Growth rates NE Pacific and Atlantic diatom isolates under various Zn and Co additions in experiments with cultures collected from R/V Thomas G. Thompson cruise TN280, along Line P in the NE Pacific, in May of 2012.

Website: https://www.bco-dmo.org/dataset/807316
Data Type: experimental, Cruise Results
Version: 1
Version Date: 2020-03-31

Project
» US GEOTRACES PMT: Cobalt Biogeochemical Cycling and Connections to Metalloenzymes in the Pacific Ocean (PMT Cobalt and Metalloenzymes)
» Marine Microbial Investigator Award: Investigator Mak Saito (MM Saito)

Program
» Marine Microbiology Initiative (MMI)

Contributors	Affiliation	Role
Saito, Mak A.	Woods Hole Oceanographic Institution (WHOI)	Principal Investigator
York, Amber D.	Woods Hole Oceanographic Institution (WHOI BCO-DMO)	BCO-DMO Data Manager

Abstract
Growth rates of two NE Pacific Line P diatom isolates and two Atlantic diatom isolates under various Zn and Co additions. Cultures were collected from the GeoMICS expedition on the R/V Thomas G. Thompson (cruise TN280), along Line P in the NE Pacific, in May of 2012.

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Coverage

Spatial Extent: **N**:54 **E**:-4 **S**:40.756 **W**:-128.666
Temporal Extent: 2012-05-17 - 2012-05-22

Dataset Description

Growth rates of two NE Pacific Line P diatom isolates and two Atlantic diatom isolates under various Zn and Co additions. Cultures were collected from the GeoMICS expedition on the R/V Thomas G. Thompson (cruise TN280), along Line P in the NE Pacific, in May of 2012.

