Nutrients and flow cytometry from station N-1200 collected in August 2017 from a cruise aboard R/V Mediterranean Explorer

Website: https://www.bco-dmo.org/dataset/874805
Data Type: Cruise Results
Version: 1
Version Date: 2025-07-22

Project
» Microbial ecosystems in silico, in the lab and in the field: understanding interactions between abundant marine bacterial taxa (HADFBA)

Contributors	Affiliation	Role
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Abstract
This dataset contains nutrients and flow cytometry from station N-1200 collected in August 2017 from a cruise aboard R/V Mediterranean Explorer.

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Coverage

Spatial Extent: **Lat**:32.456 **Lon**:34.3745
Temporal Extent: 2017-08-07

Methods & Sampling

Cruises and sample collection
A cruise was carried out on August 7, 2017 on R/V Mediterranean Explorer to study the photic zone in the Eastern Mediterranean at high depth resolution. Genetic data and nano-SIMS measurements were used to infer and quantify mixotrophy by Prochlorococcus at the base of the photic zone.

Water samples were collected using a 12-bottle rosette with 8 L Niskin bottles. Sampling depths were selected based on real-time data from a Conductivity, Temperature, Depth (CTD) profiler (Seabird 19 Plus) from the down-cast before each sample collection in the up-cast. The continuous data were processed using custom Excel files taking into account the location of each sensor and the sensor delay, and binned over 1-meter intervals.

Nutrients
Nutrient samples were collected in acid-washed 500 ml plastic containers and immediately filtered through 0.22 µm Nalgene rapid-flow filter units. The first two filtrates were discarded, and the final sub-samples were collected in new 50 ml falcon tubes and stored in the dark at 4 degrees Celsius in racks. The samples were

transported to the analytical lab at the marine station at Sdot-Yam within 12–15 h of sampling and dissolved nutrients were determined using a SEAL AA-3 autoanalyzer system (SEAL, 2011). To minimize cross-contamination in the laboratory, ammonium was analyzed first alone using an automated ophthalaldehyde (OPA) fluorescence method. Samples were placed in the sample trays and only opened immediately before the sample probe took the sample. The baseline water used for ammonium determination was freshly prepared Milli-Q (MQ) water. The next parameters to be determined were dissolved nitrate and silicate. The methods used were Cd reduction and diazo dye for nitrate & nitrite (hereafter referred to as Nox), and molybdate blue in the presence of oxalic acid for Silicate. The baseline water used for these determinations was also MQ water. Finally, a 3rd run was carried out using an ultra-low level phosphate (DIP) method involving using a 100 cm long flow cell (LWCC) and molybdate blue determination derived from Murphy and Riley (1962). The blank used for phosphate determination was surface seawater, whose DIP value was determined in a previous experiment in which we determined a refractive index correction by removing the ascorbic acid reagent. It was not possible to use MQ as the baseline, as has been used elsewhere because the MQ water in our lab (derived from desalinated water) contains ~20 nM DIP.

Picophytoplankton abundance using flow-cytometry
Water samples (1.5 ml) were collected from each sampling depth, put in cryo-vials (Nunc), and fixed with 7.5 µl 25% glutaraldehyde (Sigma). Vials were incubated in the dark for 10 min, flash-frozen in liquid nitrogen, and stored in a -80 degree C freezer. Before analysis, samples were thawed in the dark at room temperature. Analysis was performed using a BD Canto II flow-cytometer with 2 µm diameter fluorescent beads (Polysciences, Warminster, PA, USA) as a size and fluorescence standard. Three types of phytoplankton cells were identified based on their natural auto-fluorescence: *Prochlorococcus*, *Synechococcus,* and picoeukaryotes. Cells were differentiated based on cell chlorophyll (Ex482nm/Em676nm, PerCP channel) and phycoerythrin fluorescence (Ex564nm/Em574nm), and by the size of the cell (forward scatter). Data were processed using FlowJo software. Flow rates were determined several times during each running session by weighing tubes with double-distilled water, and counts of the standard beads were used to verify a consistent flow rate.

Additional data related to this study: Amplicon sequencing of picoplankton ITS (Internal Transcribed Sequence) are available as NCBI BioProject PRJNA802375. NanoSIMS analyses of flow-corted Prochlorococcus cells are presented in a manuscript by Wu et al (currently under review, doi: 10.1101/2022.01.14.476346).

