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Seasonal variability in the phytoplankton community of the North Pacific Subtropical Gyre

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Abstract. Time series measurements of in situ fluorescence, extracted particulate chlorophyll *a*, primary productivity, extracted adenosine 5′-triphosphate, and fluorescence per cell, as measured by flow cytometry, demonstrate seasonal cycles in fluorescence and chlorophyll concentrations in the North Pacific Subtropical Gyre (22° 45′N, 158° 00′W). Two opposing cycles are evident. In the upper euphotic zone (0-50 m), chlorophyll *a* concentrations increase in winter, with a maximum in December, and decrease each summer, with a minimum in June or July. In contrast, chlorophyll *a* concentrations in the lower euphotic zone (100-175 m) increase in spring, with a maximum in May, and decline in fall, with a minimum in October or November. The winter increase in chlorophyll *a* concentration in the upper 50 m of the water column appears to be a consequence of photoadaptation in response to decreased average mixed-layer light intensity rather than a change in phytoplankton biomass. In the lower euphotic zone, however, the seasonal cycle in pigment concentration does reflect a change in the rate of primary production and in phytoplankton biomass as a consequence of increased light intensity in summer. These observations have important implications for phytoplankton dynamics in the subtropical oceans and for remote sensing of phytoplankton biomass.

Introduction

Seasonal variations in light, temperature, and turbulence produce pronounced annual cycles in the epipelagic zone at middle to high latitudes. A combination of surface water cooling and wind-driven mixing brings nutrients from below the euphotic zone into the surface ocean during winter. During spring, when mixing decreases and phytoplankton are retained in illuminated surface waters, phytoplankton production and biomass increase rapidly in response to increased light and nutrient availability [*Sverdrup*, 1953]. This pattern of seasonal variability, with a pronounced spring bloom, has been well documented [e.g., *Menzel and Ryther*, 1960; *Lohrenz et al.*, 1992a; *Ducklow and Harris*, 1993].

Seasonality in the physical forces that control phytoplankton growth in the upper ocean is dampened with decreasing latitude. As a consequence, seasonality in phytoplankton dynamics is also believed to decline with decreasing latitude. Although simple models suggest that a cycle in phytoplankton biomass and production should be observed in the subtropical oceans [*Cushing*, 1959], evidence for the presence of seasonal cycles in phytoplankton production and biomass at latitudes lower than 30^0 is equivocal.

Seasonality in phytoplankton biomass and production in the subtropical oceans was not observed until the late 1960s (Indian Ocean [*Jitts*, 1969]; eastern tropical Pacific Ocean, [*Owen and Zeitzschel*, 1970]). Subsequent studies, however, have failed to confirm a seasonal cycle in the subtropics. Many of these latter studies were conducted at or near the "Climax" site at approximately 28^0N, 155^0W in the North Pacific Ocean and suggested that variability in phytoplankton pigment concentrations or biomass is stochastic and is controlled primarily by periodic mixing events [*McGowan and Hayward*, 1978; *Hayward et al.*, 1983; *Hayward*, 1987] or by inputs of iron and nitrate from the atmosphere [*DiTullio and Laws*, 1991; *Young et al.*, 1991]. However, these investigations have been hampered by infrequent visits to this remote location, particularly in winter [*Hayward*, 1987]. *Bienfang* [1981] and *Bienfang et al.* [1984] were unable to detect a seasonal cycle in chlorophyll *a* (chl *a*) or in primary production during an 18-month coastal time series study in the subtropical North Pacific near Hawaii. In contrast, satellite observations have recently indicated that the chl *a* concentrations estimated from coastal zone color scanner imagery vary approximately twofold in the subtropical oceans, with a maximum in winter [*Yoder et al.*, 1993]. These observations have been interpreted as evidence for a seasonal cycle in phytoplankton biomass which is consistent with the early model developed by *Cushing* [1959]. In addition, *Venrick* [1993] has reevaluated 17 years of phytoplankton data collected at the Climax site and has concluded that an annual cycle is indeed present in the North Pacific Subtropical Gyre (NPSG). She proposed that winter mixing transports cells normally found near the base of the euphotic zone to the near-surface waters and that this stimulates their growth during winter. *Letelier et al.* [1993], in a study of phytoplankton pigment distributions in the NPSG,

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attributed elevated chl *a* concentrations within the surface mixed layer to photoadaptation in response to the deepening mixed layer during winter months. Furthermore, *Letelier et al.* [1993] provided evidence for a seasonal cycle in chl *a* concentrations at the depth of the chlorophyll maximum layer, an effect that they attributed to increased light intensity in spring.

The inconsistency in the observations discussed above may be due, in part, to undersampling of the subtropical oceans. These areas are the most sparsely sampled regions of the world's oceans [*Blackburn*, 1981]. Typically, studies of phytoplankton biomass and production use chl *a* concentrations and rates of primary production from as few as six depths in the surface ocean. Given that seasonal changes at these latitudes are expected to be relatively small and that samples are typically collected from only a few depths at infrequent intervals, it is possible that the seasonal cycles in the subtropical oceans have not been resolved clearly because of inadequate sampling intensity.

Since October 1988, scientists in the Hawaii Ocean Time-series (HOT) Program have been making approximately monthly observations of the physical, chemical, and biological conditions at a permanent station north of the Hawaiian archipelago. The purpose of this program is to assess annual and interannual variability in the NPSG [*Karl and Winn*, 1991; *Letelier et al.*, 1993; *Winn et al.*, 1994; Karl, D.M., and R.L. Lukas, The Hawaii Ocean Time-series (HOT) Program: Background, rationale and field implementation, submitted to *Deep Sea Research, part II*, 1995; hereinafter referred to as Karl and Lukas, submitted manuscript, 1995]. We have used continuous in situ flash fluorescence, in combination with discrete measurements of chl *a*, primary production, adenosine 5′-triphosphate (ATP), and cellular fluorescence as measured by flow cytometry, to examine temporal variability at station ALOHA (A Long-term Oligotrophic Habitat Assessment).

Materials and methods

Station Location and Sampling Schedule

The time series measurements summarized here encompass the period from October 1988 to December 1993. Samples were collected at station ALOHA, located at 22°45′N, 158°00′W, approximately 100 km north of Oahu, Hawaii (Figure 1). Standard hydrographic sampling was conducted using a Sea-Bird (model SBE-09) CTD (Conductivity, Temperature, and Depth) [Karl and Lukas, submitted manuscript, 1995]. Samples were collected with PVC (polyvinyl chloride) sampling bottles attached to an aluminum CTD-rosette system [*Karl and Winn*, 1991].

Flash Fluorescence

Flash fluorescence traces were obtained with a Sea Tech (model ST-0250) flash fluorometer (ex = 425 nm, em = 625 nm) attached to the rosette and integrated with the Sea-Bird CTD. Data were collected at 24 Hz and averaged to 2 Hz. The data were then averaged in 2-dbar bins. On each cruise, 10 to 15 fluorescence profiles were collected as part of the CTD "burst" sampling strategy [*Karl and Winn*, 1991; Karl and Lukas, submitted manuscript, 1995]. Only fluorescence profiles collected at night (2000 to 0400 LT) were used in our analysis, however, to avoid light-dependent inhibition of chlorophyll fluorescence [*Lewis et al.*, 1984a; *Falkowski and Kiefer*, 1985] that would otherwise affect our data set.

The flash fluorometer was serviced by the manufacturer on a regular basis; the instrument response changed slightly each time it was returned. We normalized the instrument response for each HOT cruise using the voltage derived at two depth intervals: 400-450 m and 900-1000 m. These depths were used because the voltage output at these depth intervals remained constant over time and changed only when the instrument was serviced. To assess changes in instrument response between servicings, we also determined the relative response of the fluorometer between each HOT cruise using fluorescent plastic sheeting. The sheeting was placed at a fixed distance (approximately 2 cm) from the instrument lamp in the dark, and the output voltage was recorded. This procedure was used to confirm that the instrument was performing properly before each cruise.

Chlorophyll Concentrations

Water column chl *a* concentrations were determined by both standard fluorometric techniques [*Strickland and Parsons*, 1972] and by high-pressure liquid chromatography (HPLC) [*Bidigare et al.*, 1989]. Typically, pigment analyses were made on samples collected from at least 11 depths each month; however, a greater number of fluorometric pigment measurements were made because of the relative ease of this assay. Samples for fluorometric analysis of pigment concentrations were collected in subdued light, concentrated by vacuum filtration on Whatman GF/F glass fiber filters, extracted in 100% acetone, and stored in the dark at -20°C for analysis in the laboratory. This procedure has been shown to yield reliable estimates of total chl *a* at station ALOHA despite the ubiquity of small picoplankton [*Chavez et al.*, 1995]. Samples for HPLC analysis (4 -10 L) were filtered onto GF/F glass fiber filters by positive pressure (4-8 psi N_2), extracted in 100% acetone, and analyzed by reverse phase HPLC with the minor modifications described by *Letelier et al.* [1993].

Nitrate Plus Nitrite

Nutrient samples collected at sea were frozen immediately and stored for analysis at our shore-based laboratory. Inorganic nitrogen (nitrate plus nitrite) was measured using standard autoanalyzer techniques or using a low-level chemiluminescent technique. Autoanalyzer measurements were made on a four-channel Technicon Autoanalyzer II continuous system, after reduction of nitrate to nitrite in a copperized cadmium column, using slight modifications of the Technicon procedures for the analysis of seawater. Low-level nitrogen measurements were made in the upper 100 m of the water column using the chemiluminescent method of *Cox* [1980] as modified by *Garside* [1982]. Wherever possible, the low-level results were used in the analysis presented here.

