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Robert J. Olson

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BIOLOGY DEPARTMENT
WOODS HOLE OCEANOGRAPHIC INSTITUTION
WOODS HOLE, MA 02543

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DR. ERIC SHULENBERGER, CODE 322BC
ONR, 800 N. Quincy Street
Arlington, VA 22217-5000

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Vertical mixing in the surface layer of the ocean will affect phytoplankton growth by changing the light field to which cells are exposed. Conversely, indicators of phytoplankton photoacclimation should be diagnostic of mixing processes. A combination of laboratory and field experimental work, field observations, and theoretical models were used to quantify the relationship between vertical mixing and photoacclimation in determining the time and space evolution of single cell optical properties for the photosynthetic picoplankton, *Prochlorococcus* spp. Diel time-series observations from the Sargasso Sea revealed patterns in single-cell fluorescence distributions within *Prochlorococcus* spp. populations which correspond to decreasing mixing rates and photoacclimation during the day, and increased mixing at night. Reciprocal light shift experiments were used to quantify the photoacclimation kinetics for *Prochlorococcus* spp. fluorescence. In addition, a continuous culture system was developed which could simulate the effects of mixing across a light gradient at the level of the individual cell. When this system was operated at four different simulated diffusivities, *Prochlorococcus marinus* strain Med4 fluorescence distributions showed distinct patterns in the mean and higher moments which are consistent with a simple quasi-steady turbulent diffusion-photoacclimation model. Daytime photoacclimation drove the development of a gradient in mean fluorescence, a decrease in variance overall, and skewing of distributions away from the boundaries. These results suggest that picophytoplankton single-cell fluorescence distributions could prove to be a useful diagnostic indicator of the mixing environment.

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Final Report, ONR grant N00014-89-J-1110, Understanding Vertical Mixing and Photoacclimation Processes in the Surface Oceans Using Single Cell Analysis

Robert J. Olson
Biology Department, MS 32
Woods Hole Oceanographic Institution
Woods Hole, MA 02543-1049
phone: 508 289-2565
email: rolson@whoi.edu

Sallie W. Chisholm
Parsons Laboratory, 48-425
Massachusetts Institute of Technology
Cambridge, MA 02139
phone: 617 253-1771
email: pchisholm@mit.edu

Summary of Project

Vertical mixing in the surface layer of the ocean will affect phytoplankton growth by changing the light field to which cells are exposed. Conversely, indicators of phytoplankton photoacclimation should be diagnostic of mixing processes. A combination of laboratory and field experimental work, field observations, and theoretical models were used to quantify the relationship between vertical mixing and photoacclimation in determining the time and space evolution of single cell optical properties for the photosynthetic picoplankton, *Prochlorococcus* spp. Diel time-series observations from the Sargasso Sea revealed patterns in single-cell fluorescence distributions within *Prochlorococcus* spp. populations which correspond to decreasing mixing rates and photoacclimation during the day, and increased mixing at night. Reciprocal light shift experiments were used to quantify the photoacclimation kinetics for *Prochlorococcus* spp. fluorescence. In addition, a continuous culture system was developed which could simulate the effects of mixing across a light gradient at the level of the individual cell. When this system was operated at four different simulated diffusivities, *Prochlorococcus marinus* strain Med4 fluorescence distributions showed distinct patterns in the mean and higher moments which are consistent with a simple quasi-steady turbulent diffusion-photoacclimation model. Daytime photoacclimation drove the development of a gradient in mean fluorescence, a decrease in variance overall, and skewing of distributions away from the boundaries. These results suggest that picophytoplankton single-cell fluorescence distributions could prove to be a useful diagnostic indicator of the mixing environment.

This project supported the Ph.D. thesis research of Jeffrey Dusenberry in the MIT/WHOI Joint Program in Biological Oceanography, and the bulk of the results are embodied in this research. The thesis was completed in February 1995 and four papers are now in preparation for submission to oceanographic journals. These papers are all being prepared now, rather than over the course of the project, because they are inextricably dependent on one another. The abstracts of the thesis and of these papers follows. Other related papers which were supported at least in part by the project are listed in the Citations section.

Picophytoplankton Photoacclimation and Mixing in the Surface Oceans

Jeffrey A. Dusenberry

(Ph.D. Thesis, MIT/WHOI Joint Program in Biological Oceanography, February 1995)

Abstract

Fluctuations in light intensity due to vertical mixing in the open ocean surface layer will affect phytoplankton physiology. Conversely, indicators of phytoplankton photoacclimation will be diagnostic of mixing processes if the appropriatê kinetics are known. A combination of laboratory and field experimental work, field observations, and theoretical models were used to quantify the relationship between vertical mixing and photoacclimation in determining the time and space evolution of single cell optical properties for the photosynthetic picoplankton, *Prochlorococcus* spp. Diel time-series observations from the Sargasso Sea reveal patterns in single-cell fluorescence distributions within *Prochlorococcus* spp. populations which appear to correspond to decreasing mixing rates and photoacclimation during the day, and increased mixing at night. Reciprocal light shift experiments were used to quantify the photoacclimation kinetics for *Prochlorococcus* spp. fluorescence.

