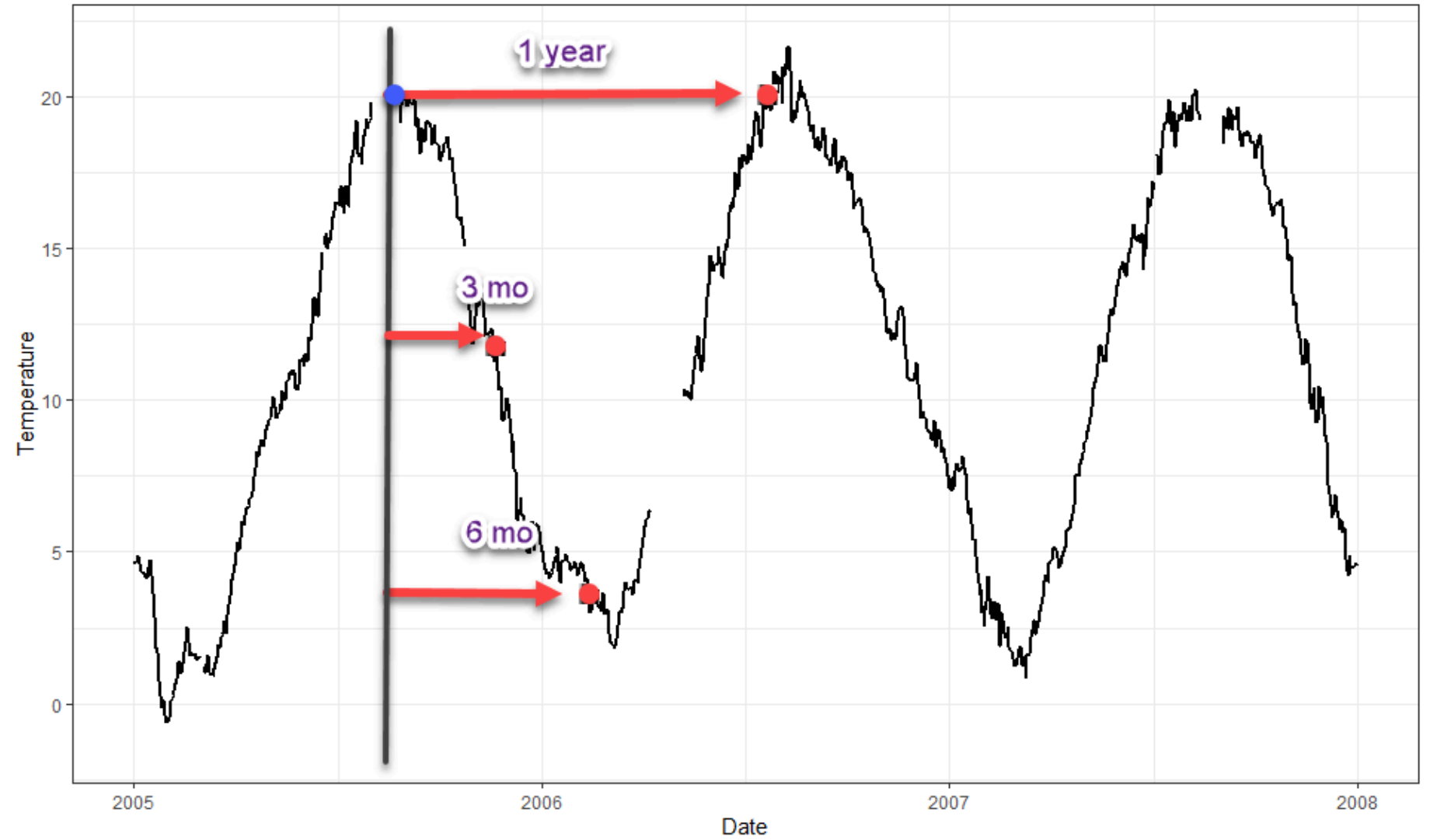


How to determine periodicity ?



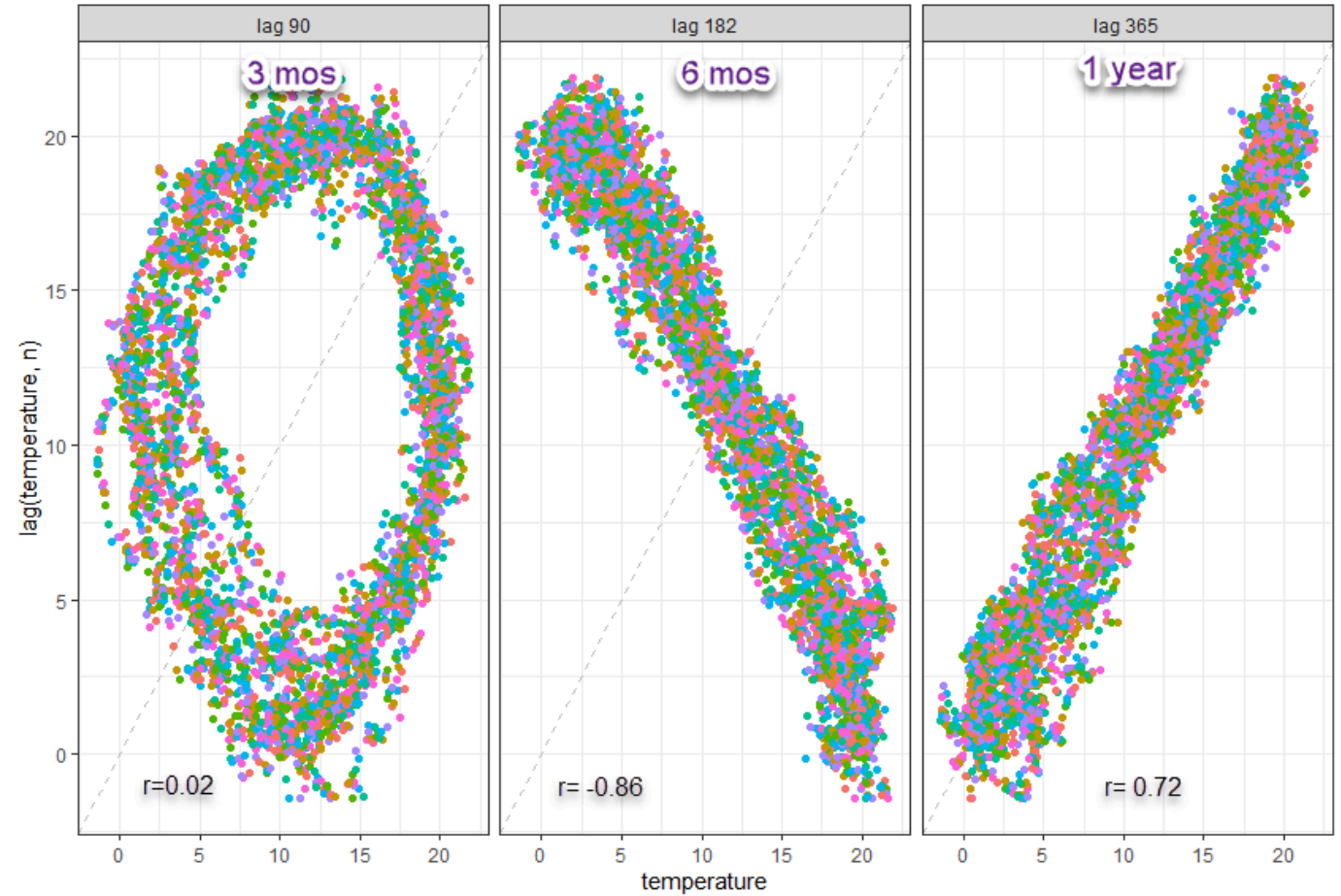
How to determine periodicity ?

- Autocorrelation



How to determine periodicity ?

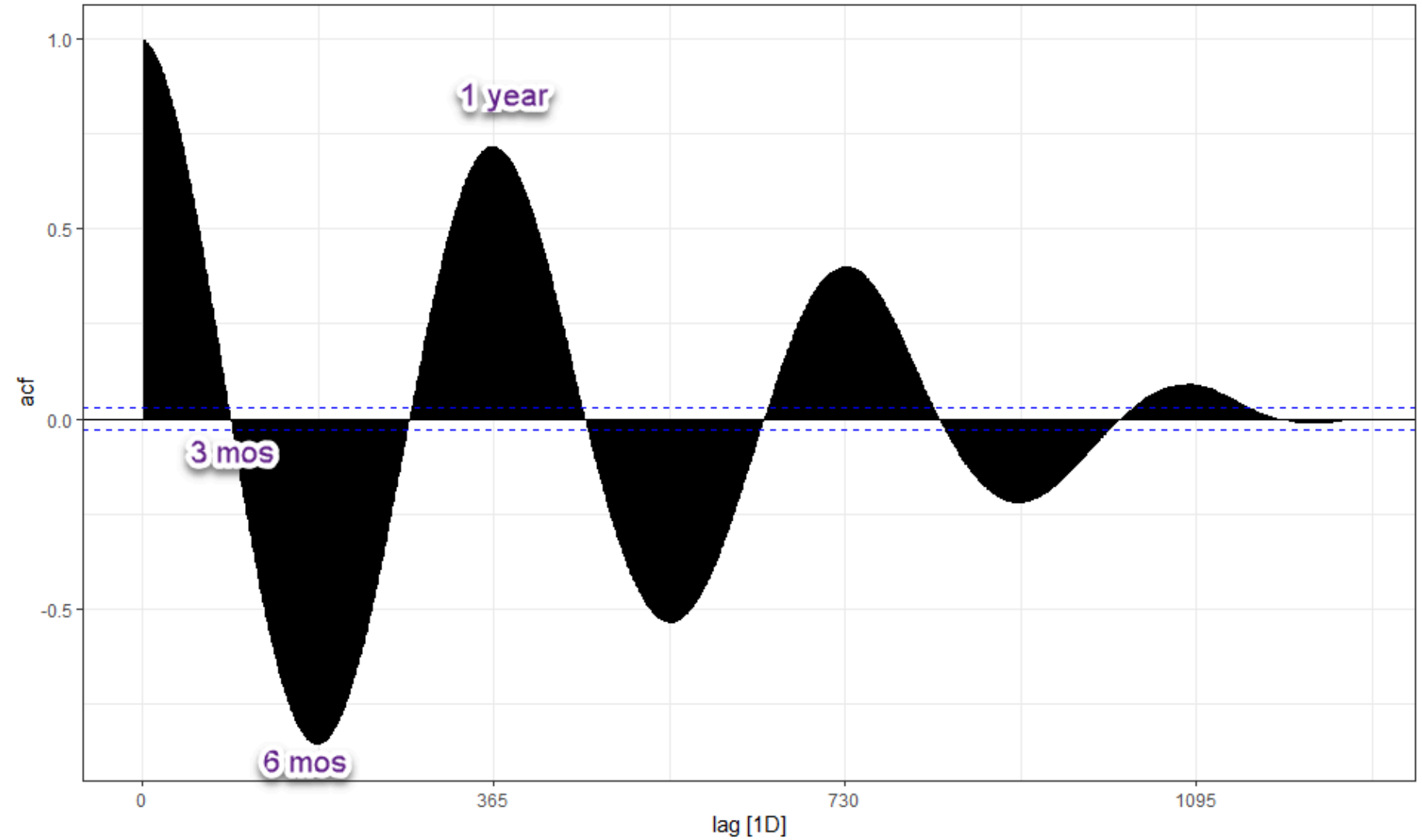
- Autocorrelation



$$r_{xy} = \frac{\sum_{i=1}^n (x_i - \bar{x})(y_i - \bar{y})}{\sqrt{\sum_{i=1}^n (x_i - \bar{x})^2} \sqrt{\sum_{i=1}^n (y_i - \bar{y})^2}}$$