Primary Productivity

The rate of primary production was determined with the ^{14}C technique using trace-metal clean methods [*Fitzwater et al.*, 1982]. Measurements were made at eight depths each month in the upper 175 m of the water column (R.M. Letelier et al., Temporal variability in photosynthetic carbon assimilation efficiencies at Station ALOHA, submitted to *Deep Sea Research., Part II, 1995*; hereinafter referred to as Letelier et al.,

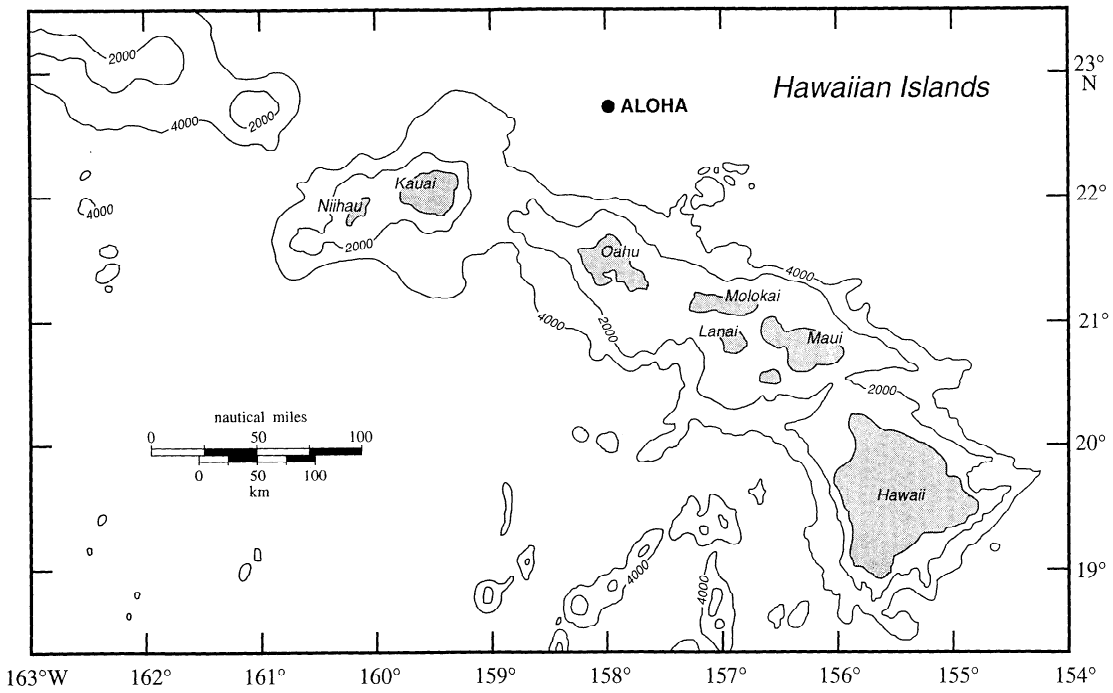


Figure 1. Location of the U.S.-World Ocean Circulation Experiment (WOCE) Joint Global Ocean Flux Study (JGOFS) Hawaii Ocean Time-series permanent station at 22° 45′N, 158° 00′W.

submitted manuscript, 1995). Subsamples (three light bottles and three dark bottles for each depth) were collected before dawn into 500-mL polycarbonate bottles from Teflon-coated Go-Flo bottles and kept in the dark until the incubation was begun. To each bottle, approximately 0.05 mCi of ^{14}C-HCO_3^- was added, and samples were incubated over the daylight period. At the end of the incubation, 200 μL of sample was preserved in β-phenylethylamine for the determination of total radioactivity. Specific activities (curies per mole) were calculated from total radioactivity and total inorganic carbon, the latter measured by CO_2 coulometry [*Winn et al.*, 1994]. The remainder of the sample was filtered onto Whatman GF/F glass fiber filters. These filters were subsequently acidified to drive off excess ^{14}C-HCO_3^- and measured by liquid scintillation counting.

From the first HOT cruise in October of 1988 until May 1990, incubations were done in an on-deck incubation system [*Lohrenz et al.*, 1992b] which simulated in situ light and temperature. Beginning in July of 1989, in situ incubations were conducted either in addition to or instead of the on-deck procedure. These incubations were done by suspending the incubation bottles from a free-floating array. The in situ incubation procedures were overlapped with the on-deck method for a total of eight cruises, in order to assess the consistency of these methods before the on-deck incubation procedure was discontinued.

Adenosine 5′-Triphosphate (ATP)

Adenosine 5′-triphosphate was measured using the firefly bioluminescence reaction [*Holm-Hansen*, 1973; *Karl and Holm-Hansen*, 1978]. These samples were filtered as soon as possible after the rosette arrived on deck. The samples were drawn through a 202-μm mesh to remove the zooplankton that would otherwise affect the precision of the analysis. Samples were filtered through 47-mm GF/F glass fiber filters, the filters were then extracted immediately in boiling TRIS (Tris[hydroxymethyl]aminomethane) buffer (0.02 *M*, *p*H 7.4 at 25°C) for approximately 5 min, and then the samples were frozen for subsequent analysis. At our shore-based laboratory, ATP was measured with firefly lantern extract (Sigma Chemical, FLE-50) on an ATP photometer manufactured by Biospherical Instruments, using peak height analysis.

Flow Cytometric Analysis of Chlorophyll Fluorescence per Cell

Samples for flow cytometric analysis were collected in duplicate, preserved with paraformaldehyde (0.2% vol/vol final concentration), and frozen in liquid nitrogen for analysis in the laboratory. Subsamples were analyzed using a Coulter (Hialeah, Florida) EPICS 753 flow cytometer equipped with dual 5-W Argon lasers and an automatic sampling device. The laser and filter setups were as previously described [*Campbell and Vaulot*, 1993; *Campbell et al.*, 1994]. *Prochlorococcus* and *Synechococcus* populations were discriminated on the basis of right-angle light scatter and pigment fluorescence. All fluorescence signals were normalized to 0.57-μm yellow-green Fluoresbrite beads (Polysciences) to maintain analytical consistency over time. Data were collected in list mode and then transferred to a personal computer and analyzed using software described by *Vaulot* [1989].

Data Analysis

The significance of the apparent seasonal cycles was assessed in two ways. First, the sum of the squares of the residuals (RSS) was calculated for the fit of the data to the function

$$X = s_x \sin((t/365)\,2\pi - \phi) + x \quad (1)$$

where t is days from January 1, x and s_x are the mean and standard deviation, respectively, of the data, and ϕ is a phase shift. The RSS was calculated for 365 values of ϕ from 0 to 2π (i.e., for intervals of 1 day). The value of ϕ that minimizes the RSS was designated ϕ_o, and the peak of the sine function occurs at $\phi_o + \pi/2$. To reduce the variability associated with changes on timescales greater than 1 year, we transformed the data to standard scores, or z scores [*Triola*, 1980], given by

$$z_i = (x_i - x_j)/s_j \quad (2)$$

where x_j and s_j are the mean and standard deviation, respectively, for the calendar year in which the cruise occurred. Once the numbers have been transformed to z scores, x=0 and s_x=1, so equation (1) reduces to

$$X = \sin((t/365)\;2\pi - \phi) \quad (3)$$

The RSS was then calculated relative to 10,000 random number sequences drawn from a normal (Gaussian) distribution with mean 0 and standard deviation 1, and the number of such sequences that gave an RSS less than that determined for the sine function (equation (3)) with $\phi=\phi_o$ was recorded.

To estimate the significance of these results, the above procedure was carried out with 1,000 random "data sets" (also Gaussian, with x=0 and s_x=1) to determine the probability that a given percentage of random number sequences would give a lower RSS than a sine curve fit to the data if the temporal variability was purely stochastic. If a given number of random number sequences returned lower RSS than the sine curve fit to the data, the probability that the temporal variability was purely stochastic is the number of sine curve fits to random number sequences that returned that number or less. The majority of variables consistently returned 0 (i.e., in 10,000 trials, no random number sequence gave a lower RSS); the probability of this was 0.058. In the analysis of our data, if the number of random number sequences that gave a lower RSS was greater than 0, a range of appropriate probabilities is given in Table 1. Because 0.058 is the minimum value possible, seasonality of a given variable can never be said to be statistically significant by this method alone. However, the probability of a large number of variables returning values in the 0.05-0.10 range is the product of those probabilities.

The second approach was to divide the data (again transformed into z scores) into two 4-month periods corresponding to "winter" and "summer" seasons centered on dates separated by 6 months (i.e., exactly one-half year out of phase), with the intervening "spring" and "fall" data excluded, and to compare seasonal means by one-way ANOVA (Analysis of Variance). This was done for three possible definitions of the seasons, corresponding to winter being January through April, December through March, or November through February.

Results

Representative Contour Plots

To examine our data set for the presence of seasonal cycles, we first contoured these data to provide a visual representation of the first 5 years of time series observations. Representative contour plots of flash fluorescence, HPLC chl *a*, and primary production show regular seasonal oscillations (Plates 1, 2, and 3). Both flash fluorescence and HPLC chl *a*, for example, show seasonal oscillations in the 0- to 50-m depth interval, with a minimum near July (Plates 1 and 2). In the 100- to 175-m interval, the oscillation appears approximately opposite in phase,

Table 1. Phase Shifts and Probability Levels for Fitting of Sinusoidal Function (Equation (3)) to Data

Data Parameter	Depth Interval, m	Peak Date*	Probability
Flash fluorescence	0-50	Jan. 11	0.058
	100-175	June 19	0.058
Chlorophyll *a*	0-50	Dec. 12	0.058
(fluorometric)	100-175	May 14	0.058-0.078
Chlorophyll *a*	0-50	Dec. 23	0.058-0.078†
(HPLC)	100-175	May 17	0.058
Primary production	0-50	NS	0.870-0.894
	100-175	June 8	0.058
ATP	0-50	NS	0.306-0.342
	100-175	April 21	0.058
Prochlorococcus	0-50	Jan. 27	0.058
(cellular fluorescence)	100-175	NS	0.372-0.402
Synechococcus	0-50	Feb. 3	0.058
(cellular fluorescence)			
Top of the nitracline		June 1	0.058
Mixed layer depth		Dec. 9	0.058-0.078

Abbreviations are ATP, adenosine 5′-triphosphate; HPLC, high-pressure liquid chromatography; and NS, not significant.

* With phase shift that minimizes RSS.

