Measurements of chemoautotrophy in samples collected on cruise LMG1801 on R/V Laurence M. Gould from January to February 2018

Website: https://www.bco-dmo.org/dataset/840078
Data Type: Cruise Results
Version: 1
Version Date: 2021-04-08

Project
» Collaborative Research: Chemoautotrophy in Antarctic Bacterioplankton Communities Supported by the Oxidation of Urea-derived Nitrogen (Oxidation of Urea N)

Contributors	Affiliation	Role
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Popp, Brian N.	University of Hawaii at Manoa (SOEST)	Co-Principal Investigator
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Abstract
This dataset presents estimates of chemoautotrophy, measured as dark incorporation of 14C-DIC, as well as QA/QC measurements in samples collected on LMG1801.

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Coverage

Spatial Extent: **N**:-64.08913 **E**:-64.40628 **S**:-68.69457 **W**:-74.47352
Temporal Extent: 2018-01-08 - 2018-02-04

Methods & Sampling

Sample Collection. Samples were collected on the Antarctic continental shelf and slope west of the Antarctic Peninsula within the PAL-LTER sampling domain (http://pal.lternet.edu/) during summer (cruise dates 30 Dec 2017 through 12 Feb 2018; sampling dates 5 Jan to 4 Feb 2018) from the ARSV Laurence M Gould (LMG 1801, PAL-LTER cruise 26, DOI: 10.7284/907858). Sampling focused on three or 4 depths at each station chosen to represent the Antarctic Surface Water (ASW, 0 -34 m depth), the Winter Water (WW, the water column temperature minimum, generally between 35 and 174 m) the Circumpolar Deep Water (CDW, 175-1000 m) and slope water (SLOPE, >1000 m, generally ~10 m above the bottom at deep stations on the slope, 2500-3048m). Water samples were collected from Niskin bottles (General Oceanics Inc., Miami, FL, USA) into opaque 2 L HDPE plastic bottles or into aged, acid-washed, sample-rinsed 250 ml polycarbonate bottles (Nalge) completely filled (~270 mL) directly from Niskin bottles as soon as possible after the rosette was secured on

deck. Subsequent processing took place in an adjacent laboratory.

Samples for DNA analysis were taken from the 2 L opaque HDPE bottles and were filtered under pressure through 0.22 um pore size Sterivex GVWP filters (EMD Millipore, Billerica, MA, USA) using a peristaltic pump. Residual seawater was expelled from the filter using a syringe filled with air, then ~1.8 ml of lysis buffer (0.75 M sucrose, 40 mM EDTA, 50 mM Tris, pH 8.3) was added to the filter capsule, which was capped and placed in a -20 °C freezer. The frozen samples were aggregated into Ziploc Freezer Bags and transferred to a -80 °C freezer for the remainder of the cruise and for shipping to the laboratory.

Two samples of the Sterivex filtrate (40 mL each into new 50 mL disposable centrifuge tubes, VWR, rinsed 3x with sample) were frozen immediately at -20 °C, then aggregated into Ziploc Freezer Bags and transferred to a -80 ° freezer for the remainder of the cruise and for shipping to the laboratory. These were used for subsequent determination of 1) urea concentration and 2) the natural abundance of 15N in the nitrite plus nitrate pools ($15NO_x$ hereinafter). An additional sample of the Sterivex filtrate was stored in a polycarbonate bottle at 4 °C for subsequent onboard determination of ammonia concentration by the Holmes et al (1999) o-phthaldialdehyde method and nitrite concentration by the diazo-coupling method (Strickland and Parsons 1972). Technical difficulties encountered during onboard analysis resulted in the loss of ammonium and nitrite data for some samples.

Samples for DNA and chemical analyses were shipped on dry ice from Punta Arenas, Chile to the Hollibaugh laboratory at the University of Georgia. Upon arrival they were stored in a -80 °C freezer until analyzed. Samples for 15N analysis were shipped on dry ice from Punta Arenas, Chile to the Popp laboratory at the University of Hawaii. Upon arrival they were stored in a -40 °C freezer until analyzed.