How to determine periodicity ?

- Autocorrelation

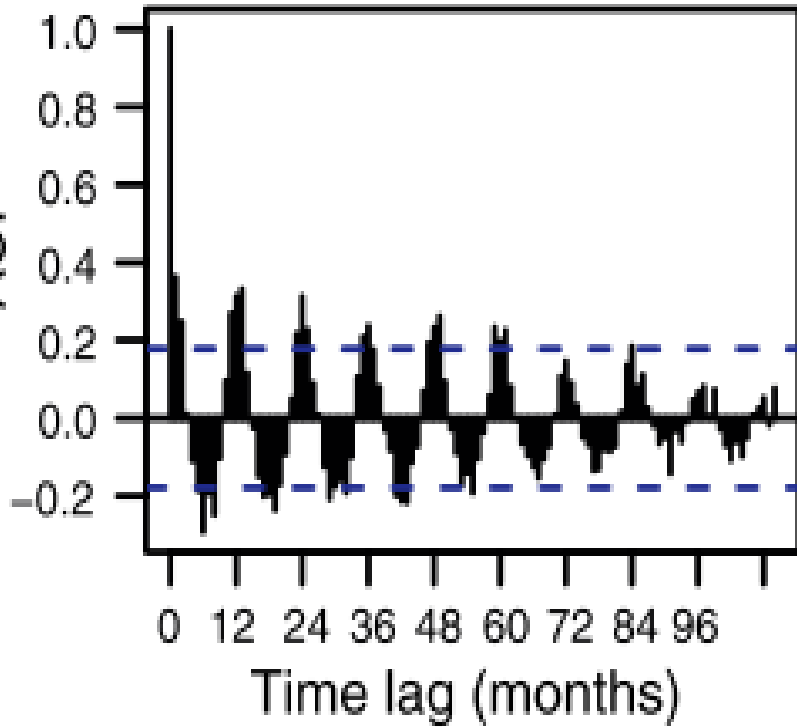


Class periodicity

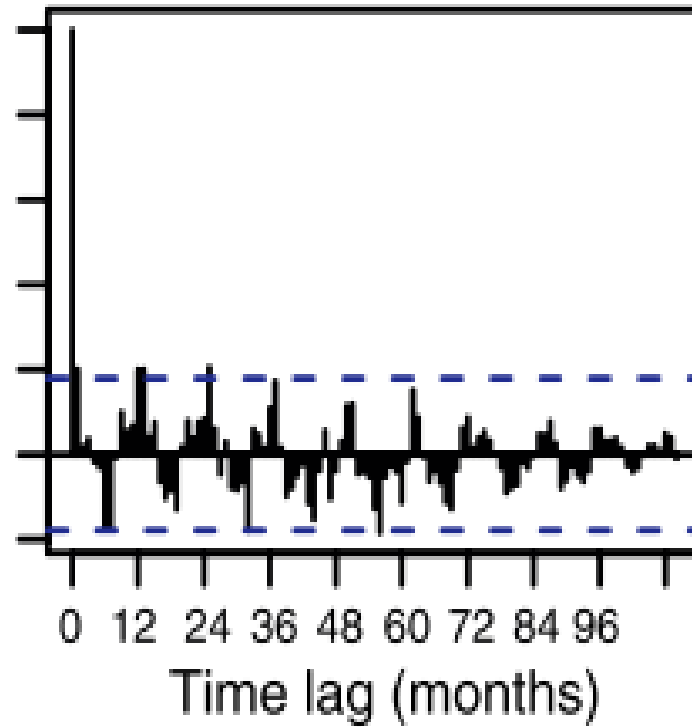
- Autocorrelation function

(b)