Methods & Sampling

Location: Northeast Pacific Line P Transect 48.8167 N 128.667 W

Media and culturing techniques
Northwest Atlantic Thalassiosira pseudonana CCMP1335 cultures were maintained in a 24°C incubator under constant fluorescent lighting (65 µmol quanta m-2 s-1 PAR). Northeast Atlantic Phaeodactylum tricornutum CCMP632 cultures were maintained in an 18°C incubator under constant fluorescent lighting (90 µmol quanta m-2 s-1 PAR). Both Pacific Pseudo-nitzschia delicatissima UNC1205 and Pacific Thalassiosira sp. UNC1203 were grown in an 18°C incubator under constant fluorescent lighting (85 µmol quanta m-2 s-1 PAR). All cultures were randomly repositioned each day to avoid any effect of subtle variation in light intensity on growth. T. pseudonana CCMP1335 and P. tricornutum CCMP632 (Bigelow Laboratory, East Boothbay, ME) were obtained from the Mincer and Saito laboratory culture collections at the Woods Hole Oceanographic Institution, respectively. Pacific P. delicatissima UNC1205 and Thalassiosira sp. UNC1203 were obtained from the Marchetti laboratory at the University of North Carolina. Both Pacific isolates were collected at station P8 of Line P, a Northeast Pacific ocean transect located off of Vancouver Island comprising 26 sampling stations and ending at Ocean Station Papa at 50°N 145°W (Fig. 1a). All cultures were axenic and maintained by sterile technique until needed.
Polycarbonate and plastic bottles were cleaned to remove trace metal contaminants before use. This procedure involved, at minimum, a 72h soak in <1% Citranox detergent, five rinses in mili-Q water, a 7d soak in 10% HCl, and five rinses with dilute acid (HCl, pH 2). Cultures were grown in microwave sterilized 28 mL polycarbonate centrifuge tubes, and all solutions were pipetted only after a tip rinse procedure consisting of three rinses with 10% HCl followed by three rinses with sterile dilute HCl (pH 2). All culture work was conducted in a Class 100 clean room.
Culture media was prepared after that used by Sunda and Huntsman for trace metal experimentation (Sunda and Huntsman 1992, 1995b). Microwave sterilized, 0.2µm-filtered Pacific seawater from the North Pacific station “Aloha” (22° 45'N, 158° 00'W) was used as the media base. Macronutrients were added to this sterile base to a final concentration of 88.2µM $NaNO_3$, 41.5µM NaH_2PO_4, and 106µM Na_2SiO_3 and were chelexed before use. Added vitamins included 2 nM biotin, 0.37 nM B12 as cyanocobalamin, and 300 nM thiamine and were also chelexed before use. Trace metals were added to final media concentrations of 10-7 M $FeCl_3$, 4.8 x 10-8 M $MnCl_2$, 4.0 x 10-8 M $CuSO_4$, 10-7 M $NiCl_2$, and 10-8 M Na_2O_3Se within a 10-4 M ethylenediamine tetraacetic acid disodium salt (EDTA, Acros Organics, $C_{10}H_{14}N_2Na_2O_8$) metal ion buffer system. All media amendments were sterile filtered through acid rinsed 0.2µm filters before addition to final media, and final media equilibrated for at least 12h before inoculation.
Established cultures of each diatom were first acclimated in low-metal media containing 1 nM total Zn or less for at least three transfers. These acclimated cultures were used to initially inoculate transfer 1 (T1) cultures at 1% volume. For all diatoms, Zn or Co limitation experiments were first performed using a range of Zn concentrations with Co omitted and vice versa. We refer to experiments using media amended with Zn or Co (while omitting the other) as “simple limitation” experiments. Experiments varying concentrations of both Zn and Co were also conducted, allowing for three-dimensional visualizations of growth rates as used previously (Saito et al. 2002; Saito and Goepfert 2008). We refer to experiments using media amended with both Zn and Co as “matrix” experiments. In vivo fluorescence (Turner Instruments TD-700) of simple and matrix experiment cultures was measured on a near-daily basis as a proxy for chlorophyll a and is reported as relative fluorescence units (RFU). Growth rates were calculated from the exponential portion of each culture’s growth curve. Computed ratios of $[Zn^{2+}]$ and $[Co^{2+}]$ to total concentrations, whose values are $10^{-3.99}$ and $10^{-3.63}$ respectively, were used to convert added metal concentrations to free ion concentrations and are the same as those used by Sunda and Huntsman (Sunda and Huntsman 1995).

Isolation sources and locations:
* Pseudonitzschia delicatissima UNC1205 and Thalassiosira UNC1203 were isolated from station P8 of the Line P transect, 48.817°N 128.666°W
* Phaeodactylum tricornutum CCMP632 was ordered from Bigelow, the strains original location of isolation was 54°N 4°W
* Thalassiosira pseudonana CCMP1335 was also from Bigelow, original location of isolation was 40.756° N 72.82° W

Data Processing Description

BCO-DMO Data Manager Processing Notes:
* Combined separate sheets in originally submitted excel file "Line P Diatom Growth Rate Tables BCO-DMO.xlsx" and extracted to csv format.
* Column "culture" added to capture information in the title of each of the original tables (e.g. Thalassiosira UNC1203)

* added a conventional header with dataset name, PI name, version date
* Missing data identifier in original data "NA" is displayed by default as "nd" in the bco-dmo data system.
* modified parameter names to conform with BCO-DMO naming conventions (only letters, numbers, and underscores)

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Data Files

File
growth_rates.csv (Comma Separated Values (.csv), 8.44 KB) MD5:cfa6de86412f5b6cb3353f1603b4c0e7 Primary data file for dataset ID 807316