Chemoautotropic production. Samples used for determining chemoautotrophic production were held in the dark at 0°C for no longer than 6 hours prior to being amended with 14C bicarbonate. Chemoautotrophy was determined by measuring the incorporation into organic matter of 14C supplied as $NaH14CO_3$. $NaH14CO_3$ (5 mCi) was diluted into 25 mL of MilliQ water made basic (pH ≈9) using NaOH. This stock solution (0.2 uCi/uL) was passed through a syringe filter (Acrodisk, 0.22 pore size) into a 30 mL polycarbonate bottle and stored at 4°C. Water from the sample depth was collected directly from the appropriate Niskin sampler into aged, acid-washed 250 mL screw-cap amber HDPE bottles (3 rinses) filled to the top (volume ~270 mL). Each experiment used two replicate treatments and a control bottle. Controls consisted of either 0.22 um filtered water from the Sterivex filtration of the same sample or of whole water that was incubated along with the 14C amended treatment, except that no 14C was added until immediately before filtering the set. Each bottle received ~ 20 uCi of $NaH14CO_3$ (100 uL of the working stock). Label was added in a darkened lab van illuminated with a dim, red-filtered light. Samples were mixed by inverting gently then placed in a water-ice bath contained in an ice chest wrapped in aluminum foil contained in a black 3 mil plastic garbage bag. Incubation temperature was maintained by adding ice as needed, which led to departures to above the desired incubation temperature for some stations. Water temperature in the bath was recorded at 5-minute time steps with HOBO TidBit loggers and these samples have been flagged. Lights were kept off except when working in the lab van, when red light was used to illuminate the working area.

See the attached supplemental files for detailed data on the water bath temperature. "LMG1801_Water_Bath_Temperature_Plot.pdf" contains a figure plotting the bath temperature, the measured chemoautotrophy value for each station and depth, and the intervals of the incubations by station. "Water_Bath_Temperature_Data.xlsx" contains the data used to create this plot.

At the end of the incubation (~48 hours) the bottles were removed from the ice bath, triplicate samples of 100 uL were taken from the filtered control (or from one of the treatments in later experiments) and radioassayed to verify the amount of tracer added. The remaining sample and all of the treatment samples were filtered through 25 mm diameter, 0.22 um pore size membrane filters (GSWP Millipore) under dim red light. Filters were rinsed two times with filtered seawater, removed from the filter holder into scintillation vials, then 100 uL of 10% HCl was added to each vial, soaking the filter in the process. After 24 hr in the fume hood (uncapped) to allow excess water and acid to volatilize, vials received 4 mL of Ultima Gold scintillation cocktail, then were counted in a Perkin-Elmer LSC.

BCO-DMO Processing Description

- renamed fields to comply with BCO-DMO naming conventions;
- converted start and end date/time fields to ISO8601 format;
- 2021-03-16: revised/updated the Methods & Sampling section of the metadata;
- 2021-04-08: replaced data file with copy received on 2021-03-18.

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Data Files

File
chemoautotrophy.csv (Comma Separated Values (.csv), 6.73 KB) MD5:b61fd408265ad730576ad095ef9de132 Primary data file for dataset ID 840078