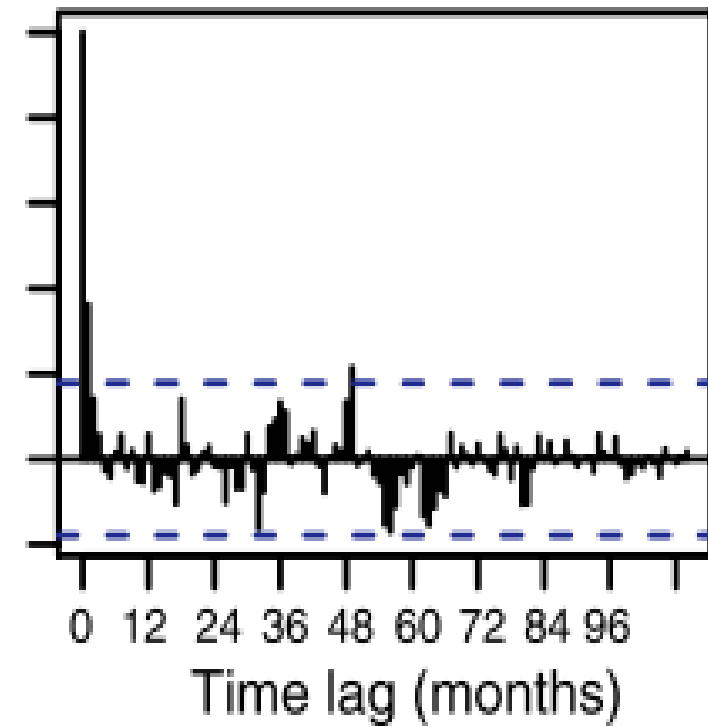
Mamiellophyceae
RI = 2.2



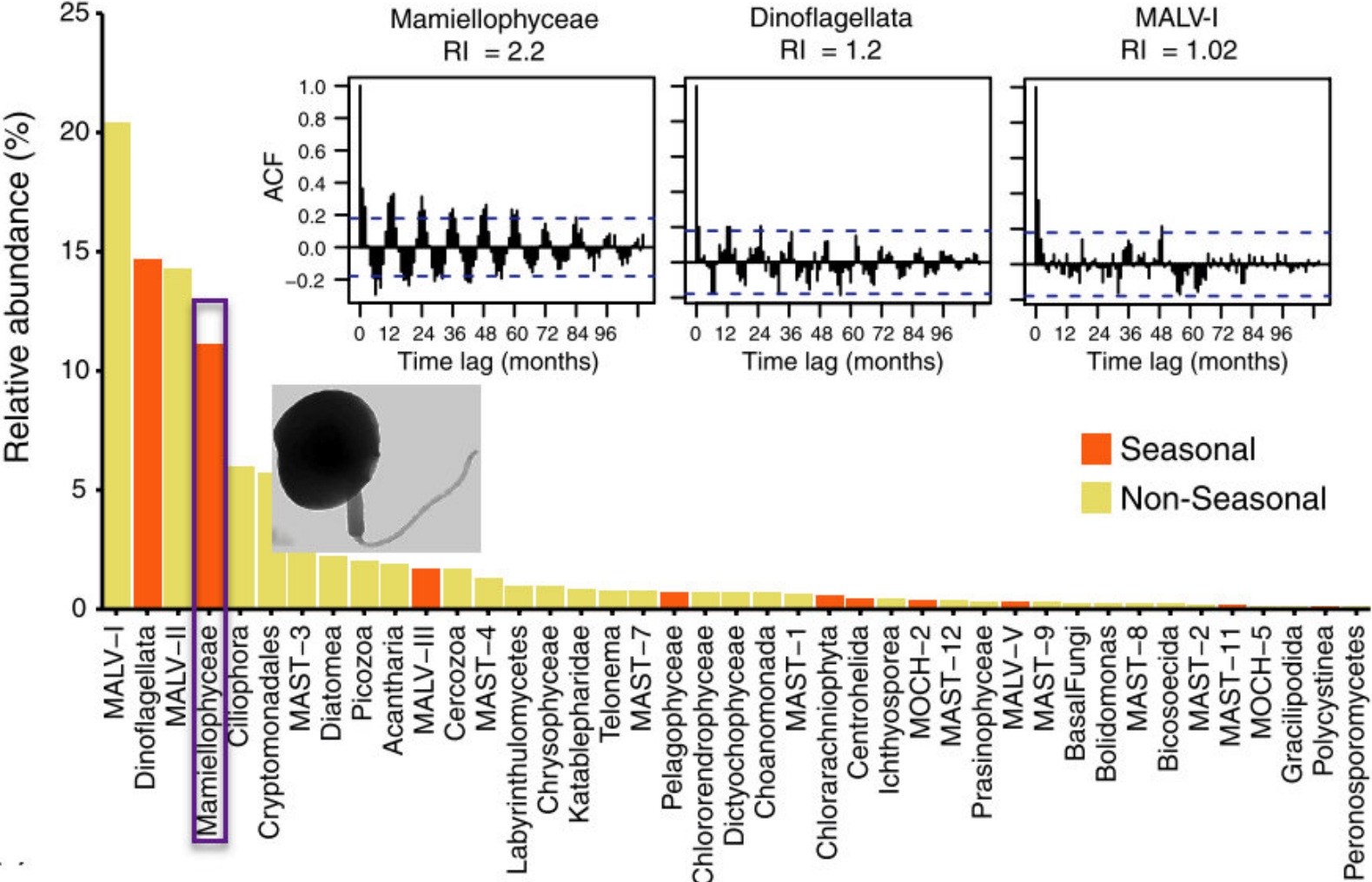
Dinoflagellata
RI = 1.2



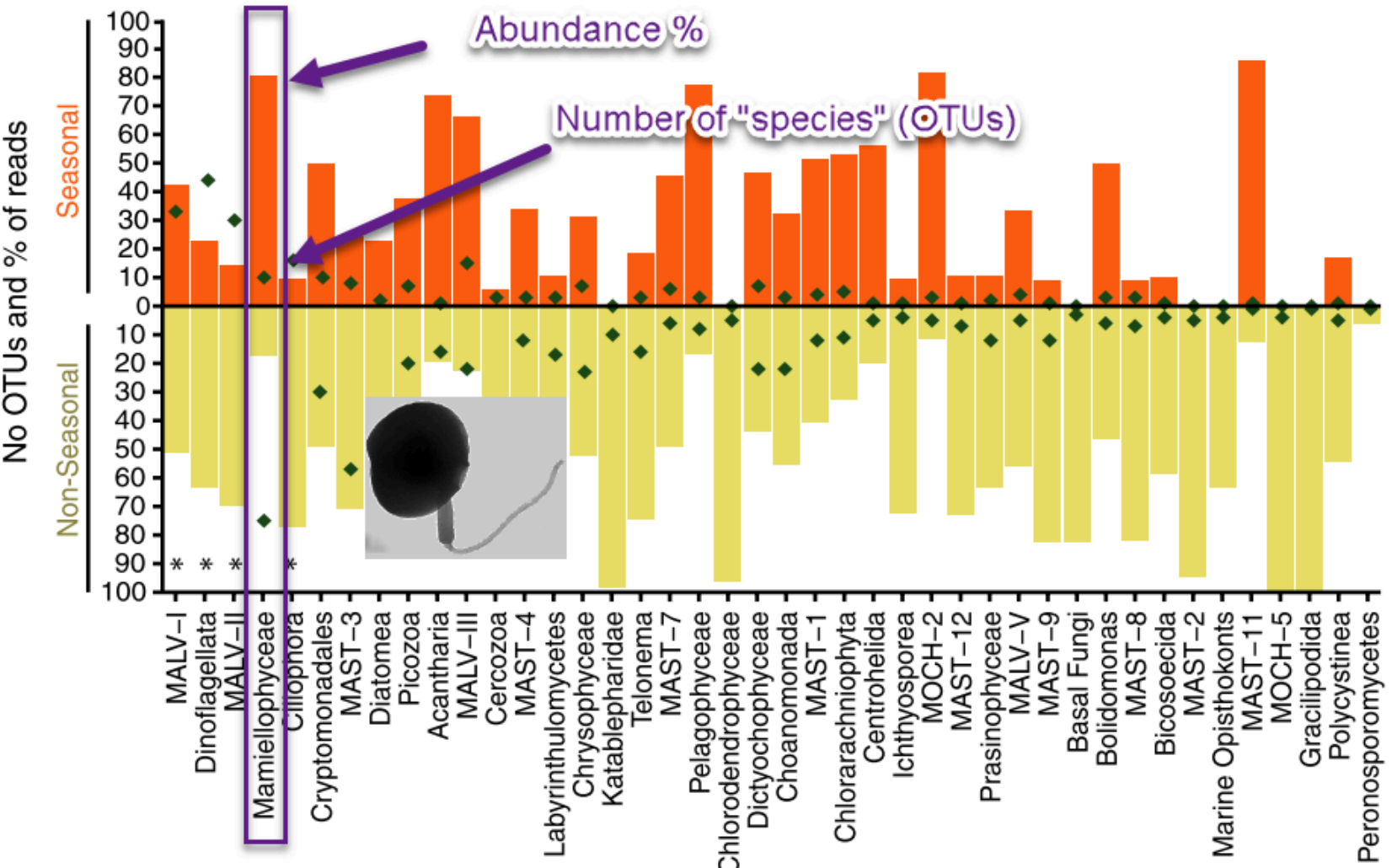
MALV-I
RI = 1.02



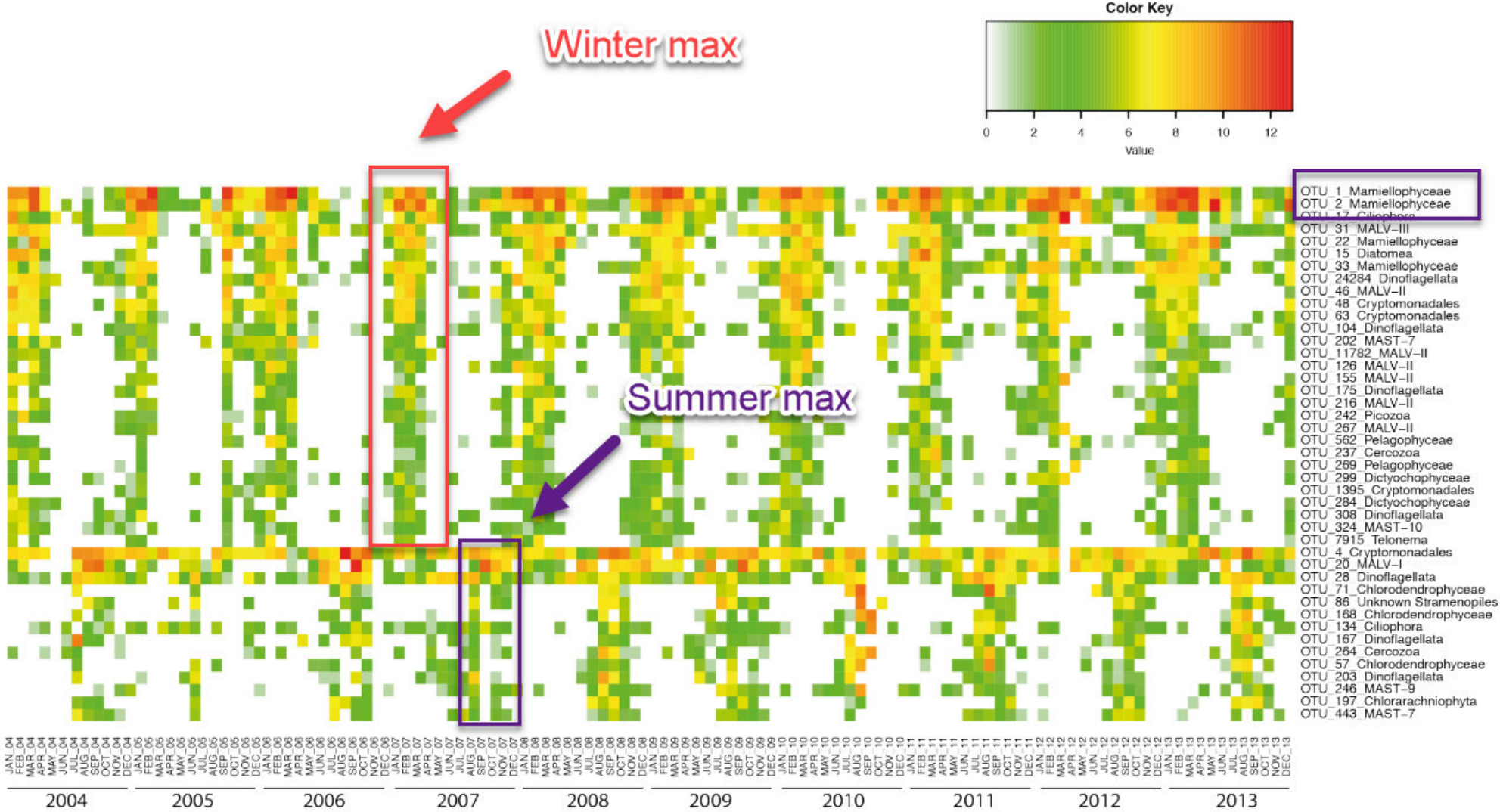
Class periodicity



Species periodicity



Species periodicity



Species dynamics

What drives the *Synechococcus* bloom

Synechococcus

- Discovered in 1979 by John Waterbury
- Epifluorescence microscopy

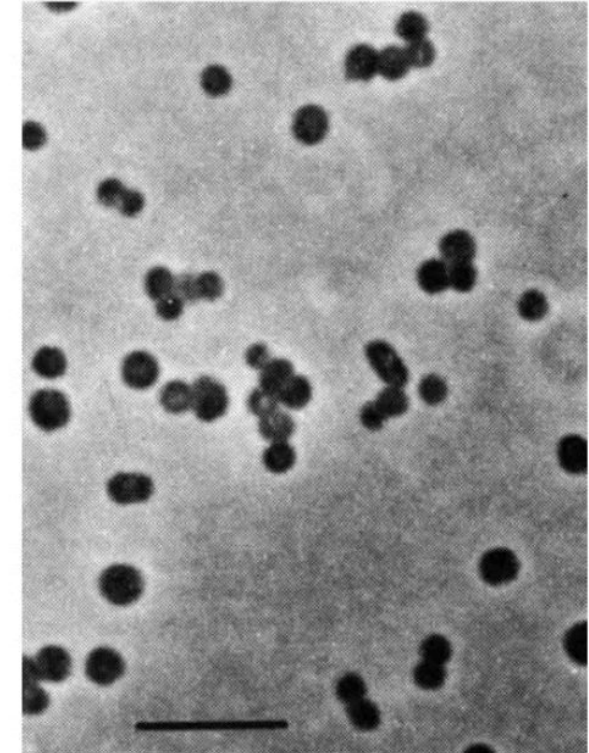
