Available online at www.sciencedirect.com

SCIENCE DIRECT®

Progress in Oceanography 64 (2005) 167–187

Progress in Oceanography

www.elsevier.com/locate/pocean

Responses of phytoplankton and heterotrophic bacteria in the northwest subarctic Pacific to in situ iron fertilization as estimated by HPLC pigment analysis and flow cytometry

Koji Suzuki [a,*], Akira Hinuma [b], Hiroaki Saito [c], Hiroshi Kiyosawa [d], Hongbin Liu [e], Toshiro Saino [b,f], Atsushi Tsuda [g]

[a] *Graduate School of Environmental Earth Science, Hokkaido University, Kita-ku, Sapporo 060-0810, Japan*
[b] *Hydrospheric Atmospheric Research Center, Nagoya University, Chikusa-ku, Nagoya 464-8601, Japan*
[c] *Tohoku National Fisheries Research Institute, Shinhama-cho 3-27-5, Shiogama 985-0001, Japan*
[d] *Marine Biological Research Institute, Shinagawa-ku, Tokyo 142-0042, Japan*
[e] *Louisiana Universities Marine Consortium, 8124 Hwy 56, Chauvin, LA 70344, USA*
[f] *CREST, Japan Science Technology Corporation, Nagoya University, Chikusa-ku, Nagoya 464-8601, Japan*
[g] *Ocean Research Institute, University of Tokyo, Nakano-ku, Tokyo 164-8639, Japan*

Available online 7 April 2005

Abstract

To verify the hypothesis that the growth of phytoplankton in the Western Subarctic Gyre (WSG), which is located in the northwest subarctic Pacific, is suppressed by low iron (Fe) availability, an in situ Fe fertilization experiment was carried out in the summer of 2001. Changes over time in the abundance and community structure of phytoplankton were examined inside and outside an Fe patch using phytoplankton pigment markers analyzed by high-performance liquid chromatography (HPLC) and flow cytometry (FCM). In addition, the abundance of heterotrophic bacteria was also investigated by FCM. The chlorophyll *a* concentration was initially ca. 0.9 $\mu g\,l^{-1}$ in the surface mixed layer where diatoms and chlorophyll *b*-containing green algae (prasinophytes and chlorophytes) were predominant in the chlorophyll biomass. After the iron enrichment, the chlorophyll *a* concentration increased up to 9.1 $\mu g\,l^{-1}$ in the upper 10 m inside the Fe patch on Day 13. At the same time, the concentration of fucoxanthin (a diatom marker) increased 45-fold in the Fe patch, and diatoms accounted for a maximum 69% of the chlorophyll biomass. This result was consistent with a microscopic observation showing that the diatom *Chaetoceros debilis* had bloomed inside the Fe patch. However, chlorophyllide *a* concentrations also increased in the Fe patch with time, and reached a maximum of 2.2 $\mu g\,l^{-1}$ at 5 m depth on Day 13, suggesting that a marked abundance of senescent algal cells existed at the end of the experiment. The concentration of peridinin (a dinoflagellate marker) also reached a maximum 24-fold, and dinoflagellates had contributed significantly (>15%) to the chlorophyll biomass inside the Fe patch by the end of the experiment. Concentrations

* Corresponding author. Tel.: +81 11 706 2370; fax: +81 11 706 2247.
E-mail address: kojis@ees.hokudai.ac.jp (K. Suzuki).

doi:10.1016/j.pocean.2005.02.007

of 19′-hexanoyloxyfucoxanthin (a prymnesiophyte marker), 19′-butanoyloxyfucoxanthin (a pelagophyte marker), and alloxanthin (a cryptophyte marker) were only incremented a few-fold increment inside the Fe patch. On the contrary, chlorophyll *b* concentration reduced to almost half of the initial level in the upper 10 m water column inside the Fe patch at the end of the experiment. A decrease with time in the abundance of eukaryotic ultraphytoplankton (<ca. 5 μm in size), in which chlorophyll *b*-containing green algae were possibly included was also observed by FCM. Overall, our results indicate that Fe supply can dramatically alter the abundance and community structure of phytoplankton in the WSG. On the other hand, cell density of heterotrophic bacteria inside the Fe patch was maximum at only ca. 1.5-fold higher than that outside the Fe patch. This indicates that heterotrophic bacteria abundance was little respondent to the Fe enrichment.

Keywords: HNLC; Iron; Phytoplankton; Heterotrophic bacteria; Pigments; Subarctic Pacific

1. Introduction

The northwest subarctic Pacific including Oyashio waters is sometimes more productive in its lower trophic levels than the northeast subarctic Pacific (e.g. Obayashi et al., 2001; Taniguchi, 1999). However, it is also evident that chlorophyll *a* levels around the center of the northwestern subarctic Pacific (i.e., Western Subarctic Gyre: WSG) is generally as low as in the northeast subarctic Pacific (e.g. Sasaoka et al., 2002), which is one of the high-nitrate, low chlorophyll (HNLC) regions (Harrison et al., 1999). Martin and Fitzwater (1988) proposed for the first time that phytoplankton growth in the northeast subarctic Pacific was limited by low levels of iron (Fe) in seawater, and thereby chlorophyll *a* concentration is kept low. Recently, Suzuki et al. (2002a) pointed out that there is an east–west gradient in the photosynthetic potential of phytoplankton in the subarctic Pacific during early summer, and that the difference is caused by Fe levels in seawater. The results of Suzuki et al. (2002a) are consistent with the east–west difference in atmospheric Fe flux into the subarctic Pacific (Duce & Tindale, 1991). In addition, Nishioka et al. (2003) showed that higher acid-labile, particulate Fe concentrations towards the west, though dissolved Fe levels were almost the same between the northeast and northwest subarctic Pacific. These data indicate that Fe that may well control the photosynthetic physiology of phytoplankton, and Fe supply has a crucial effect on biogeochemical cycles in the whole subarctic Pacific Ocean.

To verify the Fe hypothesis, an in situ iron fertilization experiment, Subarcitic Pacific Iron Experiment for Ecosystem Dynamics Study (SEEDS) was carried out in the WSG during the summer of 2001 (Tsuda et al., 2003). In previous in situ Fe enrichment experiments conducted in other HNLC regions, namely, the eastern equatorial Pacific (Coale et al., 1996; Martin et al., 1994) and the Southern Ocean (Boyd et al., 2000; Gervais, Riebsell, & Gorbunov, 2002), remarkable increases in chlorophyll *a* concentrations occurred with dramatic floristic shifts to pennate diatoms by the addition of Fe. Such phytoplankton response might lead to an increase in the flux of sinking particulate organic carbon, that is, the sequestration of CO_2 into the deep ocean. On the other hand, the proliferation of diatoms may also have serious negative consequences for the ecosystems (Adhiya & Chisholm, 2001). For example, zooplankton reproduction can be inhibited by the predominance of diatoms (Ban et al., 1997). Some pennate diatoms of the genus *Pseudo-nitzchia*, are known to produce a strong neurotoxin domoic acid and may influence marine ecosystems (Scholin et al., 2000), although domoic acid may function as a binding ligand for trace metals and increase the bioavailability of Fe for the algae (Rue & Bruland, 2001).

Other consequences of in situ Fe fertilization can be expected. In the previous in situ Fe fertilization experiments, an increase in the concentration of dimethylsulfide (DMS), which is produced mainly by prymnesiophytes and dinoflagellates (Scarratt et al., 2002), was also observed as a common phenomenon. DMS is a key chemical compound in the global sulfur cycle, and its oxidation products can influence atmospheric