Stoichiometric traits and environmental parameters from surface samples collected on GOSHIP expedition I08S in the Southern Indian Ocean from February 21 to April 1, 2024

Website: https://www.bco-dmo.org/dataset/1002701
Data Type: Cruise Results
Version: 1
Version Date: 2026-07-24

Project
» Collaborative Research: The stoichiometric trait distribution of the marine microbiome (StoichTraitD)

Contributors	Affiliation	Role
Hall, Edward	Colorado State University (CSU)	Principal Investigator
Sahoo, Deepika	Colorado State University (CSU)	Scientist
Rauch, Shannon	Woods Hole Oceanographic Institution (WHOI BCO-DMO)	BCO-DMO Data Manager

Abstract
This dataset contains analyses collected from a broad latitudinal and environmental gradient in the southern Indian Ocean. Energy Dispersive Spectroscopy (EDS) was used to analyze biomass C:N:P of individual microbes and probability density function was applied to create distributions for each stoichiometric trait (C:N, N:P, and C:P). This dataset describes the richness, evenness, and divergence from each trait distribution. Each individual cell value is accompanied by cell area and the environmental parameters from the stations where the cells were collected. This is the first study to investigate how the environment acts on individual microbes to shape the stoichiometric trait distribution of the open ocean microbiome.

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Coverage

Location: Southern Indian Ocean
Spatial Extent: **N**:-28.3181 **E**:95.008 **S**:-67.1141 **W**:76.6563
Temporal Extent: 2024-03-04 - 2024-03-28

Methods & Sampling

At each of the 66 stations, we sampled surface water (~5 meters (m)) using a (SIO) STS 36-place yellow rosette sampler. We prepared samples for single-cell trait analyses in clean conditions by resting 3 to 5 drops (~10 microliters (µL)) of seawater on SF200 Ni grids (Cat: SF200-NI, Electron Microscopy Sciences) using a micropipette. The grids were air-dried for approximately 50 seconds between each drop of seawater. We removed the excess water using a tapered rolled corner of fresh Kimwipe, to avoid excess accumulation of salt particles on the grid. The process was repeated 3 to 5 times for each grid. In total, 6 to 10 grids were prepared for the surface water samples at each station. The samples were analyzed onshore in the Analytical Research Core's (ARC) electron microscope facility at Colorado State University, Fort Collins. We measured the

dissolved nutrients, including ammonium, nitrite, nitrate, phosphate, and silicate onboard using a Seal Analytical continuous-flow AutoAnalyzer 3. Temperature, salinity, and fluorescence (chlorophyll) data were obtained from the sensors mounted on the CTD rosette sampler.

Data Processing Description

We exported the spectral output generated by the Oxford EDAX system as an '.emsa' file type and processed the spectra using the freely available NIST-based DTSA II (Desktop Spectral Analyzer) software program (Jupiter release available at: http://www.cstl.nist.gov/div837/837.02/epq/dtsa2/). After stripping the background radiation from each spectrum, we determined the total counts within the pre-selected ranges using the DTSA software. The background-stripped counts in each characteristic peak were generated for both the sample and the film, and the film signal was subsequently subtracted from the sample signal. The elemental content in counts per second was further converted to cellular elemental content in femtomoles using the C, N, and P calibration factors. From these values, we calculated the individual traits (i.e., C:N, N:P, and C:P) for each microbe. We analyzed between 24 and 88 individual cells per station. The calibration factors for C, N, and P were 13.64, 10.52, and 49.81 counts per second per femtomole, respectively. We used the probability density function to characterize the trait distributions using the "ggridges" package in RStudio (2025.05.1). Additionally, we computed the functional richness, functional evenness, and functional divergence of the trait distributions using the TPD package in RStudio. We selected an alpha value of 0.95 for each trait to make the analyses less sensitive to outliers. We also used ImageJ software to measure the cell area of microbes. It is free software available at https://imagej.net/ij/download.html. In ImageJ, we processed the SEM images of microbes, where the software automatically outlined the cells and calculated their corresponding area in square micrometers (μm^2).