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Supplemental Files

File
Water Bath Temperature Data filename: Water_Bath_Temperature_Data.xlsx (Octet Stream, 658.53 KB) MD5:c7387eb7a547679bbe878ce3efc7d7db These data were used to generate the associated plot saved in file "LMG1801_Water_Bath_Temperature_Plot.pdf". This spreadsheet reports the temperature recorded during the incubations in the ice and water bath used to maintain a target incubation temperature of 0 oC. This spreadsheet reports the temperature in the incubator used in our measurements of N oxidation rates. Data were collected using a HOBO TidBit logger (Logger S/N: 20211708, Sensor S/N: 20211708) placed in the water bath. The "Metadata" sheet within this Excel file contains descriptions of the data columns. Updated from data file received on 2021-03-18.
Water Bath Temperature Plot filename: LMG1801_Water_Bath_Temperature_Plot.pdf (Portable Document Format (.pdf), 427.88 KB) MD5:38c5901ab32eaf61cafd4dca05511785 This figure plots the bath temperature (blue line), the measured chemoautotrophy value (orange ticks, units of nmol C fixed L-1 d-1) for each station and depth, and the intervals of the incubations by station as colored bars below the x-axis, whose position is keyed to temperature (y-axis) values. The interval is the duration of the experiment, start to finish, local time, in DDMMYY HH:MM, format, 24-hour clock, GMT-5). Updated from data file received on 2021-03-18.

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Related Publications

Holmes, R. M., Aminot, A., Kérouel, R., Hooker, B. A., & Peterson, B. J. (1999). A simple and precise method for measuring ammonium in marine and freshwater ecosystems. Canadian Journal of Fisheries and Aquatic Sciences, 56(10), 1801–1808. doi:10.1139/f99-128
Methods

Strickland, J. D. H. and Parsons, T. R. (1972). A Practical Hand Book of Seawater Analysis. Fisheries Research Board of Canada Bulletin 157, 2nd Edition, 310 p.
Methods

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Parameters

Parameter	Description	Units
Event_Log_Number	Sequential numbers keyed to the bridge log of activities	unitless
Cast_Start_Time_GMT	Date and time of day for beginning CTD cast = sample collection; 24-hour clock; formatted to ISO8601 standard (UTC/GMT): YYYY-MM-DDThh:mmZ	unitless
Latitude	Latitude in decimal degrees (negative values = South)	degrees North
Longitude	Longitude in decimal degrees (negative values = West)	degrees East
Station_Description	PAL-LTER category for the station	unitless
LTER_Grid_Station	Station location on the PAL-LTER sampling grid (http://pal.lternet.edu)	unitless
Depth	Depth sampled in meters	meters (m)
Treatment	Identifies manipulation experiments or comparisons	unitless
Fraction_of_label_incorporated	Fraction of the added 14C that was incorporated into organic matter	unitless
Start_ISO_DateTime_UTC	Time at which the incubation was initiated by adding 14C to the sample; 24-hour clock; formatted to formatted to ISO8601 standard (UTC/GMT): YYYY-MM-DDThh:mmZ	unitless
End_ISO_DateTime_UTC	Time at which the incubation was terminated by filtration; 24-hour clock; formatted to formatted to ISO8601 standard (UTC/GMT): YYYY-MM-DDThh:mmZ	unitless
Incubation_Length	Duration of the incubation in days	days
Midpoint_of_incubation	Julian Day of 2018, as a decimal day (GMT)	decimal days
Chemoautotrophy	Carbon fixation rate, nmol C L-1 d-1	nanomoles C per liter per day (nmol C L-1 d-1)
Incubation_Temp_Max	Maximum temperature recorded in the incubator during an incubation	degrees Celsius
Incubation_Temp_Min	Minimum temperature recorded in the incubator during an incubation	degrees Celsius
Incubation_Temp_Median	Median temperature recorded in the incubator during an incubation	degrees Celsius
Incubation_Temp_Mean	Mean temperature recorded in the incubator during an incubation	degrees Celsius
Incubation_Temp_St_Dev	Standard Deviation of the temperature recorded in the incubator during an incubation	degrees Celsius

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Instruments

Dataset-specific Instrument Name	Perkin-Elmer LSC
Generic Instrument Name	Liquid Scintillation Counter
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 the quantify the activity of particulate emitting (ß and a) radioactive samples, it can also detect the auger electrons emitted from 51Cr and 125I samples.

