Iron and vitamin B12 metatranscriptomincs co-limitation in sea water collected from the Ross Sea in 2014 (Ross Sea Microb Ecophys project)

Website: https://www.bco-dmo.org/dataset/628290
Data Type: experimental
Version: 2015-12-07

Project
» Synergistic Effects of Iron, Carbon Dioxide and Temperature on the Fate of Nitrate: Implications for Future Changes in Export Production in the Southern Ocean (Ross_Sea_Microb_Ecophys)

Contributors	Affiliation	Role
Allen, Andrew E.	J. Craig Venter Institute (JCVI)	Principal Investigator
Bronk, Deborah A.	Virginia Institute of Marine Science (VIMS)	Co-Principal Investigator
Hutchins, David A.	University of Southern California (USC-WIES)	Co-Principal Investigator
Copley, Nancy	Woods Hole Oceanographic Institution (WHOI BCO-DMO)	BCO-DMO Data Manager

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Dataset Description

We examined the short-term transcriptional response of the 16 Jan 2013 community to cobalamin and iron addition in order to further characterize the nature and implications of this colimitation. RNA sequencing was performed on triplicate samples, 24 h after micronutrient addition. Sequencing and assembly statistics are given in Bertrand et al. 2015.

Data deposition: The data reported in this paper have been deposited in the NCBI sequence read archive (BioProject accession no. PRJNA281813; BioSample accession nos.SAMN03565520–SAMN03565531). Assembled contigs, predicted peptides, annotation, and transcript abundance data for this study can be found at https://scripps.ucsd.edu/labs/aallen/data/. These files are also accessible via the '**Get Data**' link above.

Methods & Sampling

Near-surface water samples were collected by helicopter in the Ross Sea near McMurdo Station, Antarctica.

Methodology: From Bertrand, E.M., McCrow, J.P., Moustafa, A., Zheng, H., McQuaid, J., Delmont, T., Post, A.F., Sipler, R., Spackeen, J.L., Xu, K., Bronk, D.A., Hutchins, D.A., Allen, A.E. (2015) Phytoplanktont-bacterial interactions mediate micronutrient colimitation at the coastal Antarctic sea ice edge. Proceedings of the National Academy of Sciences. 10.1073/pnas.1501615112

On January 16, 2013, seawater was collected from3mdepth at the sea ice edge in McMurdo Sound of the Ross Sea (77° 36.999' S 165° 28.464' E), using trace metal clean technique. Triplicate bottles (2.7 L) of each treatment (unamended control, + 1 nM added FeCl3, + 200 pM added cyanocobalamin, and + 200 pM cyanocobalamin and 1 nM Fe) were placed in an indoor incubator at 0 °C, ~45 µmol photonsm-2·s-1 of constant light. After 24 h, RNA sampleswere collected (450 mL) and nitrate uptake and primary productivity

rates were measured. Samples were taken for Chl a at 0, 24, and 96 h. RNA was extracted using the TRIzol reagent (Life Technologies). Ribosomal RNA was removed with Ribo-Zero Magnetic kits, and the resulting mRNA enrichment was purified and subjected to amplification and cDNA synthesis, using the Ovation RNA-Seq System V2 (NuGEN). One microgram of the resulting high-quality cDNA pool was fragmented to a mean length of 200 bp, and Truseq (Illumina) libraries were prepared and subjected to paired-end sequencing via Illumina HiSeq. Reads were trimmed and filtered, contigs were assembled in CLC Assembly Cell (CLCbio), and ORFs were predicted (41). ORFs were annotated de novo for function via KEGG, KO, KOG, Pfam, and TigrFam assignments. Taxonomic classification was assigned to each ORF using a reference dataset, as described in the SI Appendix (Bertrand, 2015), and the Lineage Probability Index (LPI, as calculated in ref. 18). edgeR was used to assign normalized fold change and determine which ORFs were significantly differentially expressed in pairwise comparisons between treatments, considering triplicates, within a given phylogenetic grouping (42). For Fig. 2, diatom ORFs [identified as diatom via LPI analyses; LPI > 0.8 (18)] were clustered using MCL (Markov cluster algorithm) (21), and these clusters were used to produce MANTA plots (20). For Oceanospirillaceae ASP10-02a genome assembly, water was collected from 10 m in the Amundsen Sea on December 19, 2010. Metagenomic libraries were created with the OVATION ultralow kit (NuGen). Overlapping and gapped metagenomic DNA libraries were prepared for sequencing on a HiSeq platform (Illumina). CLC was used to assemble scaffolds, tetranucleotide frequencies of scaffolds were analyzed (43), and draft genomes were generated via binning scaffolds clustered (hierarchical) together in well-supported clades, refined using GC content and taxonomical affiliation (44). ORFs identified in metatranscriptomic analyses were mapped to the Oceanospirillaceae ASP10-02a genome bin from the best-scoring nucleotide alignment, using BWA-MEM with default parameters (45). ORFs with >99%similarity to the genome scaffold sequences were used for subsequent analyses of gene expression patterns within this population. Complete materials and methods are given in the SI Appendix (Bertrand, 2015).