BCO-DMO Processing Description

- Imported the sample log from the original file "I08S_EDS_Sample_Log_DS_4302026.csv" as table "sample_log" in the BCO-DMO data processing system.
- Imported the primary cell-level data from the original file "IO8S_cell_info_with_environmental_DS_4302026_bco.csv" as table "cell_info" in the BCO-DMO data processing system.
- Applied find/replace on "Sample Id" in cell_info and "Sample" in sample_log to remove space-underscore patterns (" " → "") and normalize Sample IDs in both tables.
- Stripped "_Box 1" or "_Box 2" suffixes from "Sample" column in sample_log to normalize Sample IDs in both tables.
- Renamed columns in sample_log: Sample → Sample_Id, Date_UTC (mm/dd/yyyy hh:mm) → DateTime_UTC, Time_UTC (hh:mm) → Time_UTC, Lat → Latitude_sample_log, Long → Longitude.
- Renamed "Sample Id" → "Sample_Id" in cell_info to align join keys.
- Performed half-outer join of sample_log into cell_info on compound key {Sample_Id}{Niskin_num} (source) / {Sample_Id}{Niskin number}, bringing in Cruise_ID, DateTime_UTC, EXPOCODE, Grid_Locations, Grid_location_box_num, Latitude_sample_log, Longitude, Niskin_num, Sample_descrip, Time_UTC, WOCE_LINE from the sample_log.
- Converted DateTime_UTC (format "%m/%d/%Y %H:%M", UTC) to ISO 8601 datetime column "ISO_DateTime_UTC" (format "%Y-%m-%dT%H:%MZ").
- Deleted the original DateTime_UTC and Time_UTC columns.
- Multiplied Latitude_sample_log by "-1" to correct sign convention for Southern Hemisphere.
- Multiplied Latitude (from cell_info) by "-1" to correct sign convention for Southern Hemisphere; renamed to "Latitude_cell_info".
- Renamed columns to comply with BCO-DMO naming conventions.
- For rows where Sample_Id "S7_2.8 m", replaced Grid_Locations value "A1, A3, A5" with "Box 1: J4, J6, J8; Box 2: A1, A3, A5".
- For rows where Sample_Id "S7_2.8 m", replaced Grid_location_box_num value "2" with "Box 1: 1; Box 2: 2".
- Renamed table "cell_info" to "1002701_v1_io8s_cell_info".
- Filled all EXPOCODE values with "325020240221".
- Reordered columns to final order.
- Saved the final file as "1002701_v1_io8s_cell_info.csv".

- Imported original file "IO8S_stats.csv" into the BCO-DMO data processing system.
- Renamed columns to comply with BCO-DMO naming conventions.

- Saved the final file as "1002701_v1_supplemental_io8s_stats.csv".

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Related Publications

Becker, S., Aoyama, M., Woodward, E. M. S., Bakker, K., Coverly, S., Mahaffey, C., & Tanhua, T. (2020). GO-SHIP Repeat Hydrography Nutrient Manual: The Precise and Accurate Determination of Dissolved Inorganic Nutrients in Seawater, Using Continuous Flow Analysis Methods. Frontiers in Marine Science, 7. https://doi.org/10.3389/fmars.2020.581790
Methods

Carmona, C. P. (2019). TPD: Methods for Measuring Functional Diversity Based on Trait Probability Density. R package version 1.1.0. https://CRAN.R-project.org/package=TPD
Software

Carmona, C. P., de Bello, F., Mason, N. W. H., & Lepš, J. (2016). Traits Without Borders: Integrating Functional Diversity Across Scales. Trends in Ecology & Evolution, 31(5), 382–394. https://doi.org/10.1016/j.tree.2016.02.003
Methods

Manzella, M., Geiss, R., & Hall, E. K. (2019). Evaluating the stoichiometric trait distributions of cultured bacterial populations and uncultured microbial communities. Environmental Microbiology, 21(10), 3613–3626. Portico. https://doi.org/10.1111/1462-2920.14684
Methods

National Institute of Standards and Technology (NIST). NIST DTSA-II public domain software, version Jupiter. Gaithersburg, MD: National Institute of Standards and Technology. http://www.cstl.nist.gov/div837/837.02/epq/dtsa2/
Software

Posit team (2024). RStudio: Integrated Development Environment for R. Posit Software, PBC, Boston, MA. http://www.posit.co/
Software

