Chemoautotrophic assimilation of inorganic carbon within the ETNP OMZ R/V Atlantis AT50-08 from the Eastern Tropical Pacific Minimum Oxygen Zone from February to March 2023 (MicroPro project)

Website: https://www.bco-dmo.org/dataset/1005988
Data Type: Cruise Results
Version: 1
Version Date: 2026-08-25

Project
» Collaborative Research: Key Microbial Processes in Oxygen Minimum Zones: From In Situ Community Rate Measurements to Single Cells (MicroPro)

Contributors	Affiliation	Role
Pachiadaki, Maria G.	Woods Hole Oceanographic Institution (WHOI)	Co-Principal Investigator
Taylor, Gordon T.	Stony Brook University (SUNY Stony Brook)	Co-Principal Investigator
Yakubovskaya, Elena	Stony Brook University (SUNY Stony Brook)	Scientist
Butkevich, Natalie	Stony Brook University (SUNY Stony Brook)	Student
Newman, Sawyer	Woods Hole Oceanographic Institution (WHOI BCO-DMO)	BCO-DMO Data Manager

Abstract
Data profile chemoautotrophic assimilation of inorganic carbon by the microbial communities through the water column of the Eastern Tropical North Pacific's Oxygen Minimum Zone. Samples were collected on various dates in February and March of 2023 from two stations. Niskin bottle-collected whole water samples were collected in 1L bottles, dispensed into incubation septa test tubes and spiked with 14C-bicarbonate. One depth per sample set was selected for stimulation experiments. At these selected depths, replicates were additionally spiked with alternative electron donors. The unstimulated replicates from the same depth were size fractionated into 'free living' and 'particle associated' size fractions. Samples were transported to the home laboratory, suspended in scintillation cocktail and radioassayed. Samples were collected and prepared by Natalie Butkevich, Dr. Gordon Taylor and Dr. Elena Yakubovskaya. Samples were processed by Natalie Butkevich and Dr. Gordon Taylor.

Table of Contents

Coverage

Location: Eastern Tropical North Pacific
Spatial Extent: **N**:18.887 **E**:-104.89533 **S**:18.17683 **W**:-106.287
Temporal Extent: 2023-02-16 - 2023-03-10

Methods & Sampling

1L whole water samples were collected on deck under nitrogen pressure at the rosette. A three-volume overflow was performed for each sample to minimize oxygen contamination. Bottles were sealed with deoxygenated butyl rubber septa after confirming all bubbles were excluded. A He gas headspace was added in the shipboard lab no more than 15 minutes after collection. To quantify community chemoautotrophic activity, triplicate incubations with ^{14}C-bicarbonate were prepared in 16 mL Hungate tubes with degassed butyl rubber septa under nitrogen pressure. Chilled N2-purged ^{14}C-bicarbonate in an alkaline brine (pH 9.5; S = 60 on the practical salinity scale) was injected (0.92 µCi Mex1, 0.54 µCi Mex2) into the bottom before sealing (Tuttle and Jannasch 1973a). One depth per cast was selected for stimulation experiments with alternative electron donors. Selected incubations were spiked with ^{14}C-bicarbonate and one of the following: NH_4^+, DMSP, H_2S or S_2O_3. Incubations were conducted in a dark ice chest for 18-24 hours and terminated by filtration onto 25 mm nitrocellulose 0.22 µm filters (25 mm diameter Osmonics). Selected non-amended incubations were size fractionated into a “particle-associated” fraction (>2.7 µm) and a free-living size fraction (0.22 – 2.7 µm) by sequential filtration through stacked 25 mm Swinnex filter holders. Unassimilated ^{14}C-bicarbonate was purged from the filters in a saturated HCl atmosphere enclosure for >1h in a fume hood. Filters were then dried, suspended in scintillation cocktail (Hionic-Fluor) and radioassayed. Table 1 summarizes sample types. Data were corrected for isotopic fractionation (multiplied by 1.06) and for nonbiological sorption by use of control samples processed immediately after introduction of the radiotracer (T_0 samples). If blanks were higher than T_0 samples, blanks were used instead. Three T_0 sample sets were made, one for standard+stimulation experiments, one for the free-living size fraction and one for the particle-associated size fraction. Rates of dark ^{14}C-assimilation were normalized to total nanomoles of carbon per day via equation detailed under 'Data Processing' section.