Dataset-specific Instrument Name	Niskin bottles (General Oceanics Inc., Miami, FL, USA)
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	HOBO TidBit loggers
Generic Instrument Name	Temperature Logger
Generic Instrument Description	Records temperature data over a period of time.

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Deployments

LMG1801

Website	https://www.bco-dmo.org/deployment/839984
Platform	ARSV Laurence M. Gould
Start Date	2017-12-30
End Date	2018-02-12
Description	Additional cruise information is available from the Rolling Deck to Repository (R2R): https://www.rvdata.us/search/cruise/LMG1801 Cruise DOI: 10.7284/907858

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Project Information

Collaborative Research: Chemoautotrophy in Antarctic Bacterioplankton Communities Supported by the Oxidation of Urea-derived Nitrogen (Oxidation of Urea N)

Coverage: Coastal, shelf and slope waters off the West Antarctic Peninsula, PAL-LTER sampling grid, Lawrence M Gould cruise 18-01

NSF Award Abstract:
Part 1: The project addresses fundamental questions regarding the role of nitrification (the conversion of ammonium to nitrate by a two-step process involving two different guilds of microorganisms: ammonia- and nitrite-oxidizers) in the Antarctic marine ecosystem. Specifically, the project seeks to evaluate the contribution of primary production supported by the energy in nitrogen compounds to the overall supply of organic carbon to the food web of the Southern Ocean. Previous measurements indicate that nitrification could contribute about 9% to primary production supporting the Antarctic food web on an annual basis, but those measurements did not include the additional production associated with nitrite oxidation. Additionally, the project will aim to determine the significance of the contribution of other sources of nitrogen, (specifically organic nitrogen and urea released by other organisms) to nitrification because these contributions may not be assessed by standard protocols. Such work will aid in better understanding the basis of the energy for the Antarctic marine ecosystems on an annual basis as well as better aid in understanding the energetics of the ecosystem in times and places where primary production based on light energy is limited (i.e. during the polar night or under sea ice cover).

This project will result in training a postdoctoral researcher and provide undergraduate students opportunities to gain hand-on experience with research on microbial geochemistry. The Palmer Long Term Ecological Research (LTER) activities have focused largely on the interaction between ocean climate and the marine food web affecting top predators. Relatively little effort has been devoted to studying processes related to the microbial geochemistry of nitrogen cycling, yet these are a major themes at other LTER sites. This work will contribute substantially to understanding an important aspect of nitrogen cycling and bacterioplankton production in the study area. The team will be working synergistically and be participating fully in the education and outreach efforts of the Palmer LTER, including making highlights of the findings available for posting to their project web site and participating in any special efforts they have in the area of outreach.

Part 2: The proposed work will quantify oxidation rates of 15N supplied as ammonium, urea and nitrite, allowing the estimation of the contribution of urea-derived N and complete nitrification (ammonia to nitrate) to chemoautotrophy and bacterioplankton production in Antarctic coastal waters. The project will compare these estimates to direct measurements of the incorporation of 14C into organic matter in the dark for an independent estimate of chemoautotrophy. The team aims to collect samples spanning the water column: from surface water (~10 m), winter water (50-100 m) and circumpolar deep water (>150 m); on a cruise surveying the continental shelf and slope west of the Antarctic Peninsula in the austral summer of 2018. Other samples will be taken to measure the concentrations of nitrate, nitrite, ammonia and urea, for qPCR analysis of the abundance of relevant microorganisms, and for studies of related processes. The project will rely on collaboration with the existing Palmer LTER to ensure that ancillary data (bacterioplankton abundance and production, chlorophyll, physical and chemical variables) will be available. The synergistic activities of this project along with the LTER activities will provide a unique opportunity to assess chemoautotrophy in context of the overall ecosystem's dynamics- including both primary and secondary production processes.

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Funding

Funding Source	Award
NSF Office of Polar Programs (formerly NSF PLR) (NSF OPP)	OPP-1643466
NSF Office of Polar Programs (formerly NSF PLR) (NSF OPP)	OPP-1643345

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