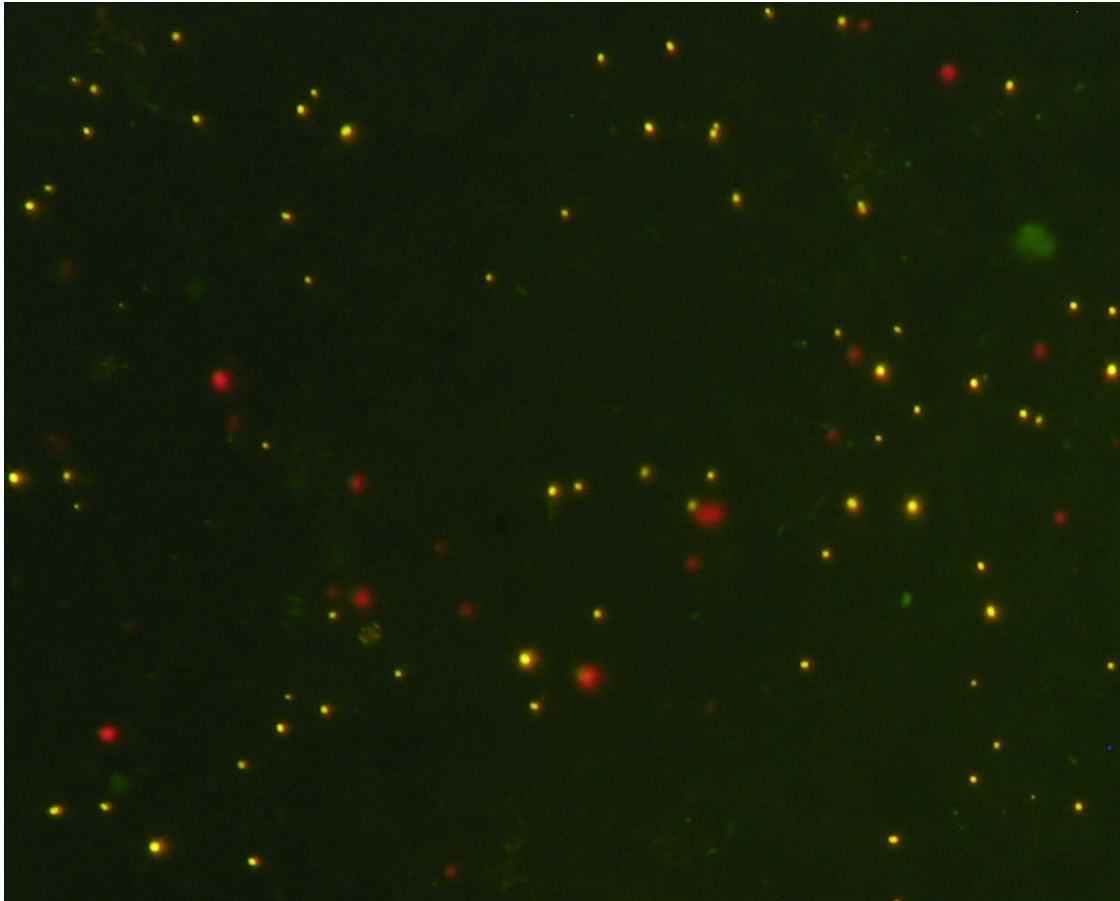
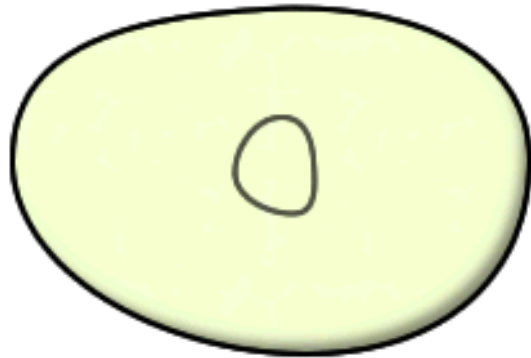


Fig. 1 Phase contrast photomicrograph of *Synechococcus* sp. (strain Syn-48) illustrating general cell morphology (scale bar, 5.0 μm).

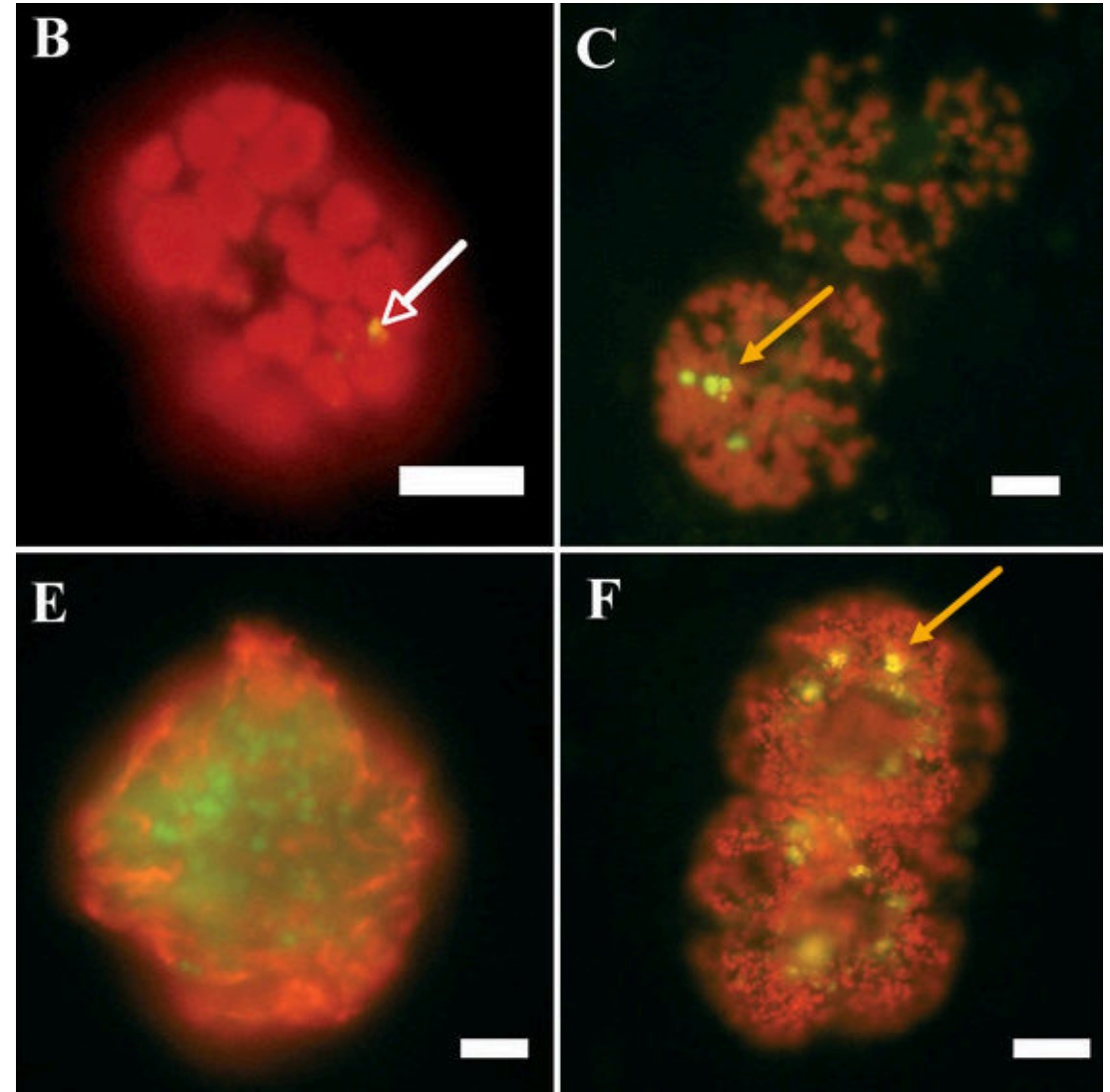
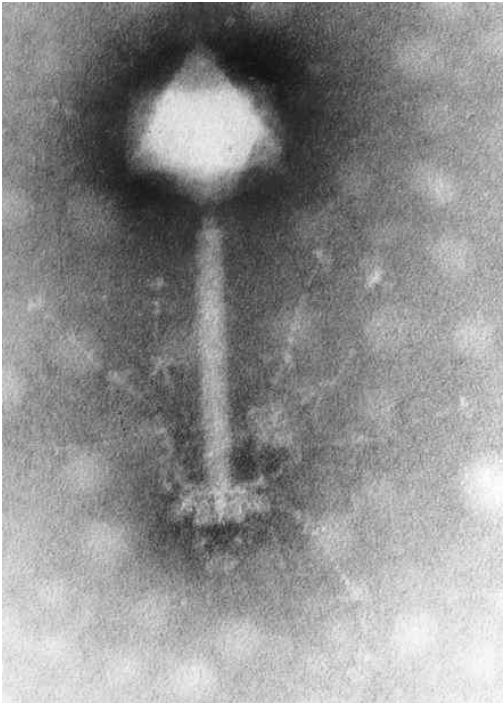
Cell multiplication

- Binary fission
- Typically once every day



Cell disappearance

- Virus
- Predation
- Cell death (UV, nutrient deprivation)