† If data from HOT cruises 2 and 3 are included, peak is Dec. 22 and probability is 0.388-0.406

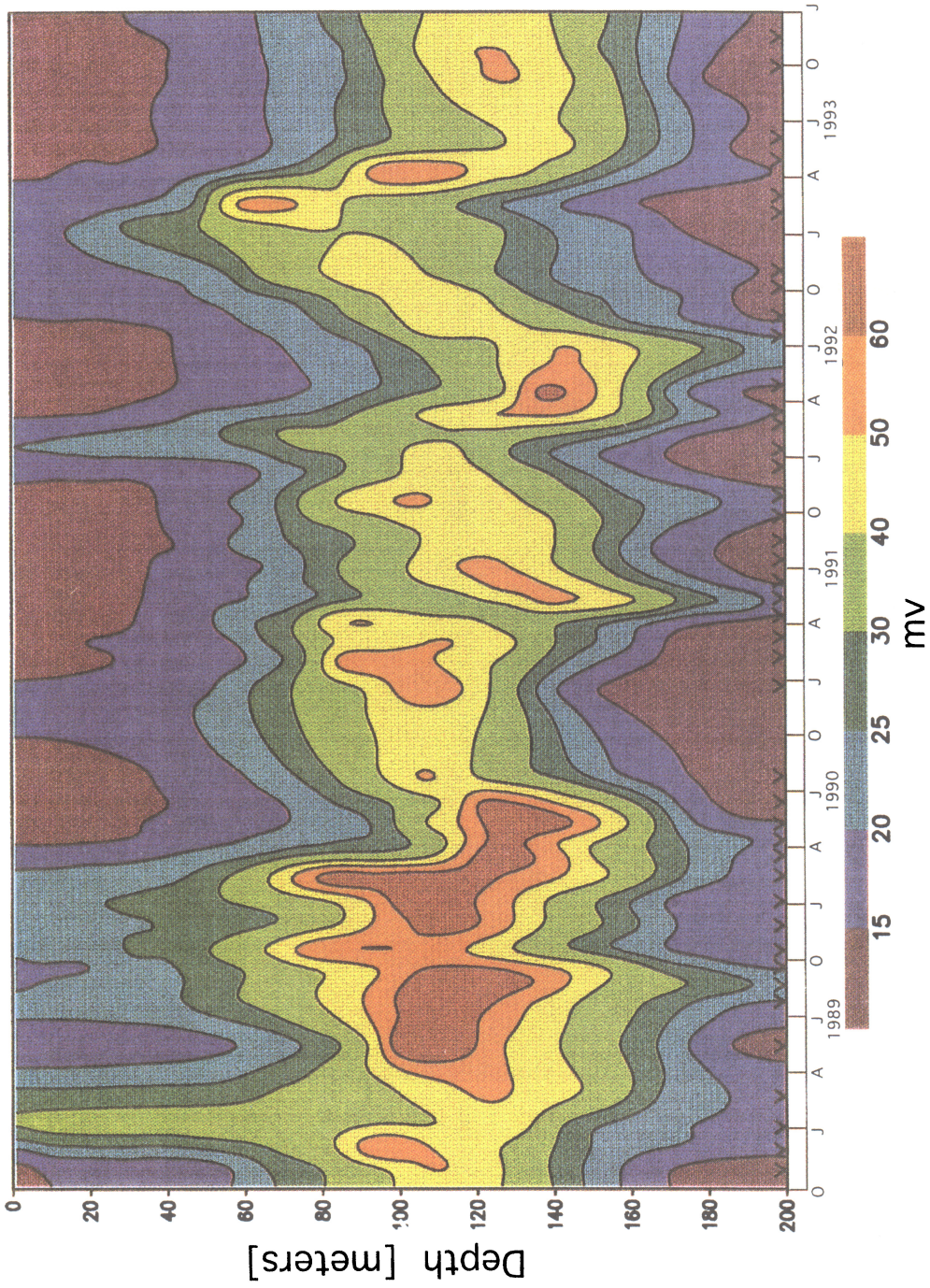


Plate 1. Contour plot of flash fluorescence in millivolts plotted against depth for the period October 1988 through October 1993. Each HOT cruise is designated by v on the horizontal axis. Contour plot was generated using a fluorescence value averaged for each 2 m.

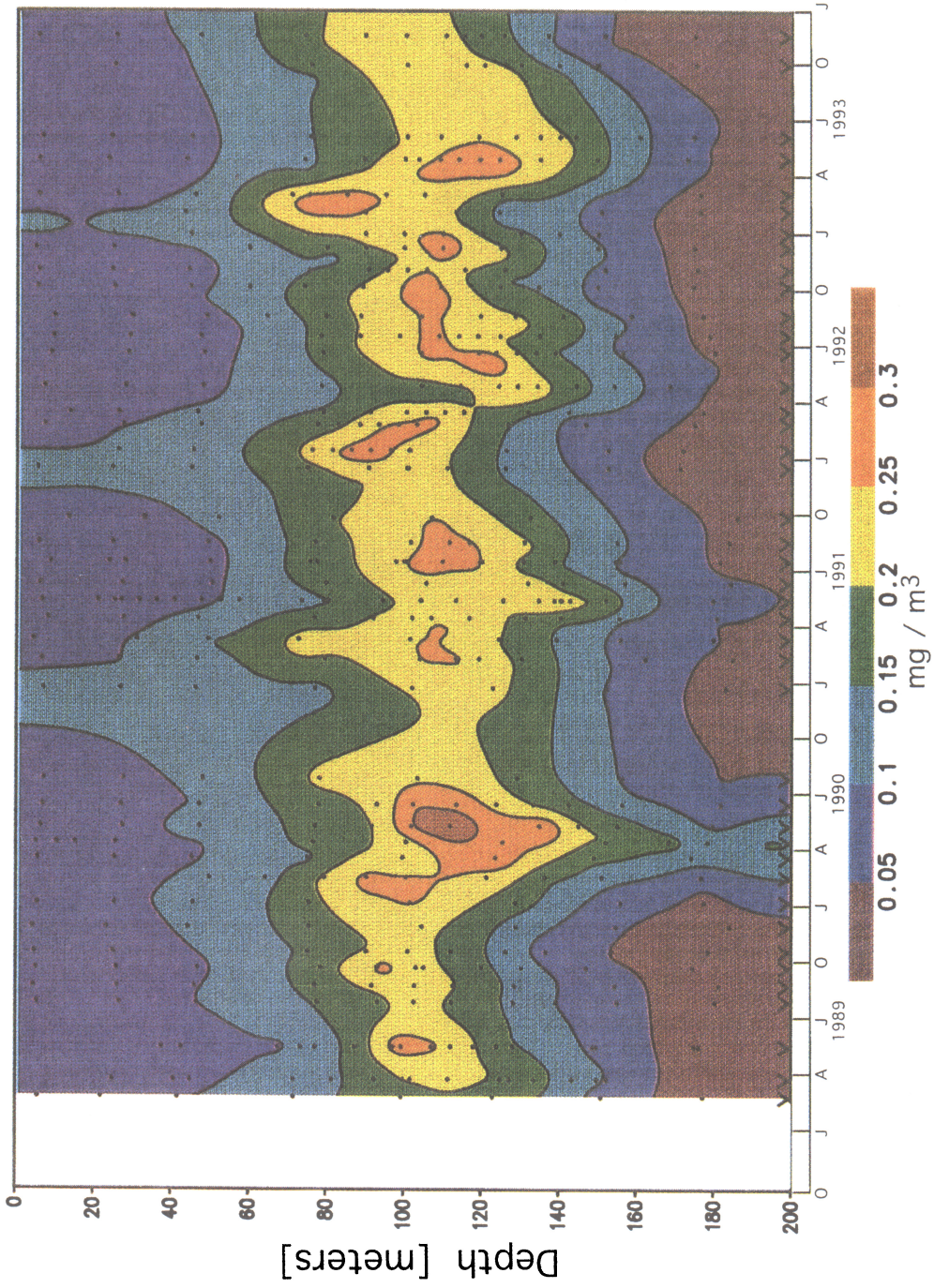


Plate 2. Contour plot of high-pressure liquid chromatography (HPLC) chlorophyll *a* in milligrams per cubic meter plotted against depth for the period October 1988 through October 1993. Each HOT cruise is designated by v on the horizontal axis. Dots represent individual samples.

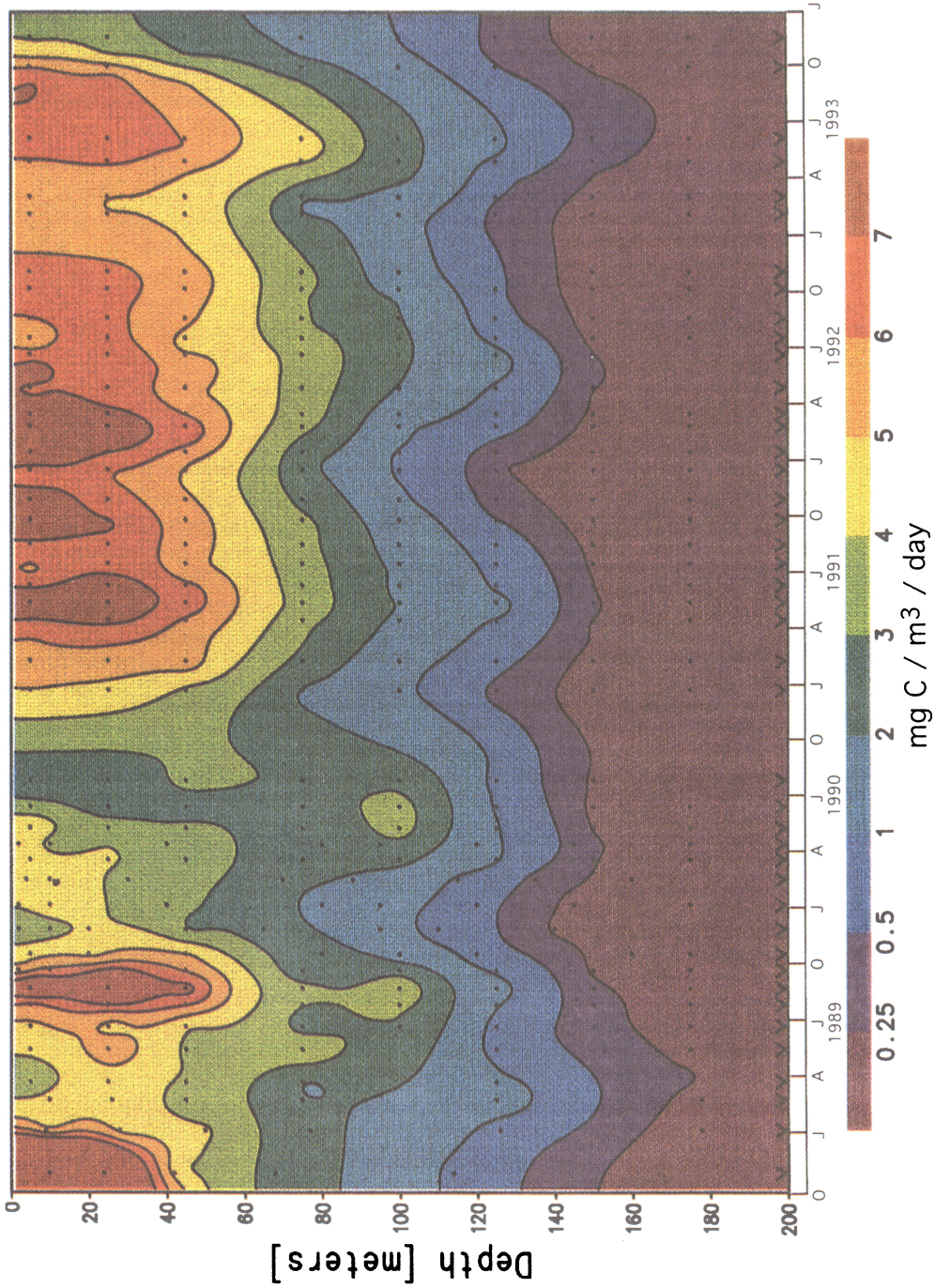


Plate 3. Contour plot of primary production in milligrams of carbon per square meter per day plotted against depth for the period October 1988 through October 1993. Each HOT cruise is designated by v on the horizontal axis. Dots represent individual samples.

with a minimum near January. A contour plot of fluorometric chl *a* (data not shown) shows similar features. The contour plot of primary production shows no evidence of a seasonal cycle in the upper 50 m of the water column, but does display a seasonal cycle between 100 and 175 m, with a maximum near July and a minimum near January (Plate 3). A contour plot of the ATP time series measurements shows a pattern of variability very similar to primary production (data not shown).