Sample code	Tracer/Amendments	Experiment type	Notes
standard	^{14}C-bicarbonate	Standard	
S_2O_3	^{14}C-bicarbonate and S_2O_3	Alternative electron donor amendment	Target stimulant concentration: 10uM. For all samples prepared Feb.19 or later, target concentration is 5uM
DMSP	^{14}C-bicarbonate and DMSP	Alternative electron donor amendment	Target stimulant concentration: 100nM. For all samples prepared Feb.19 or later, target concentration is 50nM
NH_4^+	^{14}C-bicarbonate and NH_4^+	Alternative electron donor amendment	Target stimulant concentration: 10uM. For all samples prepared Feb.19 or later, target concentration is 5uM
H_2S	^{14}C-bicarbonate and H_2S	Alternative electron donor amendment	Target stimulant concentration: 10uM. For all samples prepared Feb.19 or later, target concentration is 5uM
0.2_SF	^{14}C-bicarbonate	Size fractionation	
2.7_SF	^{14}C-bicarbonate	Size fractionation	

Data Processing Description

Assimilation equation:

rate (nanomole carbon per liter per day) = (((2*10^6) * 1.06 * max(DPM - blank or t0, 0))/specific activity value)*(24/incubation time)

Constants:

2*10^6 = dissolved inorganic carbon in seawater (from Feely et al. 1995)

1.06 = isotopic fractionation factor

29 = standard blank in DPM

30 = small size fraction blank in DPM

37 = large size fraction blank in DPM

2032708 = specific activity for mex1 in DPM, 0.916 microCurie injected per sample

1190572 = specific activity for mex2 in DPM, 0.536 microCurie injected per sample

Notes:

-separate specific activities were used for Mex1 and Mex2 samples because separate stocks were prepared. The same blank values were used across all samples.

-max(DPM-blank or t0,0) implies that if the blank/t0 subtraction resulted in a negative value, 0 was substituted instead.

BCO-DMO Processing Description

Curation Actions Performed on Data
- Loaded data from "Chemoautotrophic assimilation of inorganic carbon_FINAL.xlsx" (sheet 1), using xlsx format with formatting preservation and floating point error adjustment enabled, headers on row 1, blank headers ignored, empty rows removed, and empty string and "nd" values treated as missing; table named 1005988_v1_ETNP_OMZ_AT50_Chemoautotrophic_Assimilation_of_Inorganic_Carbon
- Created new column ISO_Cast_Start_DateTime_UTC by combining Cast Date (UTC) and Cast Time (UTC) columns, parsing with formats %Y-%m-%d and %H:%M:%S respectively, treating input as UTC, and outputting as datetime in format %Y-%m-%dT%H:%M:%SZ, also in UTC
- Renamed columns: "Sample Type" to Sample_Type, "Cast Date (UTC)" to Cast_Date_UTC, "Cast Time (UTC)" to Cast_Time_UTC, "CTD Cast Filename" to CTD_Cast_Filename, "Incubation Duration" to Incubation_Duration, "Raw DPM" to Raw_DPM, "Assimilation_nMC_d^-1" to Assimilation_nMC, "Assimilation_sd_nMC_d^-1" to Assimilation_sd_nMC, "Replicate ID" to Replicate_ID
- Reordered columns to: Depth, Sample_Type, Station, Lat, Long, ISO_Cast_Start_DateTime_UTC, Cast_Date_UTC, Cast_Time_UTC, CTD_Cast_Filename, Incubation Start (local ship time), Incubation Stop (local ship time), Incubation_Duration, Raw_DPM, Assimilation_nMC, Assimilation_sd_nMC, Replicate_ID
- Set column types and formats: Depth and Raw_DPM as integer; Sample_Type, CTD_Cast_Filename, Replicate_ID, Station as string; Lat, Long, Incubation_Duration, Assimilation_nMC, Assimilation_sd_nMC as number; Cast_Date_UTC as date (format %Y-%m-%d); Cast_Time_UTC as time (format %H:%M:%S); ISO_Cast_Start_DateTime_UTC as datetime (format %Y-%m-%dT%H:%M:%SZ); Incubation Start (local ship time) and Incubation Stop (local ship time) as datetime (format %Y-%m-%d %H:%M)
- Updated column metadata with standard name IDs, descriptions, and supplied units for all columns, including marking Depth, ISO_Cast_Start_DateTime_UTC, Lat, and Long as primary parameters
- Dumped final table to CSV output named 1005988_v1_ETNP_OMZ_AT50_Chemoautotrophic_Assimilation_of_Inorganic_Carbon.csv

Curation Actions Performed on Metadata
- Standard metadata entry and text formatting steps were performed.