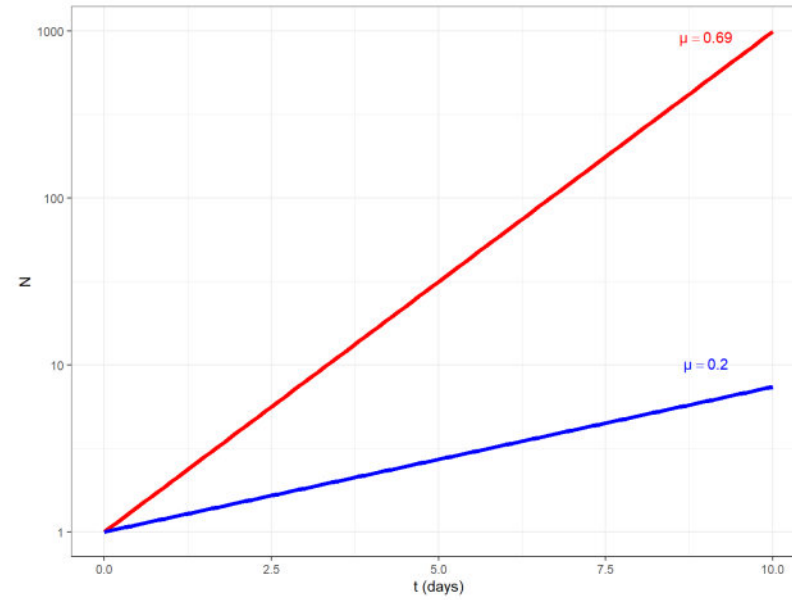
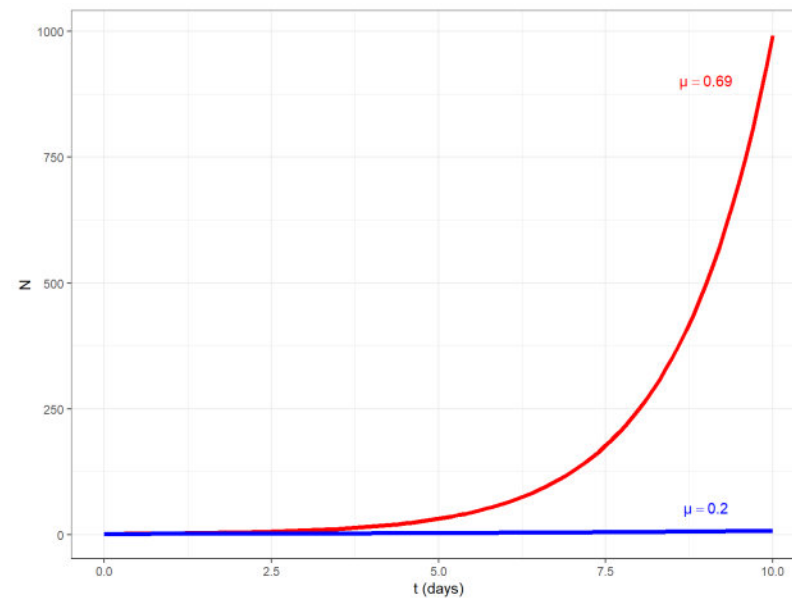
Growth rate vs Loss rate

$$\frac{dN}{dt} = \mu_{net} * N$$

$$N = N_0 \exp^{\mu_{net} * t}$$

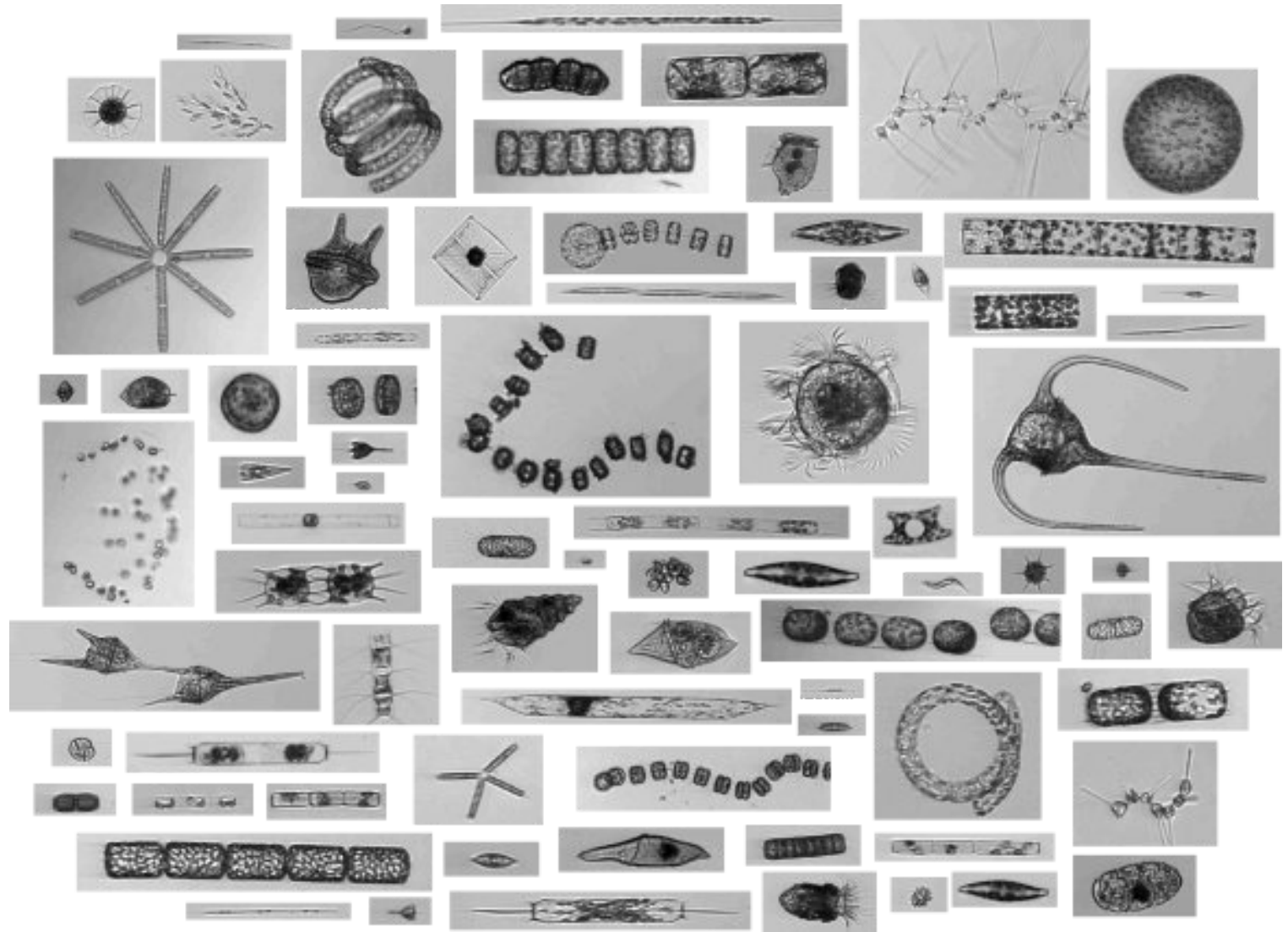
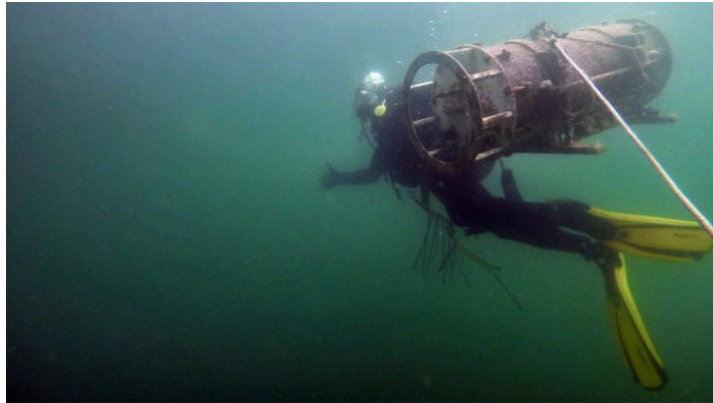
$$\mu_{net} = \mu_{growth} - \mu_{loss}$$