To assess the robustness of the seasonal oscillations that appear in these data, we have examined seasonal variations within two discrete layers of the euphotic zone. For the purpose of this discussion, we will call these the "upper" (0-50 m) and the "lower" (100-175 m) euphotic zones.

Temporal Variability in Flash Fluorescence Profiles

Fluorescence profiles observed at station ALOHA exhibited a distinct maximum at approximately 100 m. This maximum is associated with an increase in chl *a* per cell and does not represent increased phytoplankton biomass [*Eppley et al.*, 1973; *Beers et al.*, 1975, 1982].

A representative stack plot of fluorescence profiles collected during the burst sampling on a single HOT cruise plotted as a function of pressure shows significant variability (Figure 2a). This variability is due primarily to internal waves driven by the local semidiurnal tide. When plotted against potential density, therefore, fluorescence profiles show little temporal variability within a cruise (Figure 2b). In contrast, there is considerable variability in fluorescence profiles over an annual cycle.

A plot of the 5-year time series flash fluorescence in the upper and lower euphotic zones reveals distinct seasonal cycles, but there is also considerable interannual variability (Figure 3). In the upper euphotic zone, a flash fluorescence maximum was observed in winter, and a minimum was observed in summer (Figure 3a). A sine curve fit to these data showed a peak in January, with a probability level of 0.058 (i.e., of 10,000 random number sequences fit to the data, none gave a lower RSS than the sine curves). In the lower euphotic zone, a flash fluorescence minimum was generally observed in winter, and a maximum was observed in summer (Figure 3c). These oscillations are approximately 6 months out of phase with the upper euphotic zone. A sine curve fit to these data showed a peak in June (Table 1) and had the same level of significance. Winter and summer mean values in both the upper and lower euphotic zones were significantly different ($P<0.01$) for all three definitions of the seasons (Table 2). The magnitude of the change in fluorescence was approximately twofold for both the upper and the lower euphotic zones (Figures 3b and 3d).

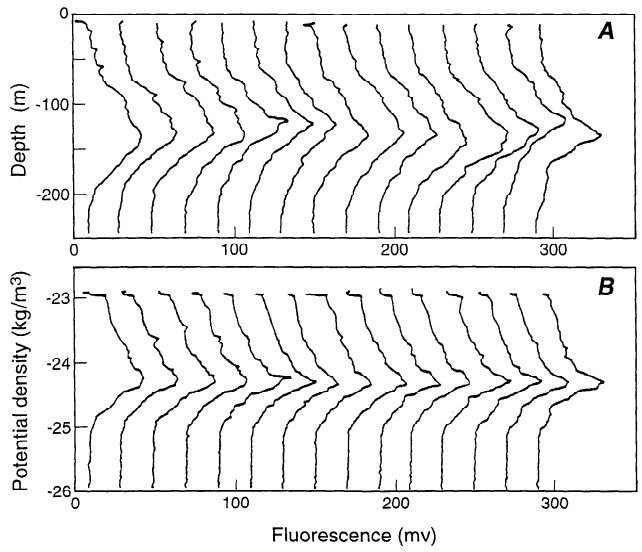


Figure 2. Flash fluorescence collected on consecutive casts on HOT cruise 30 (September 1991) showing (a) stack plot of fluorescence profiles plotted against pressureand (b) stack plot of fluorescence of profiles plotted against density. Consecutive casts are offset by 25 millivolts.

Chlorophyll *a*

Extracted chl *a* measured by fluorometry or HPLC displayed evidence of seasonal variability consistent with the flash fluorescence data (Figure 4). A sine curve fit to the data for the upper euphotic zone (Figure 4a) showed a peak in December (Table 1). A sine curve fit to the data for the lower euphotic zone (Figure 4c) showed a peak in May (Table 2), again approximately 6 months out of phase with the upper euphotic zone. Significance levels for HPLC chlorophyll in the upper euphotic zone were lower than those for fluorometric chlorophyll. This is primarily because of anomalously low values for HPLC chlorophyll on HOT cruises 2 and 3, when the HPLC technique was still under development. While we cannot say for certain that this anomaly is due to analytical error, for these two cruises the HPLC values for 0-50 m are not consistent with the fluorometric chlorophyll or flash fluorescence data, and removal of these two points from the time series greatly increases the significance of the seasonal cycle (Tables 1 and 2). For fluorometric chlorophyll in both the upper and lower euphotic zones and for HPLC chlorophyll in the lower euphotic zone, probability levels were 0.058-0.078 (Table 1).

The seasonal means for fluorometric chlorophyll in the upper euphotic zone were significantly different when winter was defined as December-March or November-February (Table 2). Seasonal means for the lower euphotic zone were significantly different for both HPLC and fluorometric chlorophyll only when the third definition (November-February) was used, but the seasonal means for HPLC were also significantly different for the second definition. The seasonal means differed by about a factor of 2 in both depth ranges (Figures 4b and 4d).

Primary Production

In the upper euphotic zone, primary production showed the least evidence of seasonality of any of the parameters examined (Figures 5a and 5b; Table 1). Seasonal means were not significantly different for any definition of the seasons (Table 2). Rates of primary production in the upper euphotic zone appear to increase significantly in 1991 and 1992 relative to the rates observed in 1989 and 1990. It has been suggested [*Karl et al.*, 1995] that this change is linked to the El Niño event of 1991-1992. In the lower euphotic zone, a relatively strong seasonal cycle was observed, with a peak in summer (Figure 5b and Table 1). Seasonal means were significantly different for two of three definitions of the seasons, overlapping those for which seasonal means of chlorophyll are also significant (Table 2). The difference between summer and winter rates of primary production in the lower euphotic zone was threefold to fourfold.

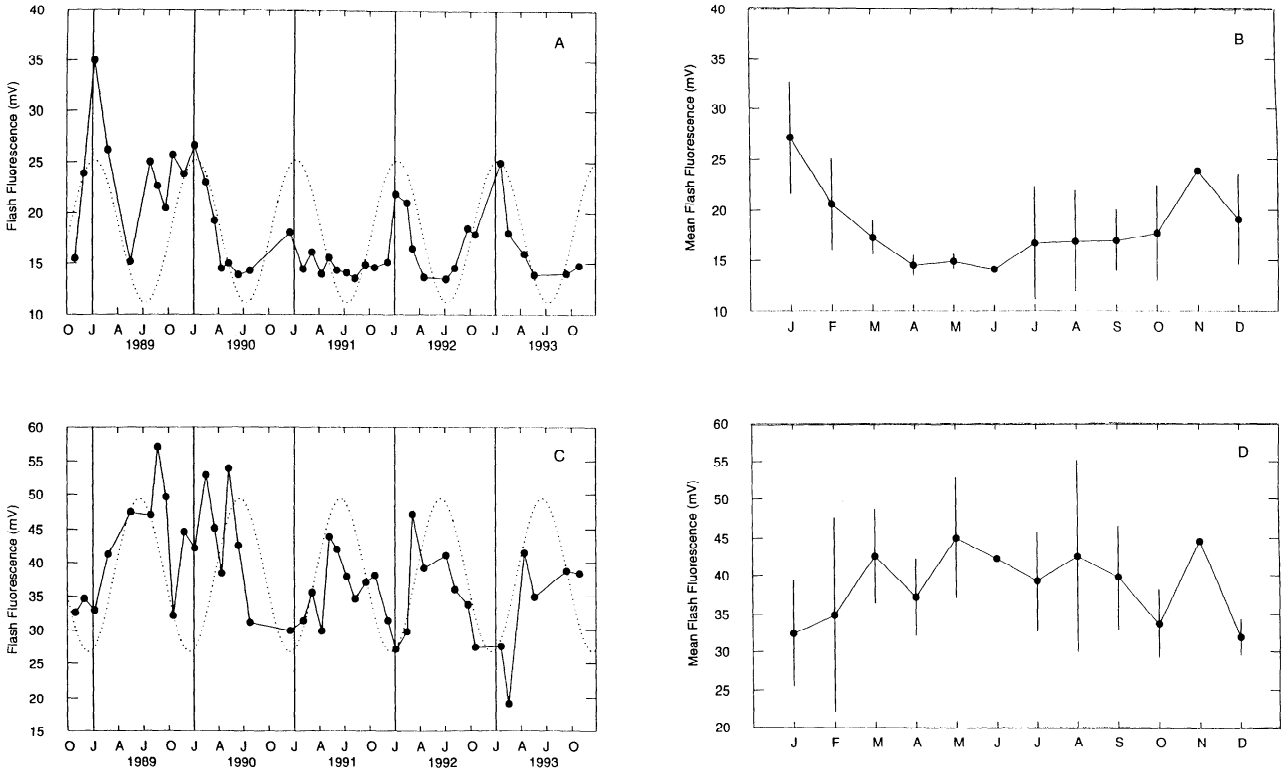


Figure 3. (a) Time-series of mean flash fluorescence in millivolts in the upper euphotic zone (0-50 m), from October 1988 through October 1993. (b) Mean values for the time series binned into monthly intervals with error bars representing the standard deviation of the binned values. (c) Time series of mean flash fluorescence in millivolts in the lower euphotic zone (100-175 m), from October 1988 through October 1993. (d) Mean values for monthly intervals, as in Figure 3b.