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Parameters

Parameter	Description	Units
Culture	Culture name (e.g. Line P Thalassiosira sp. UNC1203)	unitless
Experiment_Type	Experiment Type. One (simple) or both (matrix) metals added.	unitless
Added_Zn	Total amount of added zinc (Zn) to incubation.	nanomoles per liter (nmol/L)
Added_Co	Total amount of added cobalt (Co) to incubation.	nanomoles per liter (nmol/L)
Total_Zn	Total zinc (Zn). Added Zn and background media Zn.	nanomoles per liter (nmol/L)
Total_Co	Total cobalt (Co). Added Co and background media Co.	nanomoles per liter (nmol/L)
log_Zn2plus	Log of calculated free Zn2+ ion concentration in media. [Zn2+]	moles per liter (mol/L)
log_Co2plus	Log of calculated free Co2+ ion concentration in media. [Co2+]	moles per liter (mol/L)
Growth_rate_replicate_A	Growth rate of treatment replicate A	per day (d-1)
Growth_rate_replicate_B	Growth rate of treatment replicate B	per day (d-1)
Growth_rate_average	Average growth rate of replicates A and B	per day (d-1)
Maximum_RFU	Maximum relative fluorescence	Relative Fluorescence Units (RFU)

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Instruments

Dataset-specific Instrument Name	Turner Instruments TD-700 Fluorometer
Generic Instrument Name	Turner Designs 700 Laboratory Fluorometer
Dataset-specific Description	Turner Instruments TD-700 Fluorometer calibrated to a solid standard.
Generic Instrument Description	The TD-700 Laboratory Fluorometer is a benchtop fluorometer designed to detect fluorescence over the UV to red range. The instrument can measure concentrations of a variety of compounds, including chlorophyll-a and fluorescent dyes, and is thus suitable for a range of applications, including chlorophyll, water quality monitoring and fluorescent tracer studies. Data can be output as concentrations or raw fluorescence measurements.

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Deployments

TN280

Website	https://www.bco-dmo.org/deployment/664928
Platform	R/V Thomas G. Thompson
Start Date	2012-05-16
End Date	2012-05-22

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Project Information

US GEOTRACES PMT: Cobalt Biogeochemical Cycling and Connections to Metalloenzymes in the Pacific Ocean (PMT Cobalt and Metalloenzymes)

Coverage: Laboratory Study and Cultures from Northeast Pacific Line P Transect 48.8167 N 128.667 W

NSF abstract:

Cobalt is important for many forms of marine life, yet it is one of the scarcest nutrients in the sea. Cobalt's oceanic abundance and distribution, along with other scarce nutrients, can influence the growth of microscopic plants (phytoplankton). This in turn can influence carbon cycles in the ocean and atmosphere. Therefore, knowledge of the controls on cobalt's abundance and chemical forms in seawater is a valuable component of our ability to understand the ocean's influence on global carbon cycling. Within phytoplankton and other marine microbes, metals such as cobalt, iron, nickel, and copper are used as critical components of enzymes responsible for key cellular reactions. Since these enzymes require metals to work, they are named metalloenzymes. Participating in a Pacific Ocean cruise from Alaska to Tahiti, this project will study the oceanic distributions of dissolved cobalt and the cellular content of a group of metalloenzymes known to influence biogeochemical cycles. The project will provide scientific impact by creating new knowledge about oceanic micronutrients in regions of economic interest with regard to fisheries and deep-sea mining. Measurement of proteins in the North Pacific will provide data of broad biological and chemical interest and will be made available through a new NSF-funded "EarthCube Ocean Protein Portal" data base. Educational impact will stem from participation of a graduate student and two young technicians, as well as the PI's development of a high school chemistry curriculum for use in two local high schools, thus allowing teachers to include real oceanic and environmental data at their first introduction to chemistry.