Rasband, W. S. (n.d.). ImageJ. U.S. National Institutes of Health. https://imagej.net/ij/
Software

Wilke, C. (2025). ggridges: Ridgeline Plots in 'ggplot2'. R package version 0.5.7. https://wilkelab.org/ggridges/
Software

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Parameters

Parameter	Description	Units
WOCE_LINE	Cruise expedition track in the southern Indian Ocean	unitless
Cruise_ID	Unique identification of an expedition	unitless
EXPOCODE	Expedition code	unitless
Station	Location from where seawater was collected	unitless
Depth	Depth	meters (m)

Niskin_number	Number of niskin bottle carrying the seawater	unitless
Sample_Id	Sample Id (station + depth)	unitless
ISO_DateTime_UTC	Date and time (UTC) in ISO 8601 format	unitless
Latitude_cell_info	Latitude in degrees North (negative values = South); from original file "IO8S_cell_info_with_environmental_DS_4302026.csv"	degrees
Latitude_sample_log	Latitude in degrees North (negative values = South); from original file "I08S_EDS_Sample_Log_DS_4302026.csv"	degrees
Longitude	Longitude in degrees East (positive values = East); from original file "I08S_EDS_Sample_Log_DS_4302026.csv"	degrees
Cell_num	Cell number (sample ID + cell sample number)	unitless
C_N	carbon nitrogen ratio	unitless
N_P	nitrogen phosphorus ratio	unitless
C_P	carbon phosphorus ratio	unitless
Area	cell area	square microns
Temperature	Temperature	degrees Celsius
Salinity	Salinity	unitless
Silicate	Silicate	micromolar
Dissolved_Iron	dissolved iron	nanomolar
Ammonium	ammonium	micromolar
Nitrate	nitrate	micromolar
Nitrite	nitrite	micromolar

Nitrate_Nitrite	Nitrate and Nitrite	micromolar
Phosphate	phosphate	micromolar
Chlorophyll	Chl a	milligrams per cubic meter (mg m-3)
Grid_location_box_num	Grid Box number	unitless
Grid_Locations	Location of grid in grid box	unitless

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Instruments

Dataset-specific Instrument Name	Seal Analytical continuous-flow AutoAnalyzer 3
Generic Instrument Name	Bran+Luebbe / SEAL Analytical AutoAnalyzer 3 (AA3) continuous-flow analyzer
Dataset-specific Description	Measured the dissolved nutrients, including ammonium, nitrite, nitrate, phosphate, and silicate
Generic Instrument Description	The AutoAnalyzer 3 (AA3) is a segmented continuous-flow analyzer (continuous flow analyzer, CFA) used for automated colorimetric analysis of dissolved nutrients and other analytes in environmental, seawater, freshwater, wastewater, soil, and agricultural samples. The AA3 was originally manufactured by Bran+Luebbe and, following acquisition of the product line in 2006, has continued to be manufactured and supported by SEAL Analytical. The AA3 is the third-generation instrument in the Technicon AutoAnalyzer family and is widely used for determination of nitrate, nitrite, ammonium, phosphate, silicate, and other dissolved nutrients. See the description from the manufacturer.

Dataset-specific Instrument Name	SIO STS 36-place yellow rosette sampler
Generic Instrument Name	CTD - profiler
Dataset-specific Description	Temperature, salinity, and fluorescence (chlorophyll) data were obtained from the sensors mounted on the CTD rosette sampler
Generic Instrument Description	The Conductivity, Temperature, Depth (CTD) unit is an integrated instrument package designed to measure the conductivity, temperature, and pressure (depth) of the water column. The instrument is lowered via cable through the water column. It permits scientists to observe the physical properties in real-time via a conducting cable, which is typically connected to a CTD to a deck unit and computer on a ship. The CTD is often configured with additional optional sensors including fluorometers, transmissometers and/or radiometers. It is often combined with a Rosette of water sampling bottles (e.g. Niskin, GO-FLO) for collecting discrete water samples during the cast. This term applies to profiling CTDs. For fixed CTDs, see https://www.bco-dmo.org/instrument/869934.

