

## Effects of iron concentration on pigment composition in *Phaeocystis antarctica* grown at low irradiance

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**Abstract** Interpretation of photosynthetic pigment data using iterative programs such as CHEMTAX are widely used to examine algal community structure in the surface ocean. The accuracy of such programs relies on understanding the effects of environmental parameters on the pigment composition of taxonomically diverse algal groups. *Phaeocystis antarctica* is an important contributor to total autotrophic production and the biogeochemical cycling of carbon and sulfur in the Southern Ocean. Here we report the results of a laboratory culture experiment in which we examined the effects of ambient dissolved iron concentration on the pigment composition of colonial *P. antarctica*, using a new *P. antarctica* strain isolated from the southern Ross Sea in December 2003. Low-iron (<0.2 nM dissolved Fe) filtered Ross Sea seawater was used to prepare the growth media, thus allowing subnanomolar iron additions without the use of EDTA to control dissolved iron concentrations. The experiment was conducted at relatively low

irradiance ( $\sim 20 \mu\text{E m}^{-2} \text{s}^{-1}$ ), with *P. antarctica* primarily present in the colonial form—conditions that are typical of the southern Ross Sea during austral spring. Relative to the iron-limited control treatments (0.22 nM dissolved Fe), iron addition mediated a decrease in the ratio of 19'-hexanoyloxyfucoxanthin to chlorophyll *a*, and an increase in the ratio of fucoxanthin to chlorophyll *a*. Our results also suggest that the ratio of 19'-hexanoyloxyfucoxanthin to chlorophyll *c*<sub>3</sub> (Hex:Chl *c*<sub>3</sub> ratio) may be a characteristic physiological indicator for the iron-nutritional status of colonial *P. antarctica*, with higher Hex:Chl *c*<sub>3</sub> ratios (>3) indicative of Fe stress. We also observed that the ratio of fucoxanthin to 19'-hexanoyloxyfucoxanthin (Fuco:Hex ratio) was highly correlated ( $r^2 = 0.82$ ) with initial dissolved Fe concentration, with Fuco:Hex ratios <0.05 measured under iron-limited conditions (dissolved Fe <0.45 nM). Our results corroborate and extend the results of previous experimental studies, and, combined with pigment measurements from the southern Ross Sea, are consistent with the hypothesis that the interconversion of fucoxanthin and 19'-hexanoyloxyfucoxanthin by colonial *P. antarctica* is used as a photo-protective or light-harvesting mechanism, according to the availability of dissolved iron.

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## Introduction

*Phaeocystis antarctica* is an important phytoplankton species in the Southern Ocean, although the relative contribution of *Phaeocystis* to total autotrophic biomass and primary production in this vast oceanic region is poorly constrained, for a number of reasons. First, *P. antarctica* has a complex heteromorphic life cycle, alternating between gelatinous colonies and free-living flagellate cells, which hampers identification using simple microscopy (Rousseau et al. 1994). Second, blooms of *P. antarctica* display pronounced temporal and spatial variability, especially near the Antarctic continental margins (El-Sayed et al. 1983; Palmisano et al. 1986). And finally, although numerous studies have focused on this species in near-surface waters, *P. antarctica* may also dominate algal biomass in the lower euphotic zone in the Southern Ocean (DiTullio et al. 2003a, b). In addition, the role of *P. antarctica* in the biogeochemical cycling of carbon, sulfur and nutrient elements in the Southern Ocean remains to be elucidated. Studies on the Antarctic continental shelf suggest that *P. antarctica* plays a significant role in the regional cycling of carbon (Schoemann et al. 2005; DiTullio et al. 2000) and sulfur (Gibson et al. 1990; DiTullio and Smith 1995), as well as impacting the stoichiometry of macronutrient cycling in Southern Ocean waters (Arrigo et al. 1999; Arrigo 2005). Thus there are compelling reasons to improve our understanding of the abundance, distribution and growth requirements of *P. antarctica*.

One of the primary methods for determining algal community structure in polar waters is based on the analysis of photosynthetic pigments by high-performance liquid chromatography (HPLC). Chemotaxonomic interpretations of HPLC data using programs such as CHEMTAX (Mackey et al. 1996) rely on the input of environmentally representative initial pigment ratios for taxonomically diverse phytoplankton groups. To apply such chemotaxonomic methods to *P. antarctica* in the Southern Ocean thus requires the use of laboratory culture studies in order to establish representative pigment ratios for this species under various growth conditions. Previous studies (e.g., Buma et al. 1991) have demonstrated that *P. antarctica*

contains the two accessory pigments 19'-hexanoyloxyfucoxanthin (Hex) and fucoxanthin (Fuco), although diatoms, also common in the Southern Ocean, are also known to contain Fuco. The presence of Hex in significant concentrations relative to other photosynthetic pigments has been used to identify *P. antarctica* blooms in Antarctic waters (e.g., DiTullio and Smith 1995; Crocker et al. 1995), and a strong correlation has been observed between Hex concentrations and *P. antarctica* cell number estimated by microscopy in the southern Ross Sea (DiTullio et al. 2003b), which is dominated by *P. antarctica* blooms during the austral spring (November–December; El-Sayed et al. 1983; Arrigo et al. 1998; Smith et al. 2003).

Previous laboratory culture experiments have shown that relative concentrations of Fuco and Hex in *P. antarctica* vary as a function of ambient dissolved Fe concentration in the growth media (van Leeuwe and Stefels 1998). However, these experiments were performed under relatively high dissolved Fe concentrations (nM– $\mu$ M), compared to open Antarctic surface ocean waters, and, as in most other culture studies, the synthetic organic chelating compound EDTA was used to buffer free dissolved Fe concentrations. Gerringa et al. (2000) have presented a strong argument for the need to conduct such culture experiments using realistically low dissolved iron concentrations without the use of synthetic chelating compounds such as EDTA. Following these suggestions, we have attempted to examine the effect of iron availability on the growth and pigment composition of *P. antarctica*, using culture experiments conducted at low (i.e., sub-nanonolar) dissolved iron concentrations and without using EDTA (or other chelators) to control dissolved iron levels in the growth media. These experiments were performed using a new strain of *P. antarctica*, which was isolated from native phytoplankton collected in the southern Ross Sea during December 2003. Here we report the results of an experiment that examines the effect of sub-nanomolar iron additions on the pigment composition of colonial *P. antarctica* under relatively low irradiance—conditions that are typical of the southern Ross Sea during early austral spring—and discuss the ecological