A laboratory continuous culture system was developed which could simulate the effects of mixing across a light gradient at the level of the individual cell. This system was operated at four different simulated diffusivities. *Prochlorococcus marinus* strain Med4 fluorescence distributions show distinct patterns in the mean and higher moments which are consistent with a simple quasi-steady turbulent diffusion-photoacclimation model. In both, daytime photoacclimation drove the development of a gradient in mean fluorescence, a decrease in variance overall, and skewing of distributions away from the boundaries. These results suggest that picophytoplankton single-cell fluorescence distributions could prove to be a useful diagnostic indicator of the mixing environment.

Thesis Advisors: Sallie W. Chisholm and Robert J. Olson

Response of the photosynthetic picoplankter *Prochlorococcus* spp. to a 24-hour photoperiod and vertical mixing: I. Field observations.

J. A. Dusenberry, R. J. Olson and S. W. Chisholm

Two diel time-series sampling schemes were undertaken to quantify the effects of changing mixing dynamics on picophytoplankton optical properties. Both time-series show a shoaling of the mixed layer due to surface warming and a rain-formed mixed layer. These dynamics coupled with the diurnal cycle of solar irradiance, drove the development of a gradient in mean red fluorescence of *Prochlorococcus* spp. via photoacclimation. In addition, the distribution of fluorescence within field populations responds to changing mixing and photoacclimation dynamics, with photoacclimation in the absence of strong mixing generally resulting in reduced variance in fluorescence within sample populations. Nighttime mixing in the absence of photoacclimation reversed this process and resulted in increased variation of single-cell fluorescence. Both the effects of physical boundaries and hysteresis in photoacclimation appear to affect the third moment (skewness) of fluorescence, with boundaries causing optical properties to be skewed away from the boundary and hysteresis causing overall negative skewness. These observations show that the mean and variance, and possibly the higher moments, of single-cell optical properties reflect the physical dynamics, and should yield useful information regarding the light history of the population.

Response of the photosynthetic picoplankter *Prochlorococcus* spp. to a 24-hour photoperiod and vertical mixing: II. Determination of photoacclimation kinetics.

J. A. Dusenberry, S.W. Chisholm and R.J. Olson

To use picophytoplankton as tracers for vertical mixing, an appropriate index of photoacclimative state and the kinetics of that parameter must be defined. To determine the relevant kinetics, we did several time-series of reciprocal light shifts to *Prochlorococcus marinus* strain Med4 or natural populations of *Prochlorococcus* spp. These reveal diel patterns in fluorescence and light scatter due to phasing of cell division to the daily photoperiod; division occurs preferentially during the early part of the dark period. Laboratory experiments suggest that red fluorescence can be normalized to the cube root of forward angle light scatter to remove the signal due to phased growth and division. This normalized fluorescence may thus be an appropriate indicator of photoacclimation in *Prochlorococcus* spp. A logistic model for photoacclimation (Cullen and Lewis, 1988) was found to fit the resulting time-series well, with the most pronounced deviation from this model in the populations shifted to high irradiances. Photoacclimative rates ranged from 0.9 to 3 d^{-1}, with the highest rate observed in larger *Prochlorococcus* spp. The smaller *Prochlorococcus* yield rates that decrease with increasing temperature within the range studied (24 - 26 °C).

Response of the photosynthetic picoplankter *Prochlorococcus* spp. to a 24-hour photoperiod and vertical mixing: III. Single-cell based experimental and theoretical analysis.

J. A. Dusenberry, S. W. Chisholm and R. J. Olson

In order to quantify the effects of mixing across a light gradient on the mean and higher moments of picophytoplankton single-cell photoacclimative properties , laboratory simulations were carried out using a mixostat apparatus. This apparatus, operating on principles often used in chemostats and cyclostats, is a continuous culture system which is capable of simulating a random walk across a light gradient. This random walk is simulated at the individual cell level, so that the effects of mixing should be evident in the distribution of photoacclimative properties within a population.

Laboratory simulations of four different mixing rates yield characteristic diel patterns (10:14 light:dark cycle) in the mean and variance of normalized red fluorescence of the picophytoplankter *Prochlorococcus marinus* strain Med4. All four simulations show increasing gradients in the mean normalized red fluorescence during the daytime due to photoacclimation, and a breakdown of this gradient during the dark due to mixing. The corresponding variance also decreases during the day as a result of photoacclimation, and increases at night as the system is homogenized.

The system was modelled using a simple time-dependent photoacclimation diffusion model. Model results for the mean and the variance were similar to those found in the mixostat. These results provide a qualitative basis for interpreting field observations of picophytoplankton optical properties with respect to the mixing environment.

Steady State Single-Cell Model Simulations of Photoacclimation in a Vertically Mixed Layer

J. A. Dusenberry

Model simulations of the effects of vertical mixing and photoacclimation were done for both first-order and logistic representations of photoacclimation kinetics, in order to quantify the effects of vertical mixing and/or hysteresis on the higher moments of a photoacclimative parameter. Steady state was assumed as to simplify the model and provide a starting point for further investigations. In the simple mixed layer modelled here, hysteresis (as represented by the logistic model of photoacclimation) yields populations which are more surface acclimated at high mixing rates than the simulation without hysteresis. In both models, patterns in the mean value of a photoacclimative parameter show a simple gradient at slow mixing relative to photoacclimation which breaks down with increasing mixing and as the mixed layer approaches homogeneity. Corresponding to this pattern, the variance and higher moments of the photoacclimative parameter all show trends which are diagnostic of mixing rates or boundary effects.

Published papers which acknowledge this grant in part:

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