

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)

| Contributors | Affiliation | Role |
|-------------------------------|---|------------------------|
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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.

Acquisition Description

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 $\mu\text{mol quanta m}^{-2} \text{s}^{-1}$ PAR). Northeast Atlantic *Phaeodactylum tricornutum* CCMP632 cultures were maintained in an 18°C incubator under constant fluorescent lighting (90 $\mu\text{mol quanta m}^{-2} \text{s}^{-1}$ PAR). Both Pacific *Pseudo-nitzschia delicatissima* UNC1205 and Pacific *Thalassiosira* sp. UNC1203 were grown in an 18°C incubator under constant fluorescent lighting (85 $\mu\text{mol quanta m}^{-2} \text{s}^{-1}$ 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 milli-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, 0.2 μm -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 NaNO₃, 41.5µM NaH₂PO₄, and 106µM Na₂SiO₃ 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 FeCl₃, 4.8 x 10⁻⁸ M MnCl₂, 4.0 x 10⁻⁸ M CuSO₄, 10⁻⁷ M NiCl₂, and 10⁻⁸ M Na₂O₃Se within a 10⁻⁴ M ethylenediamine tetraacetic acid disodium salt (EDTA, Acros Organics, C₁₀H₁₄N₂Na₂O₈) 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 [Zn²⁺] and [Co²⁺] 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

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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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 Zn ²⁺ ion concentration in media. [Zn ²⁺] | moles per liter (mol/L) |
| log_Co2plus | Log of calculated free Co ²⁺ ion concentration in media. [Co ²⁺] | 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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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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