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Data Files

File
cDNA.csv (Comma Separated Values (.csv), 2.18 KB) MD5:884dbb9d36fc38e0de013ed8ed7fc85d Primary data file for dataset ID 628290

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Parameters

Parameter	Description	Units
description	description of file type	unitless
NCBI_BioProject	NCBI BioProject identification	unitless
file_size_MB	size of file available for download	MegaBytes
download_link	link to download file or to NCBI accession page	unitless

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Instruments

Dataset-specific Instrument Name	
Generic Instrument Name	Automated DNA Sequencer
Dataset-specific Description	Illumina HiSeq platform
Generic Instrument Description	A DNA sequencer is an instrument that determines the order of deoxynucleotides in deoxyribonucleic acid sequences.

Dataset-specific Instrument Name	
Generic Instrument Name	Thermal Cycler
Dataset-specific Description	Life Technologies AccuPrime PCR system Life Technologies ProFlex PCR system
Generic Instrument Description	A thermal cycler or "thermocycler" is a general term for a type of laboratory apparatus, commonly used for performing polymerase chain reaction (PCR), that is capable of repeatedly altering and maintaining specific temperatures for defined periods of time. The device has a thermal block with holes where tubes with the PCR reaction mixtures can be inserted. The cycler then raises and lowers the temperature of the block in discrete, pre-programmed steps. They can also be used to facilitate other temperature-sensitive reactions, including restriction enzyme digestion or rapid diagnostics. (adapted from http://serc.carleton.edu/microbelife/research_methods/genomics/pcr.html)

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Deployments

helicopter_Allen

Website	https://www.bco-dmo.org/deployment/628276
Platform	McMurdo Station
Start Date	2013-01-16
End Date	2015-01-23
Description	Water sample collections in the Ross Sea

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Project Information

Synergistic Effects of Iron, Carbon Dioxide and Temperature on the Fate of Nitrate: Implications for Future Changes in Export Production in the Southern Ocean (Ross_Sea_Microb_Ecophys)

Coverage: McMurdo Sound of the Ross Sea

Description from NSF award abstract:
This award provides support for "Collaborative Research: Synergistic Effects of Iron, Carbon Dioxide, and Temperature on the Fate of Nitrate: Implications for Future Changes in Export Production in the Southern

Ocean" from the Antarctic Organisms and Ecosystems program in the Office of Polar Programs at NSF. The project will use a novel combination of research approaches to evaluate the effects of temperature, carbon dioxide, and iron on three ecologically- and biogeochemically-critical Southern Ocean phytoplankton functional groups: Large centric diatoms, small pennate diatoms, and Phaeocystis antarctica.

The Southern Ocean around Antarctic is undergoing several changes including increased temperature and carbon dioxide content, as well as changing levels of biologically available iron. The project goals are to understand how the individual and combined influences of these three variables affect Southern Ocean phytoplankton community structure, and to determine how these assemblage-level responses are linked to fundamental cellular responses at the levels of nutrient physiology and gene expression. The research team will focus on three different types of marine algae: large and small diatoms, and the prymnesiophyte, Phaeocystis antarctica. Shifts between these three major algal groups have very different consequences for nutrient and carbon biogeochemistry in the rapidly changing Antarctic marine environments. However, the mechanistic underpinnings of these environmentally-driven community shifts are not known. The project includes a US-based laboratory component with Antarctic isolates, field study at McMurdo Station, and then a cruise of opportunity in the upwelling areas directly south of the Antarctic Circumpolar Current. The study also includes collection and analysis of environmental gene expression data, or meta-transcriptomics, both from the field and experimental settings. The transcriptomes will be generated under environmentally relevant conditions and will thus contain information critical for decoding the genomes of several newly sequenced polar phytoplankton species in addition to the three groups highlighted above.

Related publications:
Bertrand, E.M., McCrow, J.P., Moustafa, A., Zheng, H., McQuaid, J., Delmont, T., Post, A.F., Sipler, R., Spackeen, J.L., Xu, K., Bronk, D.A., Hutchins, D.A., Allen, A.E. 2015. Phytoplankton-bacterial interactions mediate micronutrient colimitation at the coastal Antarctic sea ice edge. Proceedings of the National Academy of Sciences. 10.1073/pnas.1501615112

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Funding

Funding Source	Award
NSF Office of Polar Programs (formerly NSF PLR) (NSF OPP)	PLR-1043671

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