- Growth rate = division
- Loss rate = cell death, predation, viruses



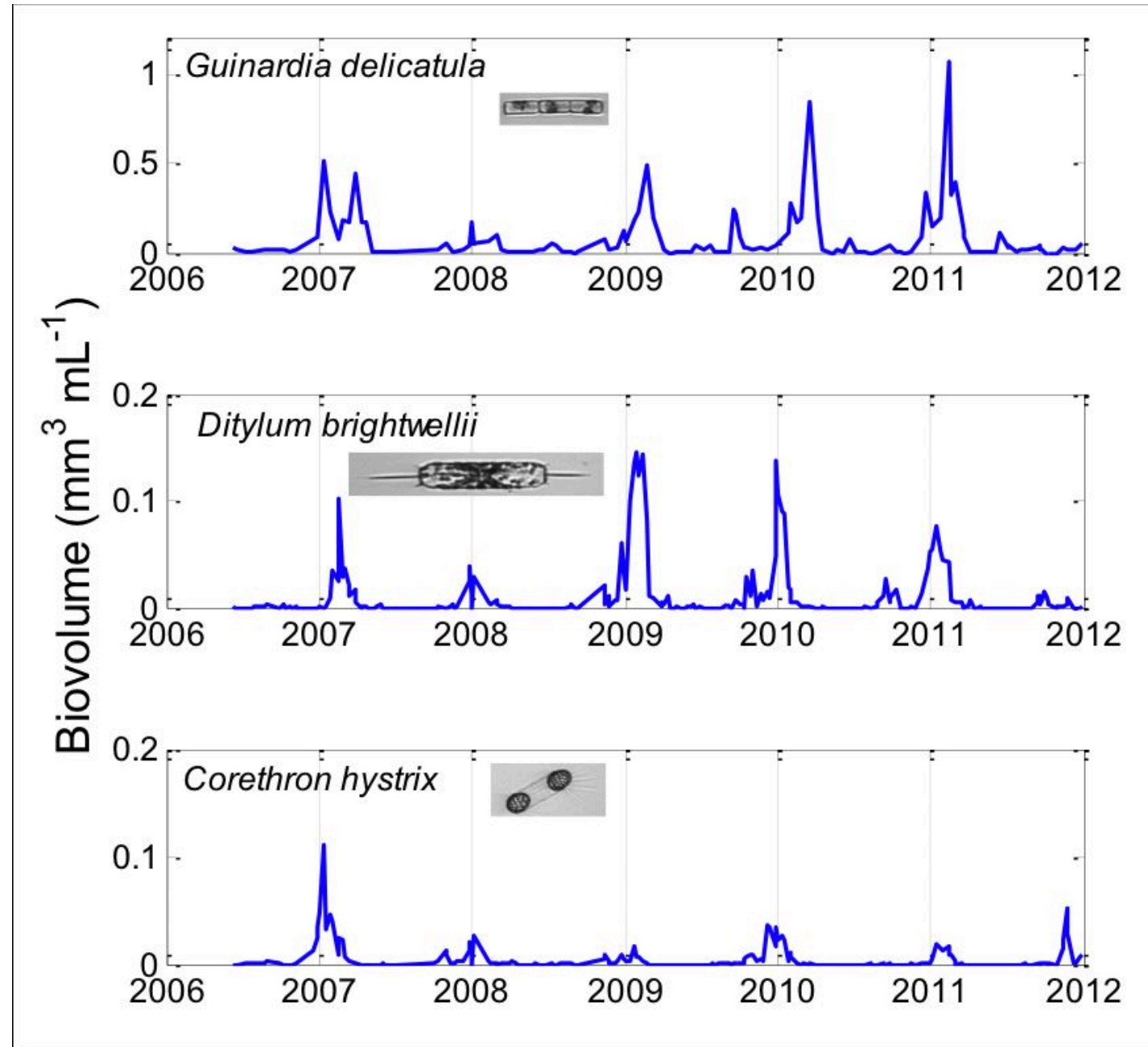
Flow Cytobot

- Imaging and flow cytometry



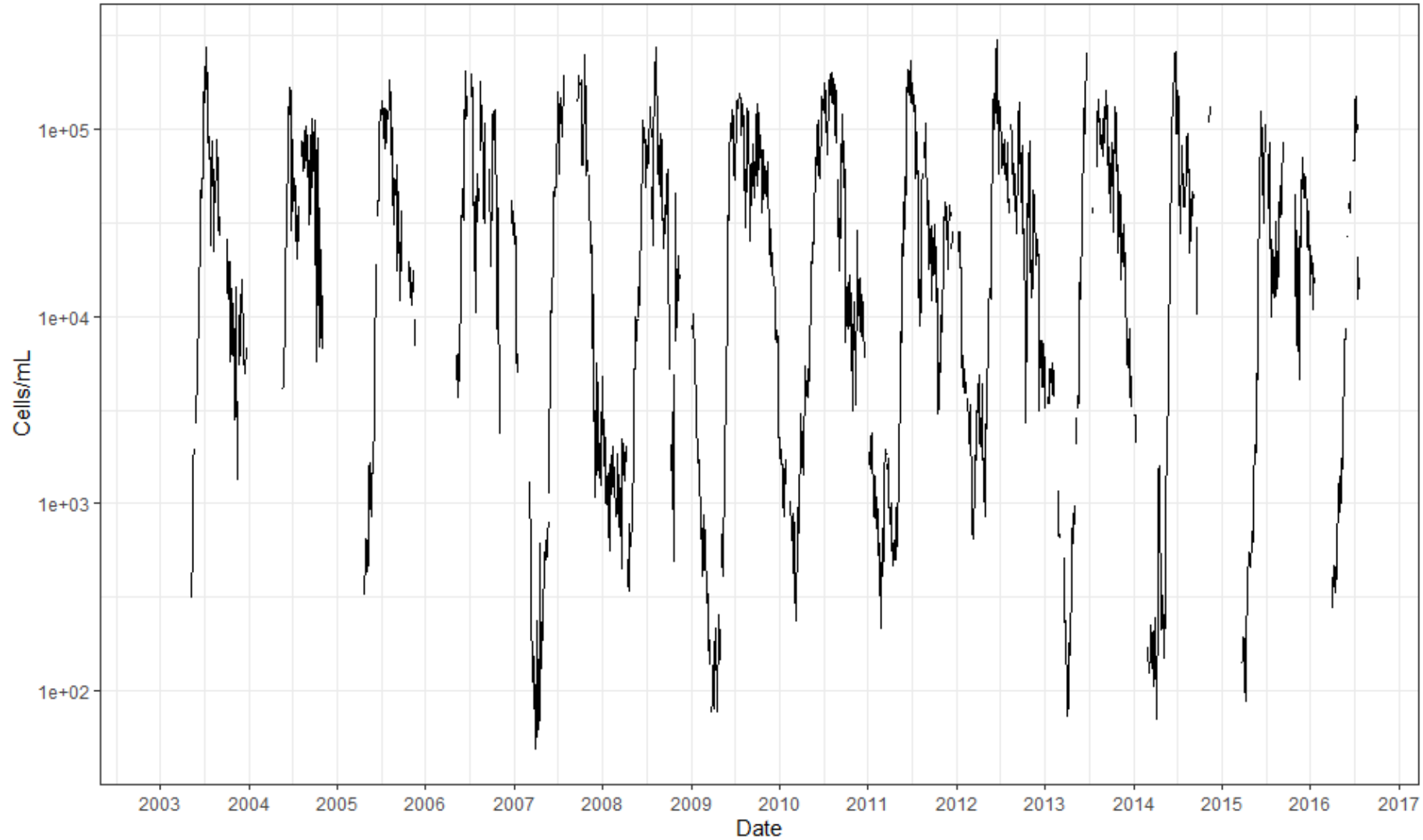
Flow Cytobot

- Diatoms



Synechococcus abundance

- Annual periodicity



Synechococcus characteristics

- Annual periodicity

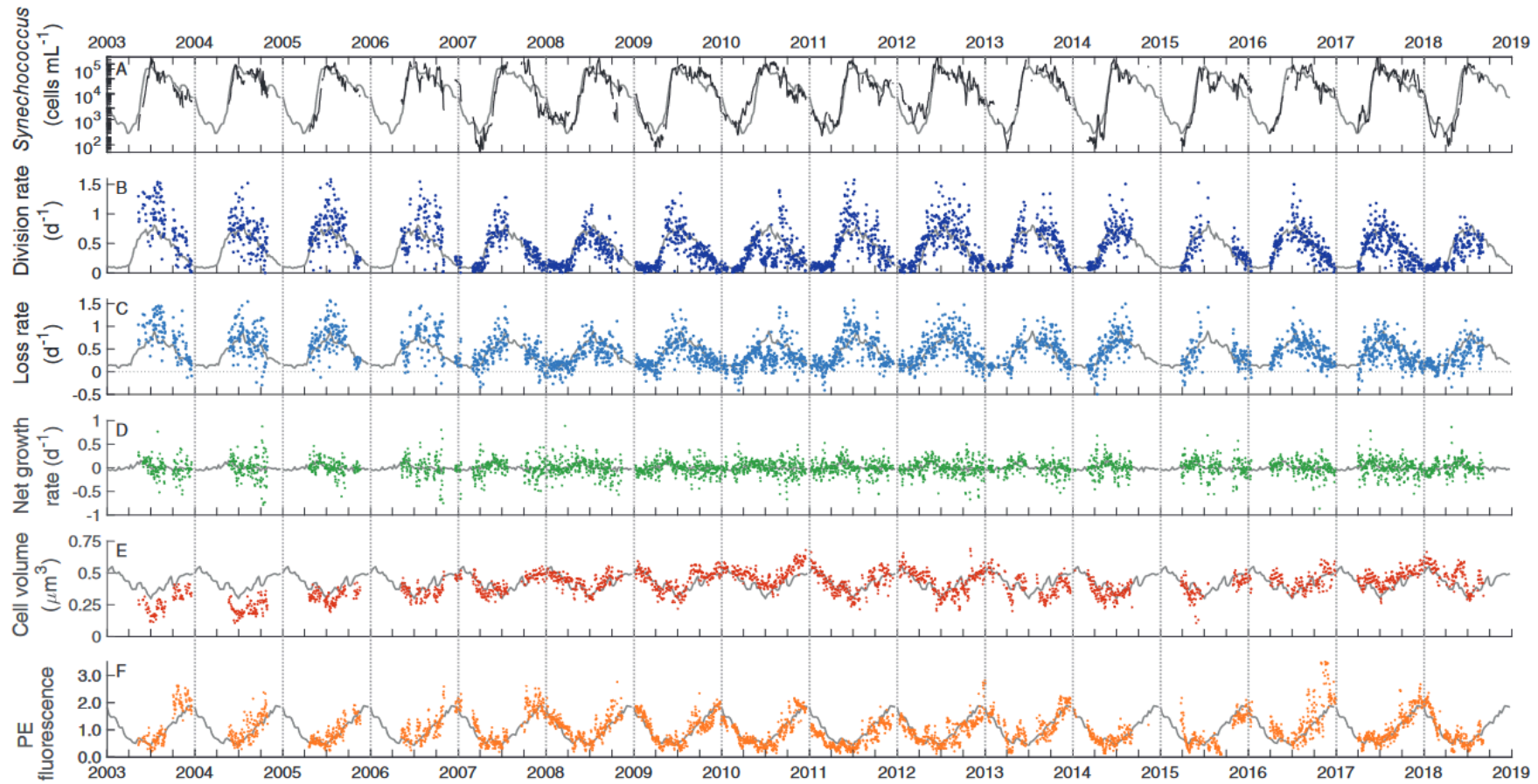


Fig. 4. Daily time series of *Synechococcus* properties for **(A)** cell concentration, log scale, **(B)** daily division rate, **(C)** calculated daily loss rate, **(D)** net growth rate, **(E)** cell volume, and **(F)** cellular PE fluorescence. Gray lines in each panel are annual patterns (weekly median climatology) for reference.