Data Processing Description

The whole dataset has undergone quality control and has no known errors. Gaps in data were represented by the value 'nd' (no data). In the final data file, missing data values are empty/blank.

BCO-DMO Processing Description

BCO-DMO Processing:
- Imported original file "High-resolution_data_for_BCO-DMO_030222.xlsx" into the BCO-DMO system.
- Marked "nd" as a missing data value (missing data are empty/blank in the final CSV file).
- Renamed fields to comply with BCO-DMO naming conventions.
- Added columns for Latitude, Longitude, and Date.
- Saved the final file as "874805_v1_flowcyt.csv".

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Data Files

File
874805_v1_flowcyt.csv (Comma Separated Values (.csv), 769 bytes) MD5:745bfd97cbc1241c6833eb1e7ce70ca8
Primary data file for dataset ID 874805, version 1

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Related Publications

Ben Ezra, T., Krom, M. D., Tsemel, A., Berman-Frank, I., Herut, B., Lehahn, Y., Rahav, E., Reich, T., Thingstad, T. F., & Sher, D. (2021). Seasonal nutrient dynamics in the P depleted Eastern Mediterranean Sea. Deep Sea Research Part I: Oceanographic Research Papers, 176, 103607. https://doi.org/10.1016/j.dsr.2021.103607
Methods

Murphy, J., & Riley, J. P. (1962). A modified single solution method for the determination of phosphate in natural waters. Analytica Chimica Acta, 27, 31–36. doi:10.1016/s0003-2670(00)88444-5
Methods

Wu, Z., Aharonovich, D., Roth-Rosenberg, D., Weissberg, O., Luzzatto-Knaan, T., Vogts, A., Zoccarato, L., Eigemann, F., Grossart, H.-P., Voss, M., Follows, M. J., & Sher, D. (2022). Significant organic carbon acquisition by Prochlorococcus in the oceans. https://doi.org/10.1101/2022.01.14.476346
Results

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Related Datasets

IsRelatedTo

Sher, D. (2025) **CTD data from station N-1200 collected in August 2017 from a cruise aboard R/V Mediterranean Explorer.** Biological and Chemical Oceanography Data Management Office (BCO-DMO). (Version 1) Version Date 2025-07-22 doi:10.26008/1912/bco-dmo.874728.1 [view at BCO-DMO]

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Parameters

Parameter	Description	Units
Depth	Depth	meters (m)
P	Soluble reactive phosphorus (DIP)	nanomolar (nM)
TOxN	Total nitrate + nitrite	nanomolar (nM)
Si	Dissolved silicate	nanomolar (nM)
NH4	Dissolved ammonium	nanomolar (nM)
Prochlorococcus	Cell counts from flow cytometry, average of duplicates from two separate casts	cells per mL
Synechococcus	Cell counts from flow cytometry, average of duplicates from two separate casts	cells per mL
Pico_eukaryotes	Cell counts from flow cytometry, average of duplicates from two separate casts	cells per mL
Latitude	Latitude of sampling location	degrees North
Longitude	Longitude of sampling location	degrees East
Date	Sample date in format YYYY-MM-DD	unitless

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Instruments

Dataset-specific Instrument Name	Seabird 19 Plus
Generic Instrument Name	CTD Sea-Bird
Generic Instrument Description	A Conductivity, Temperature, Depth (CTD) sensor package from SeaBird Electronics. This instrument designation is used when specific make and model are not known or when a more specific term is not available in the BCO-DMO vocabulary. Refer to the dataset-specific metadata for more information about the specific CTD used. More information from: http://www.seabird.com/

Dataset-specific Instrument Name	BD Canto II flow-cytometer
Generic Instrument Name	Flow Cytometer
Generic Instrument Description	Flow cytometers (FC or FCM) are automated instruments that quantitate properties of single cells, one cell at a time. They can measure cell size, cell granularity, the amounts of cell components such as total DNA, newly synthesized DNA, gene expression as the amount messenger RNA for a particular gene, amounts of specific surface receptors, amounts of intracellular proteins, or transient signalling events in living cells. (from: http://www.bio.umass.edu/micro/immunology/facs542/facswhat.htm)

Dataset-specific Instrument Name	8 L Niskin bottles
Generic Instrument Name	Niskin bottle
Generic Instrument Description	A Niskin bottle (a next generation water sampler based on the Nansen bottle) is a cylindrical, non-metallic water collection device with stoppers at both ends. The bottles can be attached individually on a hydrowire or deployed in 12, 24, or 36 bottle Rosette systems mounted on a frame and combined with a CTD. Niskin bottles are used to collect discrete water samples for a range of measurements including pigments, nutrients, plankton, etc.