Dataset-specific Instrument Name	JEOL JSM-6500F SEM
Generic Instrument Name	Scanning Electron Microscope
Dataset-specific Description	a JEOL JSM-6500F SEM equipped with an 80 mm2 Oxford X-max EDS detector
Generic Instrument Description	A scanning electron microscope (SEM) scans a focused electron beam over a surface to create an image. The electrons in the beam interact with the sample, producing various signals that can be used to obtain information about the surface topography and composition.

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Deployments

TN430

Website	https://www.bco-dmo.org/deployment/1002714
Platform	R/V Thomas G. Thompson
Start Date	2024-02-21
End Date	2024-04-01
Description	See additional information at R2R: https://www.rvdata.us/search/cruise/TN430

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Project Information

Collaborative Research: The stoichiometric trait distribution of the marine microbiome (StoichTraitD)

Coverage: Indian Ocean; Laboratory Ecophysiology

NSF Award Abstract:
The elemental ratios of carbon, nitrogen, and phosphorus (C:N:P) have been considered fixed proportions in marine environments but recent work has demonstrated changes across latitudes. Such variation can have large implications for marine biogeochemistry and atmospheric CO_2 levels. As such, future variations in ocean community C:N:P could be a key feedback to global change. The elemental composition of particulate organic matter (POM) represents the aggregate value of diverse microorganisms as well as non-living particles. However, we currently do not understand how individual cells and particles contribute to observed variation in the C:N:P of POM. This project is determining the biomass C:N:P of individual microbial cells grown under a range of conditions and sampled from diverse ocean biomes. Because different individuals are likely to have different fates (e.g., loss to sinking, lysis, predation), understanding how the trait distribution of microbial biomass C:N:P relates to cell size and trophic mode and how environmental conditions affect each trait's distribution offers new perspectives and insight into marine C, N, and P cycles. This project supports two PhD students and multiple undergraduate students. The PhD students are integrated into existing networks on microbiome science at each institution with opportunities to collaborate across diverse disciplines. Undergraduate students at both institutions are being recruited through existing training programs that target underrepresented groups. In addition, PI Hall is part of a collaborative of Ecologists and Poets at CSU, that look at ways to translate ecological relationships to non-traditional media to make it more accessible and impactful to the general public. This group is exploring the nature of individuality within the marine microbiome by creating trait distributions of written text and removing different modes of individuals (e.g., words) to better understand and communicate how individuals from the smallest organisms on the planet (marine microorganisms) can have large effects on the surrounding ecosystem (i.e., the planet). Results from this project are being incorporated into future projects of the working group at Colorado State University including public presentations, art installations, and published materials.

The aim of this project is to quantify the relationship between environmental conditions and marine microbial C:N:P by assessing the individuality in microbial elemental stoichiometry. To achieve this the project uses energy dispersive spectroscopy (EDS) to assess the stoichiometric trait distribution of populations and communities under different resource and temperature treatments and oceanographic field work across a broad latitudinal gradient. The researchers hypothesize that the relationship between the stoichiometry of particulate organic matter (POM) and environmental conditions are masked by distinct responses of individual constituents of the marine microbiome. They hypothesize that these distinct responses result in a multi-modal distribution of particulate carbon (C), nitrogen (N), and phosphorus (P) of the marine microbiome. The investigators are characterizing the distribution of three stoichiometric traits (biomass C:N, C:P, and N:P) for 50 marine isolates (both autotrophs and heterotrophs) under different resource and temperature environments. They are characterizing the same trait distribution for marine communities sampled from different regions of the ocean for the same resource and temperature environments as the population experiments. They are participating on an ocean going cruise to characterize stoichiometric trait distribution of the marine microbiome across natural gradients of resources and temperature. Understanding how constituent members of microbial communities alter their biomass in response to environmental change is providing a missing link between the variation in the ocean's environment and particulate C:N:P ratios for diverse marine environments.

This award reflects NSF's statutory mission and has been deemed worthy of support through evaluation using the Foundation's intellectual merit and broader impacts review criteria.

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Funding

Funding Source	Award
NSF Division of Ocean Sciences (NSF OCE)	OCE-2134950
NSF Division of Ocean Sciences (NSF OCE)	OCE-2135035

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