Synechococcus annual cycle

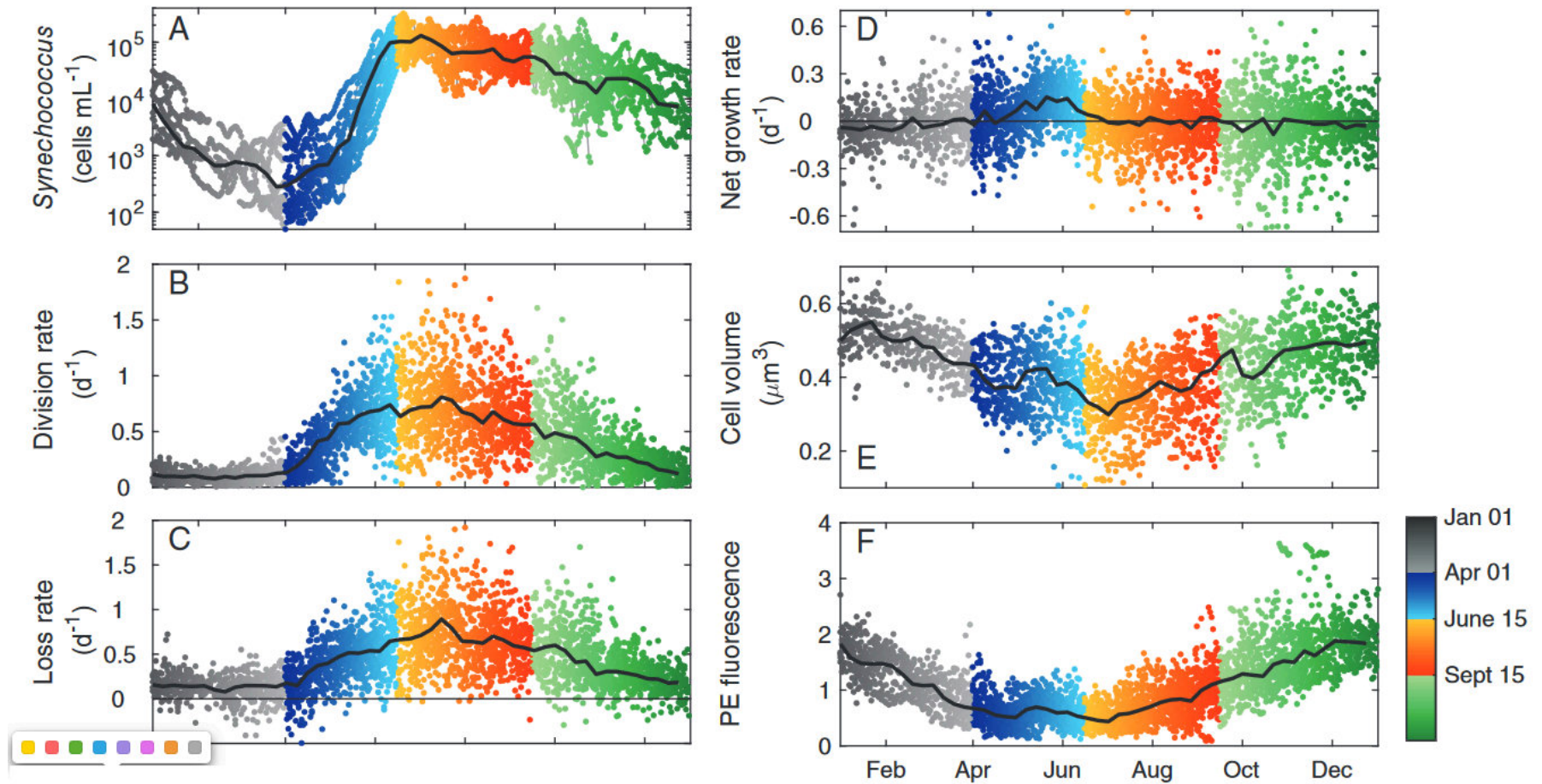
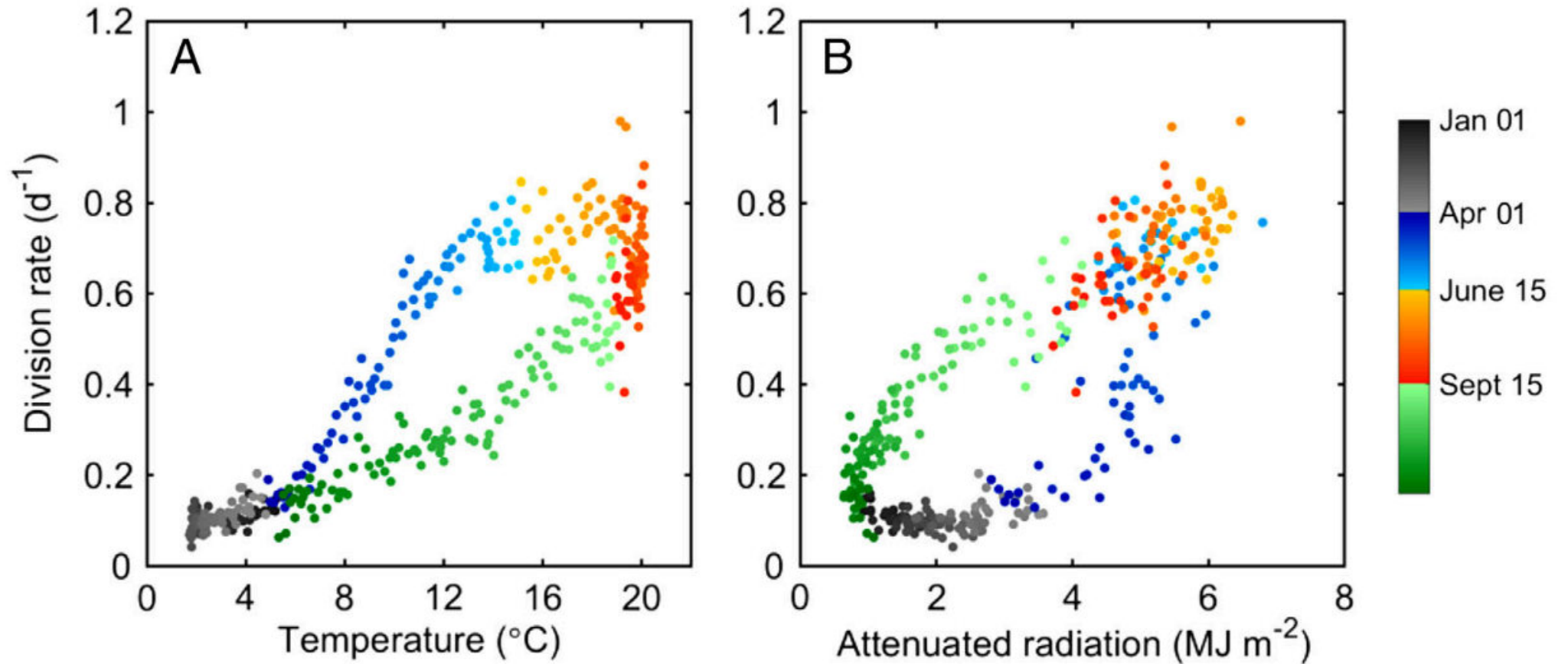


Fig. 5. Scatter plots of (A) *Synechococcus* concentration, (B) division rate, (C) loss rate, (D) net growth rate, (E) cell volume, and (F) cellular PE fluorescence by year day for data from 2003 to 2018. Black line in each panel is weekly median climatology. Color indicates season and year day.