Adenosine 5′-Triphosphate

Adenosine 5′-triphosphate concentrations in the upper and lower euphotic zones showed patterns of variability very similar to those observed for primary production. In the upper euphotic zone, a sine curve fit to the time series data set showed weak seasonality and was out of phase with those for chlorophyll and flash fluorescence (Table 1). Seasonal means were not significantly different for any definition of the seasons (Table 2). In the lower euphotic zone, the seasonal variation was fairly strong and was to some extent in phase with those in chlorophyll, flash fluorescence, and primary production (Table 1). However, seasonal means were significantly different for only one definition of the seasons (winter being January-April), which did not overlap the seasons for which seasonal means of chlorophyll were significantly different.

Cellular Fluorescence

At station ALOHA, *Prochlorococcus* is the dominant member of the phytoplankton community [*Campbell and Vaulot,* 1993] and represents approximately 50% of the phytoplankton biomass [*Letelier et al.,* 1993; *Campbell et al.,* 1994]. *Synechococcus* is the second most abundant phytoplankton group, but the number of cells declines rapidly below 100 m [*Campbell and Vaulot,* 1993].

Flow cytometric analysis of red fluorescence per cell for samples collected from 1991 through 1993 provides an estimate of pigment per cell for these two picoplankton taxa. A representative plot for *Prochlorococcus* in the upper euphotic zone is shown in Figure 6. In the upper euphotic zone, sine curves fit to the time series of cellular fluorescence for both *Prochlorococcus* and *Synechococcus* have peaks in late January or early February (Table 1). The seasonal means were significantly different ($P<0.01$) for all definitions of the seasons (Table 2). In the lower euphotic zone, seasonality of *Prochlorococcus* cellular fluorescence was weak (Table 1). However, seasonal means were significantly different ($P<0.05$) when the third definition of the seasons (winter being November-February) was used. In the lower euphotic zone, *Synechococcus* is not present in sufficient numbers for red fluorescence per cell to be determined.

Nitracline

The depth of the top of the nitracline (defined as the depth at which $[NO_2^- + NO_3^-] > 100$ n*M*) was calculated from $[NO_2^- + NO_3^-]$ profiles, as in the work by *Dore and Karl* [1995]. In this procedure, the isopycnal occurring at a pressure halfway between the first sample at which $[NO_2^- + NO_3^-] > 100$ n*M* and the sample above it, was taken to be the density surface of the top of the nitracline. The average pressure at which this density surface was found on a particular cruise (Figures 7a and 7b) was

Table 2. Comparison of Seasonal Means by ANOVA

Parameter	Depth Interval, m	Season*	Number of Winter Observations	Number of Summer Observations	F distribution	Probability
Flash fluorescence	0-50	1	65	60	44.37	<0.01
		2	52	55	88.12	<0.01
		3	44	46	123.90	<0.01
	100-175	1	65	60	3.14	NS
		2	52	55	15.65	<0.01
		3	44	46	26.37	<0.01
Chlorophyll *a* (fluorometric)	0-50	1	16	14	0.12	NS
		2	16	14	8.39	<0.01
		3	14	13	15.05	<0.01
	100-175	1	17	14	1.99	NS
		2	17	14	0.10	NS
		3	15	13	11.89	<0.01
Chlorophyll *a* (HPLC)	0-50†	1	16	13	0.68	NS
		2	15	14	5.06	<0.05
		3	13	14	6.28	<0.25
	100-175	1	17	13	0.29	NS
		2	17	14	4.96	<0.05
		3	15	14	4.37	<0.05
Primary production	0-50	1	16	14	<0.01	NS
		2	14	15	0.17	NS
		3	12	15	0.27	NS
	100-175	1	16	14	1.18	NS
		2	14	15	12.44	<0.01
		3	12	15	22.10	<0.01
ATP	0-50	1	17	14	0.12	NS
		2	16	14	0.01	NS
		3	13	14	0.57	NS
	100-175	1	17	14	5.38	<0.05
		2	16	14	2.21	NS
		3	13	14	1.91	NS
Prochlorococcus (cellular fluorescence)	0-50	1	10	10	12.23	<0.01
		2	10	9	25.55	<0.01
		3	9	8	20.34	<0.01
	100-175	1	10	10	0.08	NS
		2	10	9	1.46	NS
		3	9	8	5.16	<0.05
Synechococcus (cellular fluorescence)	0-50	1	10	10	20.36	<0.01
		2	10	9	43.51	<0.01
		3	9	8	20.77	<0.01
Top of the nitracline		1	17	16	0.09	NS
		2	16	15	2.52	NS
		3	15	15	13.84	<0.01
Mixed layer depth		1	17	15	0.22	NS
		2	17	16	2.11	NS
		3	15	15	6.74	<0.025

* Three different definitions of seasons as described in text: 1, winter being Jan. through April; 2, winter being Dec. through March; and 3, winter being Nov. through Feb.

† If the HPLC results from HOT cruises 2 and 3 are included, there are no significant differences for any definition of season.

determined using CTD profiles collected at 3-hour intervals for at least 36 hours on each HOT cruise. This "density averaging" was done to remove the effect of internal waves, which would otherwise seriously bias our time series of nitracline depths. A strong seasonal cycle was observed with a maximum in summer (Table 1). Seasonal means are significantly different only when winter is defined as November through February (Table 2).

Mixed-Layer Depths

The depth of the mixed layer, although difficult to define precisely, is important for the interpretation of the data that we present here. Using a threshold density gradient of 5 g m^{-4} to estimate mixed-layer depth (i.e., mixed layer is defined as the shallowest depth where the density gradient equals or exceeds this value), we calculate that mixed-layer depths at station ALOHA varied between approximately 20 and 60 m (Figures 7c and 7d). The seasonal cycle in mixed-layer depth is strong and almost exactly in phase with that of mixed-layer chlorophyll (Table 1). Seasonal means are significantly different only for the third definition of the seasons (Table 2), which overlaps the seasons which give significant differences among mean chlorophyll (fluorometric) concentration in the upper euphotic zone.

Discussion

Phytoplankton ecologists frequently divide the euphotic zone in the subtropical oceans into two distinct regions. In this "two-

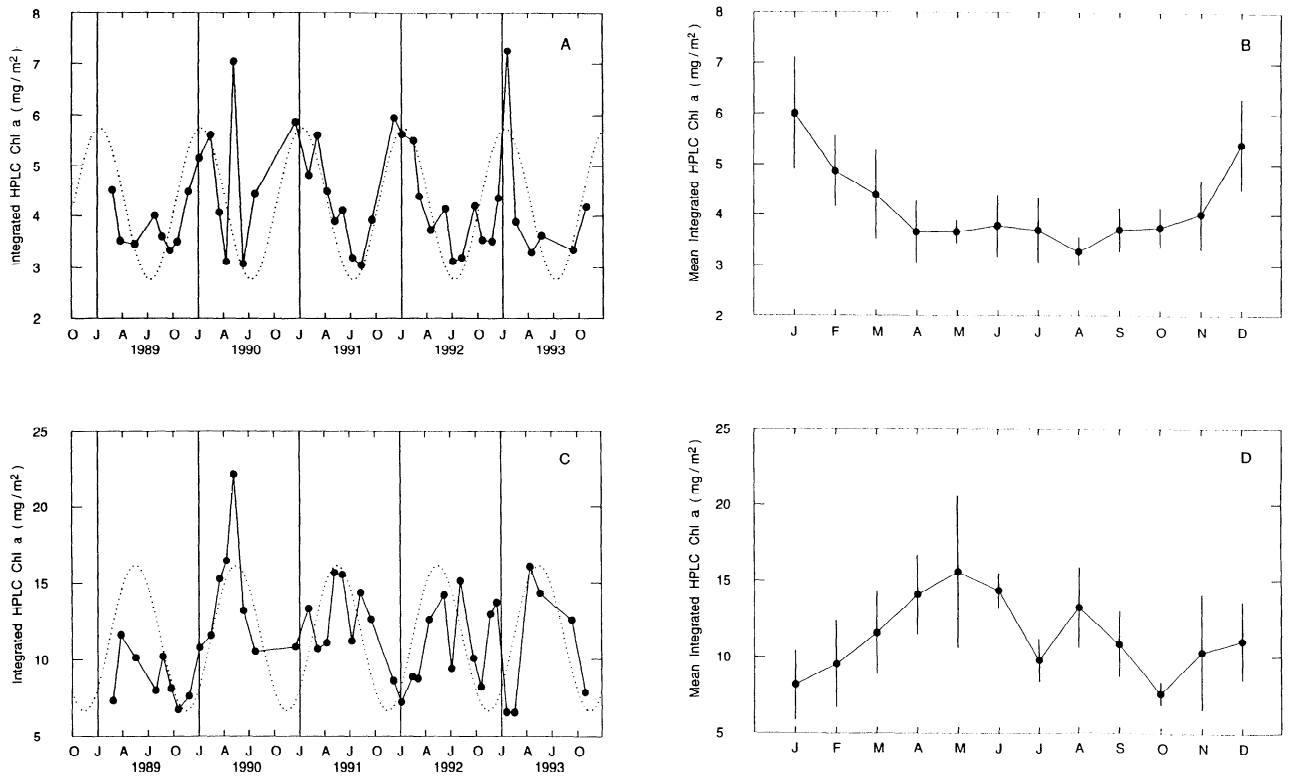


Figure 4. (a) Time series of integrated HPLC chl *a* in milligrams per square meter in the upper euphotic zone (0-50 m), from October 1988 through October 1993. (b) Mean values for the time series binned into monthly intervals with error bars representing the standard deviation of the binned values. (c) Time series of the integrated HPLC chl *a* in milligrams per square meter for the lower euphotic zone (100-175 m), from October 1988 through October 1993. (d) Mean values for monthly intervals, as in Figure 4b.

layer model," a nutrient-limited upper layer overlies a light-limited lower layer. In the NPSG, two distinct phytoplankton communities have been observed in these layers [*Venrick*, 1982]. These regions have also been described in terms of "regenerated" versus "new" production [*Dugdale and Goering*, 1967; *Eppley and Peterson*, 1979], where nutrient cycling is tightly coupled in the upper region with new production being very low (<5%), while in the deeper portion of the water column, nutrient cycling is less tightly coupled and "new" production is relatively large [*Coale and Bruland*, 1987].

Our time series observations of in situ fluorescence and chl *a* concentrations at station ALOHA show clear seasonal cycles that can be explained within the context of the two-layer model. We interpret the cycles in the upper and lower portions of the euphotic zone to be due to the responses of phytoplankton populations to seasonal cycles in insolation and upper ocean mixing.