Problem Description

No problems or issues have been reported by the authors for this dataset.

[table of contents | back to top]

Related Publications

Feely, R. A., Wanninkhof, R., Cosca, C. E., Murphy, P. P., Lamb, M. F., & Steckley, M. D. (1995). CO2

distributions in the equatorial Pacific during the 1991–1992 ENSO event. Deep Sea Research Part II: Topical Studies in Oceanography, 42(2-3), 365–386. https://doi.org/10.1016/0967-0645(95)00027-n https://doi.org/10.1016/0967-0645(95)00027-N
Methods

Steeman-Nielsen E. (1952). The use of radioactive carbon (14C) for measuring organic production in the sea. J Conseil 18: 117–140. https://doi.org/10.1093/icesjms/18.2.117
Methods

Taylor, G. T., Iabichella, M., Ho, T.-Y., Scranton, M. I., Thunell, R. C., Muller-Karger, F., & Varela, R. (2001). Chemoautotrophy in the redox transition zone of the Cariaco Basin: A significant midwater source of organic carbon production. Limnology and Oceanography, 46(1), 148–163. Portico. https://doi.org/10.4319/lo.2001.46.1.0148
Methods

[table of contents | back to top]

Parameters

Parameter	Description	Units
Depth	Recorded depth at which Niskin bottle was closed and water sample was taken. Manually recorded from CTD cast at time of sample acquisition.	meters (m)
Sample_Type	Denotes type of sample, full description available in Table 1 of methods section	unitless
Station	Broadly categorizes cast location into 'Mex1' or 'Mex2', which corresponds to a nearshore or an offshore station respectively.	unitless
Lat	Latitude at which sample was taken, recorded in CTD cast header. A positive value indicates a northern latitude coordinate.	decimal degrees
Long	Longitude at which sample was taken, recorded in CTD cast header. A negative value indicates a western longitude coordinate.	decimal degrees
ISO_Cast_Start_DateTime_UTC	Start datetime of the associated cast in UTC. The values of this column were created by combining the original Cast Date and Cast Time column.	unitless
Cast_Date_UTC	Date the cast was made as recorded by CTD cast.	unitless
Cast_Time_UTC	Time the cast began recorded by CTD cast.	unitless
CTD_Cast_Filename	Filename associated with CTD cast corresponding to these samples.	unitless

Incubation_Start_Local_Ship_Time	The time (local time of the ship at the time of recording) the incubation was started by injecting tracer in local time, recorded to calculate incubation duration.	unitless
Incubation_Stop_Local_Ship_Time	The time (local time of the ship at the time of recording) the incubation was terminated by filtration in local time, recorded to calculate incubation duration.	unitless
Incubation_Duration	Incubation duration - the length of time the sample was incubated with the tracer and (if applicable) alternative electron donor	hours
Raw_DPM	Disintegrations per minute for each sample, recorded from scintillation counter output.	disintegrations per minute
Assimilation_nMC	Calculated mean carbon assimilation.	nanomole carbon per day
Assimilation_sd_nMC	Standard deviation was calculated by taking the sum of all assimilation values (nM C d^{-1}), dividing it by the number of samples, squaring the result, and then taking the square root (Standard deviation = sqrt(((sum of all assimilation_nMC_d^-1)^2)/#samples).	nanomole carbon per day
Replicate_ID	Triplicate identifier.	unitless

[table of contents | back to top]