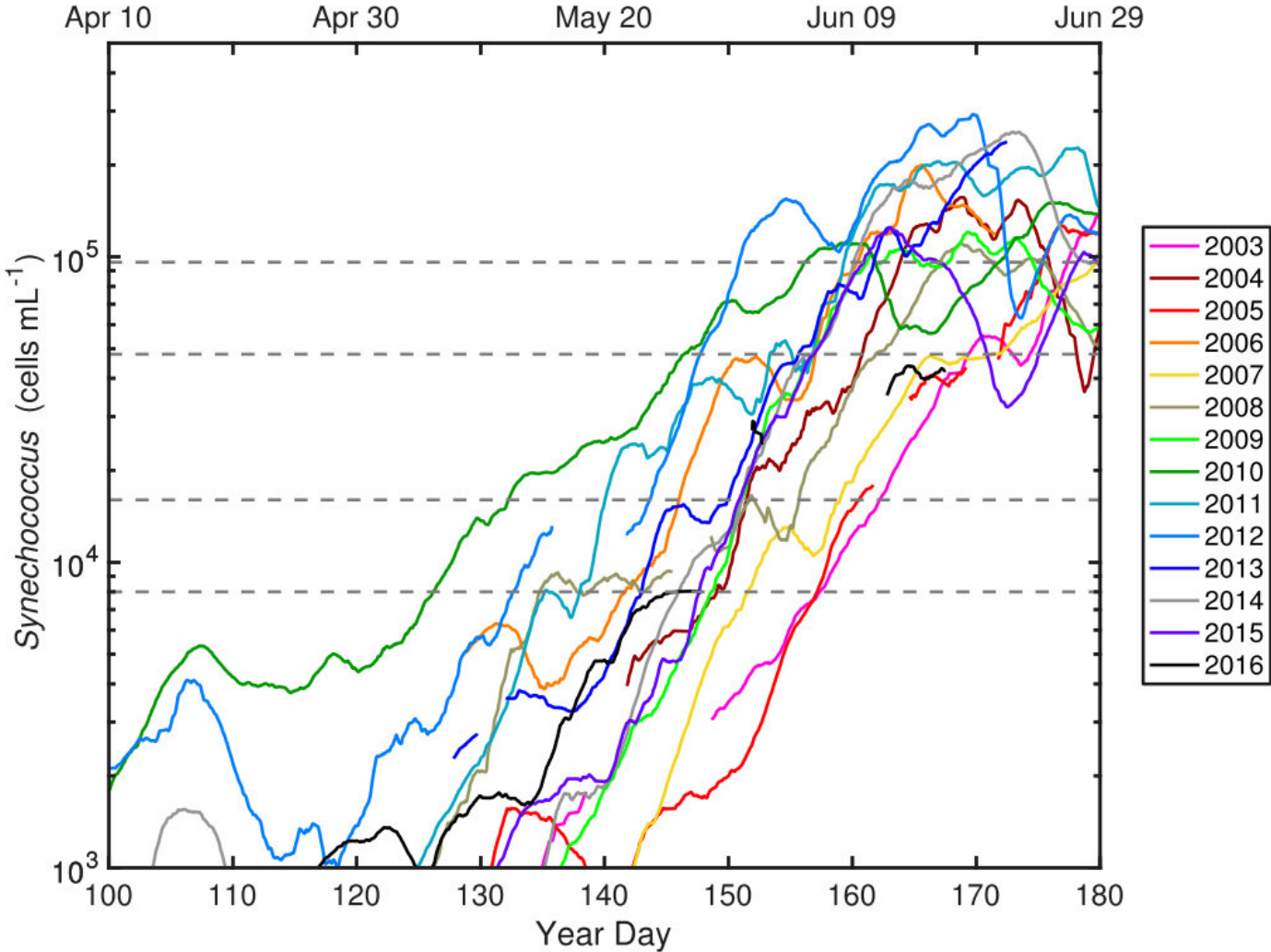
Synechococcus annual cycle

- Division rate
 - Temperature
 - Radiation
- Hysteresis



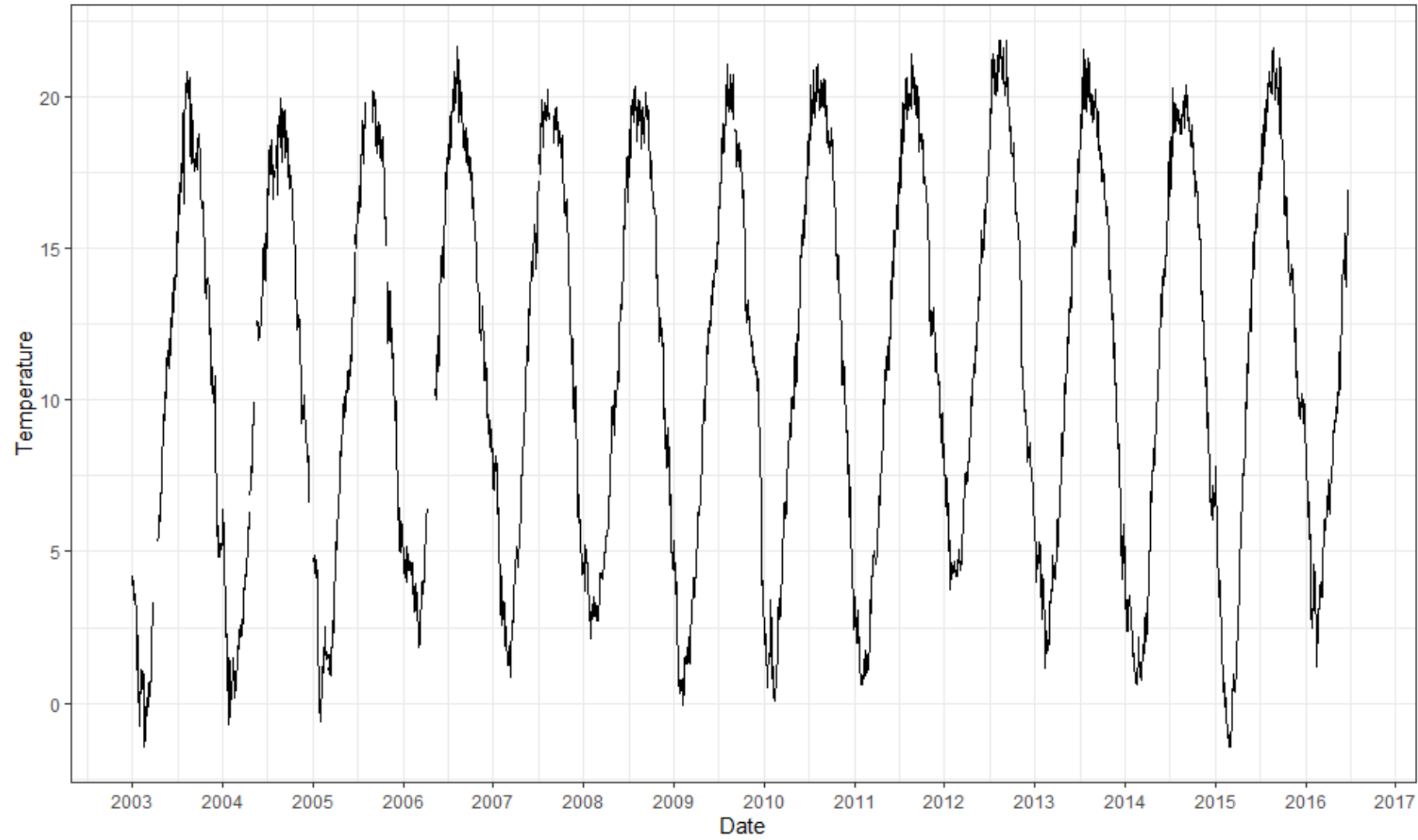
Synechococcus abundance

- Varies from one year to next



Temperature

- Can temperature explain ?



Temperature anomaly

- Some years are warmer than others

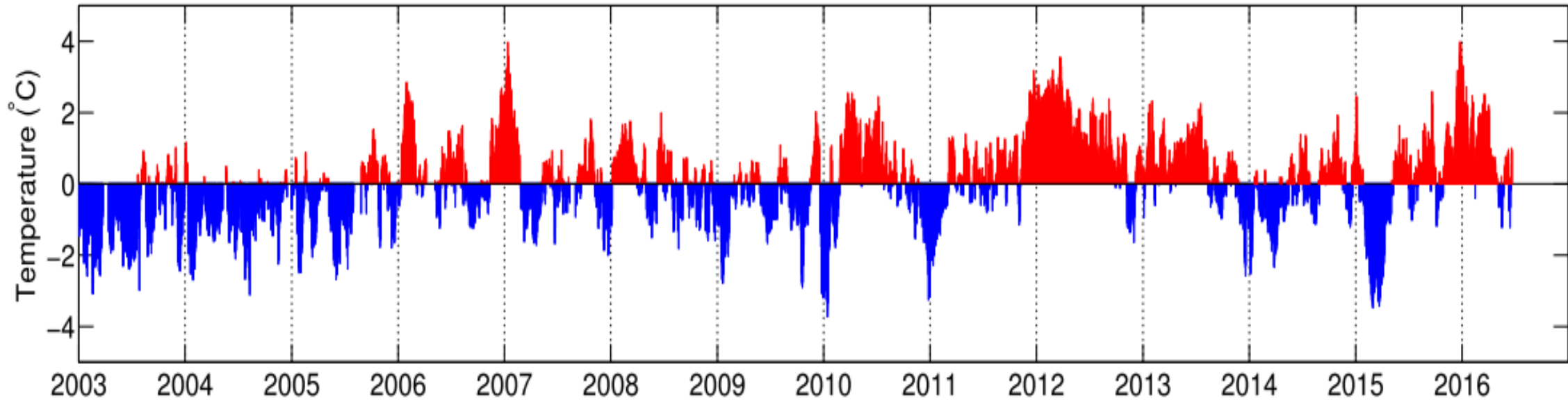


Figure S3. Daily temperature anomalies ($^{\circ}\text{C}$) from daily climatological average for 2003 to available 2016. Red values indicate positive anomalies, while blue indicates negative anomalies.

Synechococcus vs. Temperature

- Day when temperature > threshold
- Matches *Syn* concentration > threshold

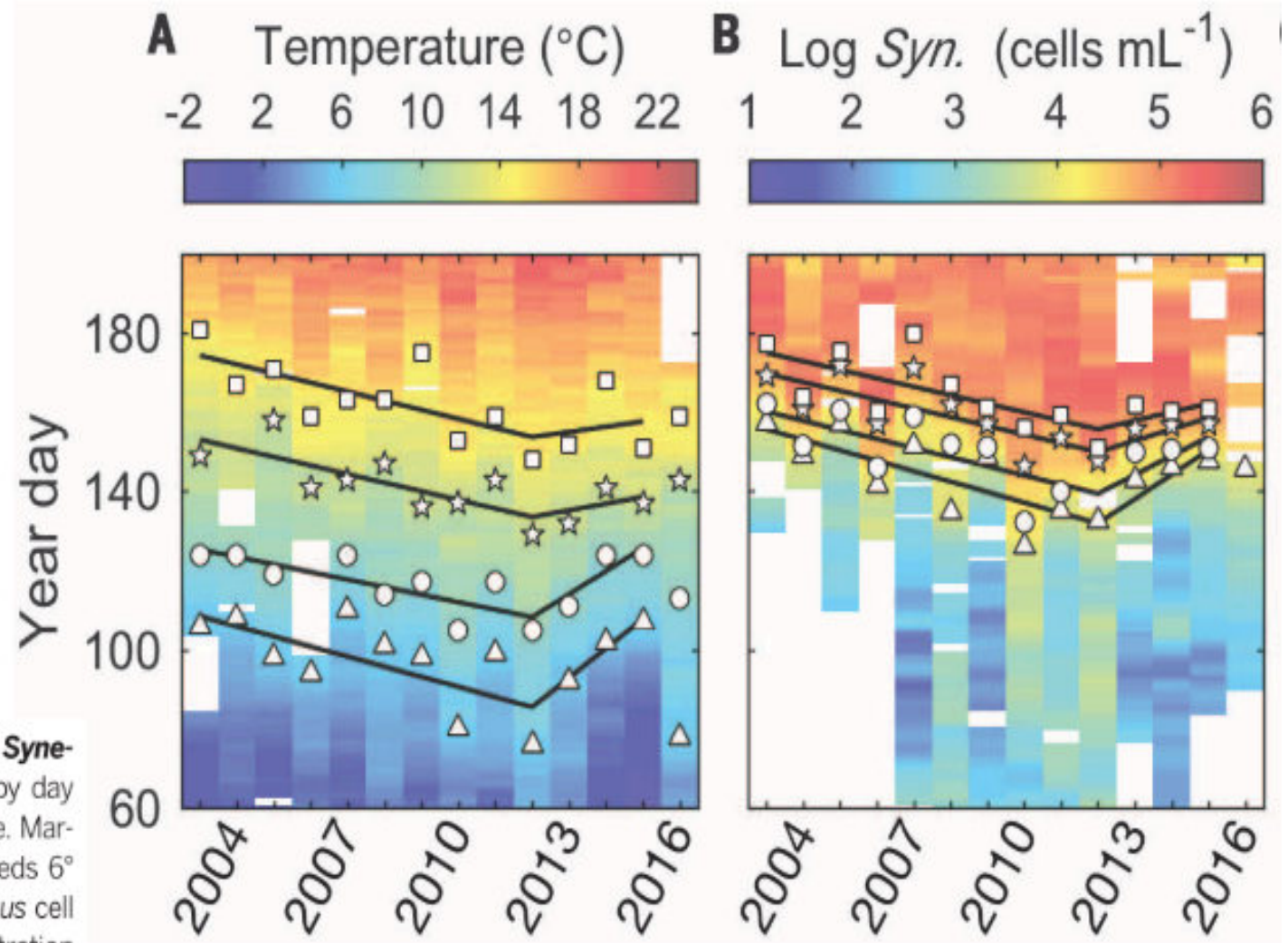


Fig. 2. Multiyear trends showing spring temperature changes and *Synechococcus* bloom shifts from 2003 to 2016. The data are shown by day of the year (vertical axis), with values denoted by color. **(A)** Temperature. Markers indicate the day in each year when water temperature first exceeds 6° (triangles), 9° (circles), 12° (stars), or 15°C (squares). **(B)** *Synechococcus* cell concentration. Markers indicate the day in each year when cell concentration exceeds 8×10^3 (triangles), 1.6×10^4 (circles), 4.8×10^4 (stars), or 9.6×10^4 (squares) cells ml⁻¹. **(C)** Integrated division rate (cumulative summed division

Take home messages

- Long term observation is key to understand:
 - what changes?
 - what drives the change?
- Some examples
 - CPR allowed to detect “Atlantification”
 - Hawaii long term change in Pacific
- More basic research questions
 - Which species re-occur from one year to the next?
 - What causes year-to-year variability for one given species?

Questions ?