Annual Variability in the Upper Euphotic Zone

Seasonal cycles in flash fluorescence and chl *a* concentrations are observed in the upper euphotic zone (Plates 1 and 2 and figures 3a,3b,4a and 4b). Although some interannual variability is obvious, maxima are observed in January or December for flash fluorescence and for chl *a* measured by both fluorometry and HPLC (Table 1). In contrast, there is no corresponding seasonal cycle in primary productivity or in ATP in this depth horizon. Cellular fluorescence of *Prochlorococcus* and *Synechococcus* in the upper euphotic zone also increases each winter (Table 1). However, cell abundance of *Prochlorococcus* shows no strong seasonal cycle [*Campbell and Vaulot*, 1993] and actually may decrease in winter (L. Campbell et al., unpublished data 1993). We interpret the primary production and ATP data from this study, combined with the *Prochlorococcus* results, as evidence for a lack of a seasonal cycle in phytoplankton biomass and production in the upper euphotic zone. We conclude that the seasonal changes in chlorophyll concentrations are due to a seasonal change in chlorophyll cell quota, with a maximum in the cellular chlorophyll concentrations each winter. It should be noted, however, that ATP is found in all living cells and is not exclusive to phytoplankton. In addition, although ATP is frequently used as a surrogate for biomass, ATP concentration may nevertheless vary independently of biomass, especially in phosphorus-limited systems [*Karl et al.*, 1995]. In spite of these potential complications, the data summarized here are most consistent with the hypothesis that the seasonal change in pigment concentrations in the upper euphotic zone is a consequence of seasonal changes in the quantity of chlorophyll per cell and not a consequence of a seasonal change in phytoplankton biomass.

As proposed by *Letelier et al.* [1993], we hypothesize that the seasonal change in chl *a* concentration per cell in the upper euphotic zone is due to the well-known photoadaptive response

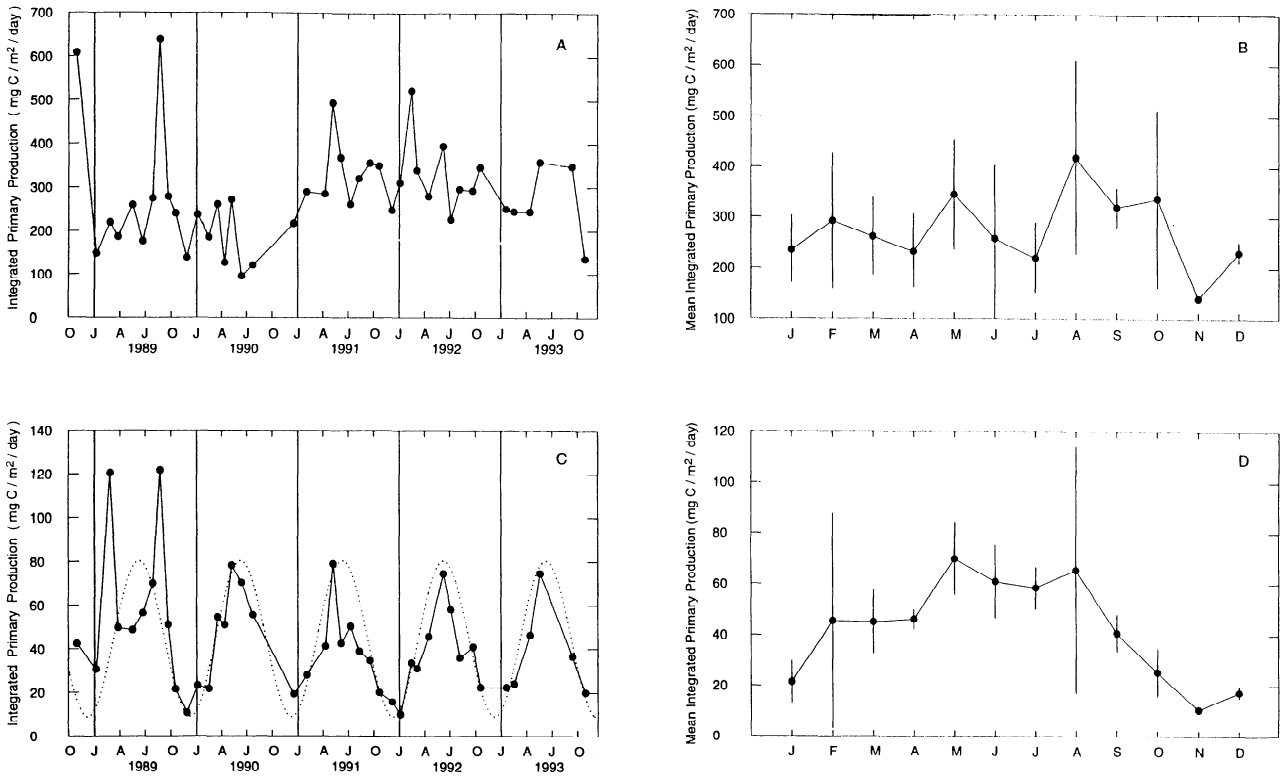


Figure 5. (a) Time series of integrated primary production in millgrams of carbon per square meter per day for the upper euphotic zone (0-50 m), from October 1988 through October 1993. (b) Mean value of integrated primary production for the time series binned into monthly intervals with error bars representing the standard deviation of the binned values. (c) Time series of mean primary production production in millgrams of carbon per square meter per day for the lower euphotic zone (100-175 m), from October 1988 through October 1993. (d) Mean values for monthly intervals, as in Figure 5b.

of phytoplankton to changes in light intensity [*Falkowski*, 1980; *Lewis et al.*, 1984ab; *Cullen and Lewis*, 1988; *Therriault et al.*, 1990]. At the latitude of station ALOHA, daily light flux at the sea surface increases from approximately 25 MJ m^{-2} d^{-1} in January to approximately 40 MJ m^{-2} d^{-1} in June [*Kirk*, 1983], approximately a 60% increase between winter and summer. In addition, a combination of an increase in wind-driven turbulence and a decrease in ocean temperature deepens the mixed layer in winter. Our observations suggest that mixed-layer depths vary between approximately 20 and 60 m between winter and summer (Figures 7c and 7d). Assuming a constant light flux at the sea surface, a deepening of the mixed layer from 20 to 60 m will produce a 40% decline in the average light intensity to which phytoplankton in the mixed layer are exposed (i.e., the average light intensity in a 60-m-deep mixed layer will be approximately 60% of the average light intensity in a 20-m-deep mixed layer). The combination of these two effects (i.e., lower light intensity in winter coupled with deeper mixed-layer depths) suggests that even at the relatively low latitude of station ALOHA, the average light intensity to which phytoplankton in the mixed layer are exposed in winter will be less than one half of that to which they are exposed in summer.

It is also possible to attribute the change in chlorophyll concentration per cell in the upper euphotic zone not to photoadaptation but to a seasonal change in the nutrient flux into the euphotic zone. It has been shown in continuous culture that chlorophyll per cell declines at low dilution rates [*Herzig and Falkowski*, 1989]. Therefore it is conceivable that chlorophyll concentrations in the upper euphotic zone decline in summer due to a decrease in the rate of nutrient input to this portion of the euphotic zone as a consequence of increased water column stability (Figures 7c and 7d). We believe photoadaptation is a more viable explanation for the observed oscillation in pigment concentration for a number of reasons. First, nutrient concentrations in the upper 50 m show no seasonal variation. This is true whether nutrients are measured with standard autoanalyzer techniques or with the more sensitive chemiluminescent method (D.M. Karl et al., Seasonal and interannual variability in primary production and particle flux at Station ALOHA, submitted to *Deep Sea Research, part II,* 1995; hereinafter referred to as Karl et al., submitted manuscript, 1995). In addition, winter mixing at the latitude of station ALOHA appears to reach the depth of the nitracline rarely, and significant nutrient injection into the upper euphotic zone as a consequence of deep mixing has been observed only infrequently in over 5 years of sampling at station ALOHA (Karl et al., submitted manuscript, 1995). Furthermore, if nutrient flux into the upper 100 m increased significantly each winter, it would be reasonable to expect that phytoplankton production would also increase in winter. Our measurements of $H^{14}CO_3^-$ uptake show no evidence of this. Given these factors, the most straightforward explanation for the observed seasonal cycle in chlorophyll concentrations is a

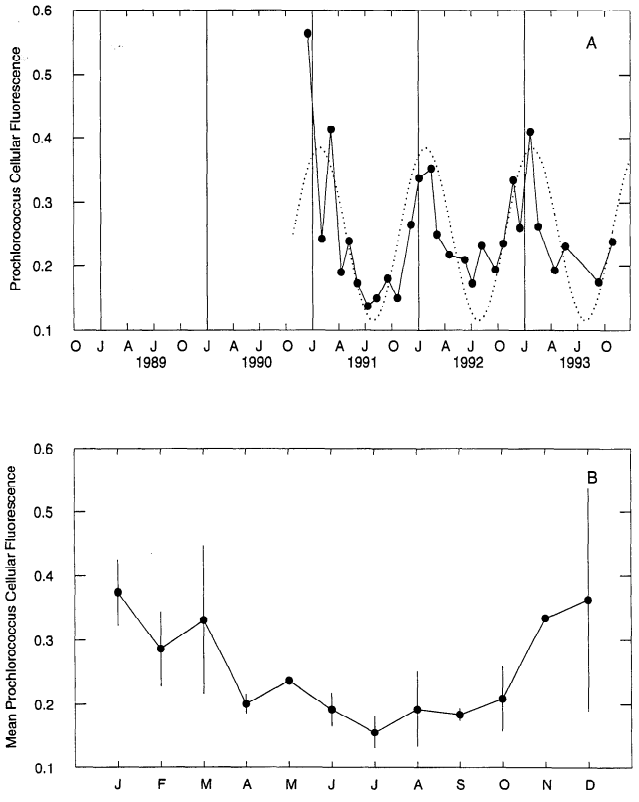


Figure 6. (a) Time series of mean *Prochlorococcus* cellular fluorescence (arbitrary units) in the upper euphotic zone (0-50 m) from December 1990 through December 1993. (b) Mean values for the time series binned into monthly intervals with error bars representing the standard deviation of the binned values.

change in the quantity of chlorophyll per cell as a consequence of photoadaptation.

Venrick [1993] has reported seasonal changes in upper ocean chl *a* concentrations, with increased concentrations in winter, as we report here. Venrick suggests that deep mixing in winter brings the larger cells found in the lower euphotic zone to the surface and that these chlorophyll-rich cells are responsible for the change in chl *a* concentrations in the surface ocean. Whereas our data do not necessarily contradict this scenario, photoadaptation within the mixed layer offers an alternative, or complementary, explanation. Our flow cytometric data, however, do provide direct evidence of photoadaptation within a specific population (Figure 6). Although it is possible that changes in community composition may contribute to the observed seasonal increases in chl *a*, *Prochlorococcus*, which contributes a large fraction of the chlorophyll in the upper euphotic zone, shows a twofold increase in chlorophyll per cell between summer and winter.