Instruments

Dataset-specific Instrument Name	Scintillation counter: TRICARBB281000, serial number SGTC05150535
Generic Instrument Name	Liquid Scintillation Counter
Dataset-specific Description	A scintillation counter was used to measure the radioactivity of ^14C retained on the filters after incubation. Following removal of unassimilated ^14C-bicarbonate, the filters were dried and suspended in Hionic-Fluor scintillation cocktail. The measured radioactivity represented biologically assimilated ^14C and was used to calculate rates of dark carbon assimilation and community chemoautotrophic activity. Measurements were corrected using time-zero controls or blanks and for isotopic fractionation.
Generic Instrument Description	Liquid scintillation counting is an analytical technique which is defined by the incorporation of the radiolabeled analyte into uniform distribution with a liquid chemical medium capable of converting the kinetic energy of nuclear emissions into light energy. Although the liquid scintillation counter is a sophisticated laboratory counting system used to quantify the activity of particulate emitting (ß and a) radioactive samples, it can also detect the auger electrons emitted from 51Cr and 125I samples. Liquid scintillation counters are instruments assaying alpha and beta radiation by quantitative detection of visible light produced by the passage of rays or particles through a suitable scintillant incorporated into the sample.

[table of contents | back to top]

Deployments

AT50-08B

Website	https://www.bco-dmo.org/deployment/995472
Platform	R/V Atlantis
Start Date	2023-02-10
End Date	2023-03-16
Description	Project: Collaborative Research: Key Microbial Processes in Oxygen Minimum Zones: From In Situ Community Rate Measurements to Single Cells Chief: Pachiadaki, Maria G Start port: Putarenas, Costa Rica End port: Puntarenas, Costa Rica See additional information at R2R: https://www.rvdata.us/search/cruise/AT50-08B

[table of contents | back to top]

Project Information

Collaborative Research: Key Microbial Processes in Oxygen Minimum Zones: From In Situ Community Rate Measurements to Single Cells (MicroPro)

Coverage: East Tropical North Pacific Ocean

NSF Award Abstract:

Oxygen availability shapes the distributions and activities of marine organisms. Ongoing human activities and climate change are expected to lead to expansion and intensification of already large oxygen-stressed areas of the coastal and open ocean. Decreases in ocean oxygen have significant ecological consequences, including habitat loss for migratory and bottom-dwelling organisms, modification of the marine food web, and production of trace gases with pronounced feedbacks on climate, such as methane and nitrous oxide. Intense

chemical cycling by microorganisms occurs in oxygen-depleted marine habitats. However, a full understanding of the consequences for marine ecosystems is hampered by limited knowledge of actual rates of key microbiological processes and dynamics of the microorganisms mediating them. This study combines novel methods and sampling techniques to understand how these processes are influenced by changes in oxygen concentration to inform predictions of important chemical exchanges within a changing ocean and its production of climate-active gases. This deeply collaborative project trains undergraduates (four of whom participate on the cruise), a graduate student and a postdoctoral fellow. Outreach takes place in middle and high schools and through social media. Data and samples from the cruise are integrated in coursework.

Oxygen depletion alters cycling of major elements (especially carbon, nitrogen, and sulfur) as well as food web functionality. This project addresses major gaps in our knowledge of oxygen minimum zone (OMZ) processes by applying in situ approaches to more accurately measure rates of several key microbial processes (chemoautotrophy, denitrification, anammox, sulfate reduction and sulfide oxidation) central to marine biogeochemical cycling. This work studies the Eastern Tropical North Pacific OMZ, the largest open ocean oxygen-depleted system, to 1) determine the in situ rates of microbial processes involved in carbon, nitrogen, and sulfur cycling, 2) reveal the genomic blueprint of active single cells involved in these processes, and 3) obtain estimates of the relative contributions of the dominant chemoautotrophic and heterotrophic groups to the measured rates. This work include applies cutting-edge equipment for in situ sampling and incubations that minimize artifacts associated with traditional water sampling approaches, allowing more accurate estimates of rates of important biogeochemical processes. Additionally, rate measurements of relatively undisturbed bulk and fractionated water samples make it easier to distinguish the potential role of particle-associated microorganisms in these OMZ processes. Single cell sorting of microorganisms using a fluorescent dye indicative of cell activity together with metatranscriptomics informs on metabolic pathways used for key processes by active microbial community members, as well as the potential coupling of chemoautotrophy and nitrogen or/and sulfur cycling. By combining stable isotope probing, fluorescence in situ hybridization and single cell Raman microspectrometry the relative activity levels of different microbial phylotypes involved in chemoautotrophic and heterotrophic elemental cycling are assessed.

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.

[table of contents | back to top]

Funding

Funding Source	Award
NSF Division of Ocean Sciences (NSF OCE)	OCE-1924492
NSF Division of Ocean Sciences (NSF OCE)	OCE-1924424

[table of contents | back to top]