Dataset-specific Instrument Name	SEAL AA-3 autoanalyzer system
Generic Instrument Name	Nutrient Autoanalyzer
Generic Instrument Description	Nutrient Autoanalyzer is a generic term used when specific type, make and model were not specified. In general, a Nutrient Autoanalyzer is an automated flow-thru system for doing nutrient analysis (nitrate, ammonium, orthophosphate, and silicate) on seawater samples.

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Project Information

Microbial ecosystems in silico, in the lab and in the field: understanding interactions between abundant marine bacterial taxa (HADFBA)

Coverage: Eastern Mediterranean Sea

Every drop of seawater contains around one million microorganisms (bacteria, small algae and other organisms such as ciliates and diatoms). These marine microbes feed the entire marine ecosystem, modulate global cycles of carbon and other elements, and impact climate. With the advances in genome-sequencing technology, we can now identify the microbes and assess their genetic and metabolic capacities, yet we still cannot deduce from the genomes of these organisms how they will grow - and interact - in nature. The proposed project will tackle this challenge through a tightly integrated combination of mathematical modeling, laboratory experiments and field work in the Eastern Mediterranean, to identify genes and pathways dictating how environmentally-relevant microbes grow and interact in the sea. We will produce genome-scale

mathematical models of the metabolism of *Prochlorococus*, the numerically-dominant photosynthetic bacteria in large swaths of the ocean, and of *Alteromonas*, abundant marine bacteria which make their living by consuming and respiring organic molecules produced by *Prochlorococcus* and other photosynthetic microbes. We will test these models using laboratory cultures of these organisms grown alone and together, and determine to what extent the models and laboratory cultures represent the growth and death of these organisms in the Eastern Mediterranean. This study will be useful for scientists of many disciplines, including not only marine biology, oceanography and ecology but also genetics, medicine and agriculture. Our results will shed light on the dynamics of some of the most common organisms in the world, responsible for the production of up to 20% of the oxygen we breathe. Our collaborative study will foster the development and training of the next generation of marine scientists, and will be used in outreach activities designed to share with high-school students and the general public the excitement of marine research and the need to responsibly utilize and sustain the oceans for the sake of future generations.

The proposed project will tackle the challenge of understanding microbial interactions from the underlying genetic data through a tightly integrated combination of genome scale modeling, laboratory experiments and field work in the Eastern Mediterranean. We aim to identify genomic traits dictating how environmentally-relevant primary producers and heterotrophic bacteria interact. Genome-scale (dynamic flux balance analysis, dFBA) models of *Prochlorococus* MED4 and of *Alteromonas* HOT1A3 will be produced and calibrated using high-throughput measurements of growth and physiological parameters in laboratory batch cultures, combined with detailed analysis of specific metabolites; The dFBA models will be combined *in-silico* and the results compared to laboratory co-cultures. Model-data discrepancies will provide opportunities to revisit the models, suggesting the mediation of alternative processes such as allelopathy or other types of chemical signaling. Finally, time-series data on the community composition and function during the summer/fall *Prochlorococcus* bloom in the hyper-oligotrophic Eastern Mediterranean, combined with field experiments (microcosms), will provide a test of hypotheses generated in the lab. This study will provide the first detailed "roadmap" linking genomic traits (genes and metabolic pathways) and rate measurements with species interactions in environmentally-relevant marine microbes. Genome-scale models will likely be embedded in a not-so-distant future in global-scale models of the Earth System, and the proposed study will provide a critical stepping-stone towards predicting how marine microbial systems will evolve in a changing world. The strong human impact on marine ecosystems, and the need for quantitative and predictive understanding of how they will respond to a changing environment, calls for interdisciplinary research and training for the next generation of scientists and decision makers. Models and data generated by our work will be integrated into a novel educational exploration-focused, web- and field-based educational module. This module will introduce key concepts in microbiology, environmental sciences and oceanography to intermediate- and high-school students.

(Note: acronym HADFBA = Heterotroph-Autotroph Dynamic Flux Balance Analysis)

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Funding

Funding Source	Award
NSF Division of Ocean Sciences (NSF OCE)	OCE-1635070

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