Annual Variability in the Lower Euphotic Zone

A seasonal cycle of in situ fluorescence and of chl *a* is also observed in the lower euphotic zone (Plates 1 and 2 and Figures 3c, 3d, 4c and 4d). In this depth range, all of these parameters display a maximum in May or June (Table 1). In contrast to the upper euphotic zone, seasonal trends in primary production rates and in ATP concentrations are present (Table 1). In addition, the seasonal cycles observed in primary production and in ATP are in phase with the oscillation in chlorophyll concentrations and display peaks in June and April (Table 1). We interpret this pattern of temporal variability to be due to a change in the rate of primary production and in phytoplankton biomass in the lower euphotic zone. In this depth horizon, seasonality in *Prochlorococcus* cellular fluorescence is weak or absent (Tables 1 and 2). Taken together, these observations are consistent with the hypothesis that chlorophyll concentrations track phytoplankton production and biomass reasonably well in this portion of the water column.

We hypothesize that the change in pigment concentrations and in primary production in the lower euphotic zone is a consequence of the seasonal cycle in incident solar irradiance at the sea surface. Increased solar irradiance in spring and summer moves the isolumes downward between winter and summer. Although the attenuation of this light at the base of the euphotic zone may increase as a consequence of the growth of phytoplankton, the increased daily light flux would be expected to stimulate phytoplankton growth and production in this portion of the water column where light is limiting and inorganic nutrients are available.

Seasonal Variability in the Subtropical Oceans

One of the primary objectives of the HOT program is to document and to quantify annual variability in the North Pacific Subtropical Gyre. It is useful therefore to use our 5-year time series record to estimate the range of annual variability in the parameters that we have discussed here. In the upper euphotic zone, fluorescence varies on a seasonal basis by a factor of approximately 2. This is consistent with recent satellite observations in this region of the ocean [*Yoder et al.*, 1993]. Chl *a* also varies with season by approximately this same magnitude in the upper euphotic zone. In this region, chl *a* as measured by HPLC varies between approximately 3 and 6 mg m^{-2} (Figure 4b). Expressed as an average concentration in the 0-to 50-m depth interval, this equates to a range of approximately 0.06 to 0.12 mg m^{-3}.

In the lower euphotic zone, integrated HPLC chl *a* varies from a minimum of approximately 7 mg m^{-2} to a maximum of about 15 mg m^{-2} (Figure 4d). Fluorometric chl *a* also displays regular oscillations (Table 1), although interference by other pigments diminishes the accuracy of these data, especially at the base of the euphotic zone [*Trees et al.*, 1985]. Primary production also varies seasonally by about a factor of 3 to 4 in the lower euphotic zone, with rates of primary production averaging approximately 70 mg C m^{-2} in this depth interval in summer and about 20 mg C m^{-2} in winter (Figures 5c and 5d). Integrated ATP concentrations in the lower euphotic zone also vary by about a factor of 2 between winter and summer in this depth interval.

The chl *a* concentration peaks in the upper and lower euphotic zones do not appear to be exactly 6 months out of phase. In the upper euphotic zone, a maximum in chl *a* concentrations is observed in winter (December and January), when solar irradiance is lowest. In the lower euphotic zone, however, a maximum in chl *a* concentration, ATP concentration, and primary production is observed in spring (April and May). We hypothesize that this is due to seasonal variation in the depth of the nitracline in response to the underwater irradiance field;

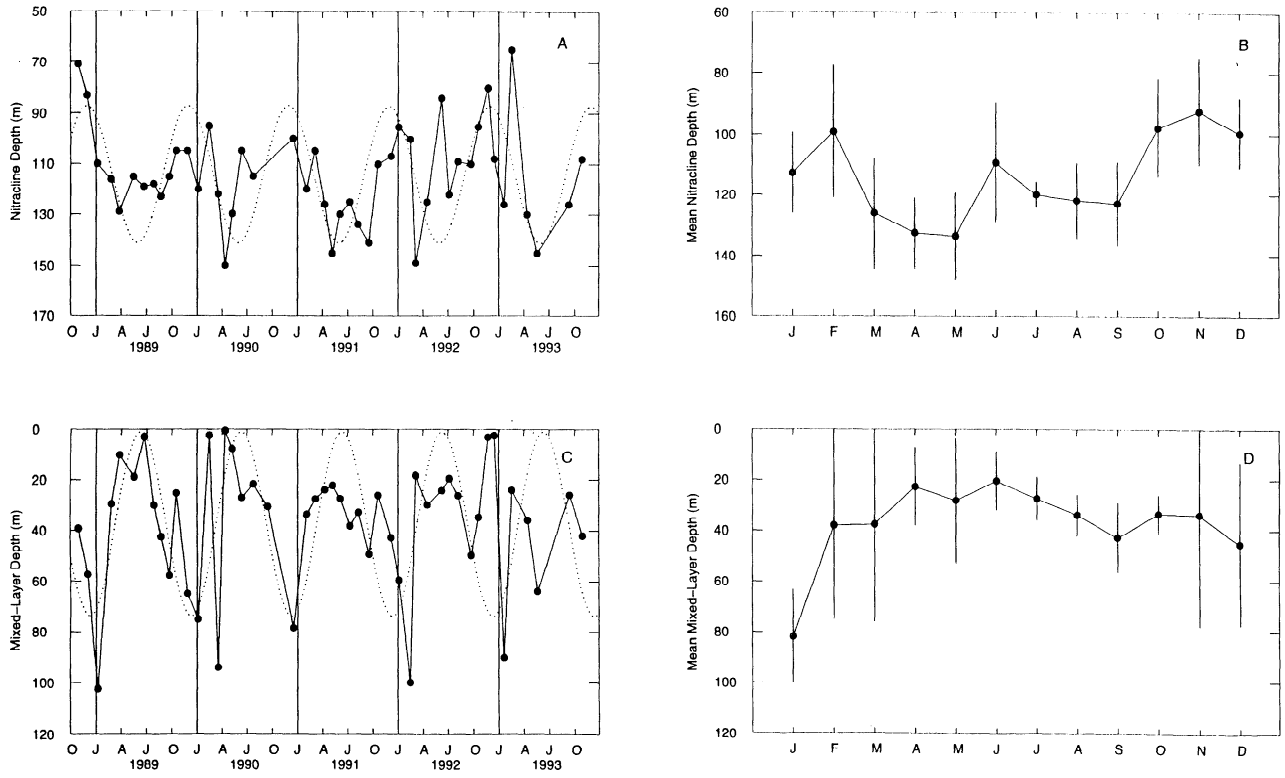


Figure 7. (a) Nitracline depths at station ALOHA from October 1988 through October 1993. (b) Mean values for the time series binned into monthly intervals with error bars representing the standard deviation of the binned values. (c) Mixed-layer depths at station ALOHA from October 1988 through 1993 with mixed-layer depths being defined on the basis of the density gradient as described in the text. (d) Mean values for monthly intervals, as in Figure 7b. Note the inverted scale on y axis in each plot.

that is, phytoplankton biomass and production are stimulated in spring, when increased irradiance increases phytoplankton production in the nitracline. The increase in phytoplankton productivity apparently produces an oscillation in the depth of the nitracline, with a maximum in May (Table 1; Figure 7a and 7b); a similar seasonal pattern in the depth of the 5- and 25-n*M* nitrite isopleths was observed by *Dore and Karl* [1995].

Middle Depths in the Euphotic Zone

Our discussion has focused on the upper and lower euphotic zones. If we define the euphotic zone as reaching a depth of 175 m (based on the depth where net photosynthesis has been shown to take place) [Letelier et al., submitted manuscript, 1995], then the discussion above has left out the region between 50 and 100 m, or almost one third of the euphotic zone. Seasonal variability in the region between 50 and 100 m is probably difficult to define, because both of the processes we have described above occur there. It is reasonable to assume that a gradual transition occurs in this region from approximately 50 m, where seasonal variability characteristic of the upper euphotic zone predominates, to below 100 m, where the characteristics more closely associated with the lower euphotic zone predominate. However, the gradual transition between the upper and the lower euphotic zones is probably significantly affected by variability in mixed layer depths.

Conclusions

The data that we present here have important implications for the study of the subtropical oceans. We have shown that seasonal variability occurs with distinct patterns in the upper and the lower euphotic zones at the latitude of station ALOHA. These seasonal cycles can be easily explained as a consequence of the annual oscillation in solar declination and related seasonal changes in upper ocean mixing. These variations are most easily understood within the context of the two-layer model of the subtropical euphotic zone [*Dugdale*, 1967; *Eppley et al.*, 1973]. In many circumstances, it may be inappropriate to study temporal variability in phytoplankton processes (e.g., pigment concentrations and primary production) using measurements integrated over the entire euphotic zone.

We suspect that the pattern of seasonal variability that we describe here may be common to large areas of the subtropical oceans. We note that the pattern of variability in upper ocean chl *a* concentrations that we have observed at station ALOHA has also been observed at the Climax site [*Venrick*, 1993] and over vast regions in the subtropical gyres via satellite [*Yoder et al.*, 1993]. Assuming that seasonal variability of this kind is common to the subtropical gyres between approximately 10° and 30°, this pattern of variability may exist over approximately 7 x 10^7 km^2, or roughly 15% of the Earth's surface.

The data that we summarize here also have implications for the use of chlorophyll fluorescence for the study of temporal variability, at least in the subtropical oceans. Although fluorescence is not a direct measure of chl *a* concentration, our data indicate that the use of night time fluorescence profiles is a valid approach to studying temporal variability in chl *a* concentrations in the subtropical oceans. This approach has the important advantage of providing for frequent measurements in both space and time.

Our observations are also relevant for remote sensing of phytoplankton processes in the subtropical oceans. Our results suggest that seasonal changes in chl *a* concentrations in the surface mixed layer at station ALOHA are largely a consequence of photoadaptation and are not related to changes in phytoplankton biomass [*Yoder et al.*, 1993]. It is reasonable to suggest that this might also be the case for the vast expanse of the subtropical seas. As a consequence, we suggest that satellite-based estimates of chlorophyll concentrations in these regions [*Yoder et al.*, 1993] cannot be used to describe temporal variability in biomass without correcting for changes in the quantity of chlorophyll per cell. In addition, our data show that significant seasonal variability in chlorophyll concentrations, primary production, and phytoplankton biomass occurs near the base of the euphotic zone. This region of the water column cannot be monitored from space. In contrast to the surface water, in this deeper portion of the subtropical euphotic zone, chlorophyll concentrations appear to be a reasonable surrogate for phytoplankton biomass.