Cobalt has a complex biogeochemical cycle. Both its inorganic and organic forms are used by biology in the upper ocean and it is removed from solution by being scavenged in the intermediate and deep ocean. This scavenging removal results in cobalt having the smallest oceanic inventory of any biologically utilized element. Recent studies, however, have found that large dissolved cobalt plumes occur in major oxygen minimum zones due to a combination of less scavenging and additions from sedimentary and remineralization fluxes. The GP15 US GEOTRACES Pacific Meridional Transect (PMT) provides an opportunity to examine the influence of oxygen depletion on cobalt chemistry. Moreover, the study of the protein component of microbial communities using new proteomic techniques will provide evidence of how different major microorganisms respond to the chemical environment (e.g. through transporter production for specific nutrients and micronutrients) as well as the biochemical basis for metal requirements related to the use of specific metalloenzymes. Specifically, the PMT provides an opportunity to confirm that the Pacific oxygen minimum zones contain a large amount of cobalt and to test the hypotheses that simultaneous zinc scarcity could induce wide-scale biochemical substitution of cobalt for zinc in the North Pacific Ocean.

Marine Microbial Investigator Award: Investigator Mak Saito (MM Saito)

In support of obtaining deeper knowledge of major biogeochemically relevant proteins to inform a mechanistic understanding of global marine biogeochemical cycles.

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Program Information

Marine Microbiology Initiative (MMI)

Website: https://www.moore.org/initiative-strategy-detail?initiativeId=marine-microbiology-initiative

A Gordon and Betty Moore Foundation Program.

Forging a new paradigm in marine microbial ecology:

Microbes in the ocean produce half of the oxygen on the planet and remove vast amounts of carbon dioxide, a greenhouse gas, from the atmosphere. Yet, we have known surprisingly little about these microscopic organisms. As we discover answers to some long-standing puzzles about the roles that marine microorganisms play in supporting the ocean’s food webs and driving global elemental cycles, we realized that we still need to learn much more about what these organisms do and how they do it—including how they evolved and contribute to our ocean's health and productivity.

The Marine Microbiology Initiative seeks to gain a comprehensive understanding of marine microbial communities, including their diversity, functions and behaviors; their ecological roles; and their origins and evolution. Our focus has been to enable researchers to uncover the principles that govern the interactions among microbes and that govern microbially mediated nutrient flow in the sea. To address these opportunities, we support leaders in the field through investigator awards, multidisciplinary team research projects, and efforts to create resources of broad use to the research community. We also support development of new instrumentation, tools, technologies and genetic approaches.

Through the efforts of many scientists from around the world, the initiative has been catalyzing new science through advances in methods and technology, and to reduce interdisciplinary barriers slowing progress. With our support, researchers are quantifying nutrient pools in the ocean, deciphering the genetic and biochemical bases of microbial metabolism, and understanding how microbes interact with one another. The initiative has five grant portfolios:

Individual investigator awards for current and emerging leaders in the field.
Multidisciplinary projects that support collaboration across disciplines.
New instrumentation, tools and technology that enable scientists to ask new questions in ways previously not

possible.
Community resource efforts that fund the creation and sharing of data and the development of tools, methods and infrastructure of widespread utility.
Projects that advance genetic tools to enable development of experimental model systems in marine microbial ecology.
We also bring together scientists to discuss timely subjects and to facilitate scientific exchange.

Our path to marine microbial ecology was a confluence of new technology that could accelerate science and an opportunity to support a field that was not well funded relative to potential impact. Around the time we began this work in 2004, the life sciences were entering a new era of DNA sequencing and genomics, expanding possibilities for scientific research – including the nascent field of marine microbial ecology. Through conversations with pioneers inside and outside the field, an opportunity was identified: to apply these new sequencing tools to advance knowledge of marine microbial communities and reveal how they support and influence ocean systems.

After many years of success, we will wind down this effort and close the initiative in 2021. We will have invested more than $250 million over 17 years to deepen understanding of the diversity, ecological activities and evolution of marine microbial communities. Thanks to the work of hundreds of scientists and others involved with the initiative, the goals have been achieved and the field has been profoundly enriched; it is now positioned to address new scientific questions using innovative technologies and methods.

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Funding

Funding Source	Award
Gordon and Betty Moore Foundation: Marine Microbiology Initiative (MMI)	GBMF3782
NSF Division of Ocean Sciences (NSF OCE)	OCE-1736599

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