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References

Beers, J.R., F.M.H. Reid, and G.L. Stewart, Microplankton of the North Pacific Central Gyre: Population structure and abundance, June 1973, *Int. Rev. Gesamten. Hydrobiol.*, *60*, 607-638, 1975.

Beers, J.R., F.M.H. Reid, and G.L. Stewart, Seasonal abundance of the microplankton population in the North Pacific Central Gyre, *Deep Sea Res.*, *28*, 981-1000, 1982.

Bidigare R. R., O. Schofield, and B. B. Prézelin, Influence of zeaxanthin on quantum yield of photosynthesis of *Synechococcus* clone WH7803 (DC2), *Mar. Ecol. Prog. Ser.*, *56*, 177-188. 1989.

Bienfang, P.K., and J.P. Szyper, Phytoplankton dynamics in the subtropical Pacific Ocean off Hawaii, *Deep Sea Res.*, *28a*, 981-1000, 1981.

Bienfang, P.K., J.P. Szyper, M.Y. Okamoto, and E.K. Noda, Temporal and spatial variability of phytoplankton in a subtropical ecosystem, *Limnol. Oceanogr.*, *29*, 527-539, 1984.

Blackburn, M., Low latitude gyral regions, in *Analysis of Marine Ecosystems*, edited by A.R. Longhurst, pp. 3-29, Academic, San Diego, Calif., 1981.

Campbell, L. and D. Vaulot, Photosynthetic picoplankton community structure in the subtropical North Pacific Ocean near Hawaii (station ALOHA), *Deep Sea Res., Part I*, *40*, 2043-2060, 1993.

Campbell, L., H.A. Nolla, and D. Vaulot, The importance of *Prochlorococcus* to community structure in the central North Pacific Ocean, *Limnol. Oceanogr.*, *39*, 954-961, 1994.

Chavez, F.P., K.R. Buck, R.R. Bidigare, D.M. Karl, D. Hebel, M. Latasa, L. Campbell, and J. Newton, On chlorophyll *a* retention properties of glass-fiber GF/F filters, *Limnol. Oceanogr.*, *40*, 428-433, 1995.

Coale, K.H., and K.W. Bruland, Oceanic stratified euphotic zone as elucidated by ^{234}Th:^{238}U disequilibria, *Limnol. Oceanogr.*, *32*, 189-200, 1987.

Cox, R.D., Determination of nitrate at the parts per billion level by chemiluminescence, *Anal. Chem.*, *52*, 332-335, 1980.

Cullen, J.J. and M.R. Lewis, The kinetics of algal photoadaptation in the context of vertical mixing, *J. Plankton Res.*, *10*, 1039-1063, 1988.

Cushing, J.D., The seasonal variation in oceanic production as a problem in population dynamics, *J. Cons. Cons. Int. Explor. Mer.*, *24*, 455-464, 1959.

DiTullio, G.T., and E.A. Laws, Impact of atmospheric-oceanic disturbances on phytoplankton community dynamics in the North Pacific Central Gyre, *Deep Sea Res.*, *38*, 1305-1329, 1991.

Dore, J.E., and D.M. Karl, Nitrite distributions and dynamics at Station ALOHA. *Deep Sea Res.*, part II, in press. 1995.

Ducklow, H.W., and R.P. Harris, Introduction to the JGOFS North Atlantic bloom experiment, *Deep Sea Res., Part II*, *40*, 1-8, 1993.

Dugdale, R.C., Nutrient limitation in the sea: Dynamics, identification and significance, *Limnol. Oceanogr.*, *12*, 685-695, 1967.

Dugdale, R.C., and J.J. Goering, Uptake of new and regenerated forms of nitrogen in primary productivity, *Limnol. Oceanogr.*, *12*, 196-206, 1967.

Eppley, R.W., and B.J. Peterson, Particulate organic matter flux and planktonic new production in the deep ocean, *Nature*, *282*, 677-680, 1979.

Eppley, R.W., E.H. Renger, E.L. Venrick, and M.M. Mullin, A study of phytoplankton dynamics and nutrient cycling in the central gyre of the North Pacific Ocean, *Limnol. Oceanogr.*, *18*, 534-551, 1973.

Falkowski, P.G., Light-shade adaptation in marine phytoplankton, in *Primary Productivity in the Sea*, edited by P.G. Falkowski, pp. 99-120, Plenum, New York, 1980.

Falkowski, P., and D.A. Kiefer, Chlorophyll *a* fluorescence in phytoplankton: Relationship to photosynthesis and biomass, *J. Plankton Res.*, *7*, 715-731, 1985.

Fitzwater, S.E., G.A. Knauer, and J.H. Martin, Metal contamination and its effect on primary productivity measurements, *Limnol. Oceanogr.*, *27*, 544-551, 1982.

Garside, C., A chemiluminescent technique for the determination of nanomolar concentrations of nitrate and nitrite in seawater, *Mar. Chem.*, *11*, 185-197, 1982.

Hayward, T.L., The nutrient distribution and primary production of the central North Pacific, *Deep Sea Res.*, *34*, 1593-1627, 1987.

Hayward, T.L., E.L. Venrick, and J.A. McGowan, Environmental heterogeneity and plankton community structure in the central North Pacific, *J. Mar. Res.*, *41*, 711-729, 1983.

Herzig, R., and P.G. Falkowski, Nitrogen limitation of *Isocrysis galbana*. I. Photosynthetic energy conversion and growth efficiencies, *J. Phycol.*, *25*, 462-471, 1989.

Holm-Hansen, O., Determination of total microbial biomass by measurement of adenosine triphosphate, in *Estuarine Microbial Ecology*, edited by L.H. Stevenson and R.R. Caldwell, pp. 73-89, Univ. S. C. Press, Columbia, 1973.

Jitts, J.R., Seasonal variations in the Indian Ocean along 110° E, IV, primary production, *Australian J. Mar. Freshwater Res.*, *20*, 65-75, 1969.

Karl, D.M., and O. Holm-Hansen, Methodology and measurement of adenylate energy charge ratio in environmental samples, *Mar. Biol. Berlin*, *48*, 185-197, 1978.

Karl, D.M., and C.D. Winn, A sea of change: Monitoring the oceans' carbon cycle, *Environ. Sci. Technol.*, *25*, 1977-1980, 1991.

Karl, D.M., R.M. Letelier, D.V. Hebel, L.M. Tupas, J.E. Dore, J.R.Christian, and C.D. Winn, Ecosystem changes in the North Pacific Subtropical Gyre attributed to the 1991-1992 El Niño, *Nature*, *373*, 230-234, 1995.

Kirk, J.T.O., *Light and Photosynthesis in Aquatic Ecosystems*. 401 pp., Cambridge Univ. Press, New York, 1983.

Letelier, R.M., R.R. Bidigare, D.V. Hebel, M. Ordrusek, C.D. Winn, and D.M. Karl, Temporal variability of phytoplankton community structure at the U.S.-JOGFS Station ALOHA (22° 45' N, 158° W) based on HPLC pigment analysis, *Limnol. Oceanogr.*, *38*, 1420-1437, 1993.

Lewis, M.R., J.J. Cullen, and T. Platt, Relationships between vertical mixing and photoadaptation of phytoplankton: Similarity criteria, *Mar. Ecol. Prog. Ser., 15*, 141-149, 1984a.

Lewis, M.R., E.P.W. Horne, J.J. Cullen, N.S. Oakey, and T. Platt, Turbulent motions may control phytoplankton photosynthesis in the upper ocean, *Nature, 311*, 49-50, 1984b.

Lohrenz, S.E., G.A. Knauer, V.L. Asper, M. Tuel, A.H. Knap, and A.F. Michaels, Seasonal variability in primary production and particle flux in the northwestern Sargasso Sea: U.S.-JGOFS Bermuda Atlantic Time Series, *Deep Sea Res., 39*, 1373-1391, 1992a.

Lohrenz, S.E., D.A. Weisenberg, and C.R. Rein, A comparison of in situ and simulated in situ methods for estimating oceanic primary productivity, *J. Plankton Res., 14*, 201-215, 1992b.

McGowan, J.A., and T.L. Hayward, Mixing and oceanic productivity, *Deep Sea Res., 25*, 771-793, 1978.

Menzel D.W., and J.H. Ryther, The annual cycle of primary production in the Sargasso Sea off Bermuda, *Deep Sea Res., 6*, 351-367, 1960.

Owen, R.W., and B. Zeitzschel, Phytoplankton production: Seasonal change in the oceanic eastern tropical Pacific, *Mar. Biol. Berlin, 7*, 32-36, 1970.

Strickland, J.D.H., and T.R. Parsons, *A Practical Handbook of Seawater Analysis*. Fish. Res. Board of Canada, 167 pp. Ottawa, Canada, 1972.

Sverdrup, H.U., On conditions for the vernal blooming of phytoplankton, *J. Cons. Cons. Perm. Int. Explor. Mer., 18*, 287-295, 1953.

Therriault, J.C., D. Booth, L. Legendre, and S. Demers, Phytoplankton photoadaptation to vertical excursion as estimated by an in vivo fluorescence ratio, *Mar. Ecol. Prog. Ser., 60*, 97-111, 1990.

Trees, C.C., M.C. Kennicutt, and J.M. Brooks, Errors associated with the standard fluorometric determination of chlorophylls and phaeopigments, *Mar. Chem., 17*, 1-12, 1985.

Triola, M. F., *Elementary Statistics*, 4th ed., 784 pp. Benjamin Cummings, Redwood City, California, 1980.

Vaulot, D. , CYTOPC: Processing software for flow cytometric data, *Signal Noise, 2*, 8, 1989.

Venrick, E.L., Phytoplankton in an oligotrophic ocean: Observations and questions. *Ecol. Monogr., 52*, 129-154, 1982.

Venrick, E.L., Phytoplankton seasonality in the central North Pacific: The endless summer reconsidered, *Limnol. Oceanogr., 38*, 1135-1149, 1993.

Winn, C.D., F.T. Mackenzie, C.J. Carrillo, C.L. Sabine, and D.M. Karl, Air-sea carbon dioxide flux in the North Pacific Subtropical Gyre: Implications for the global carbon budget, *Global Biogeochem. Cycles, 8*, 157-163, 1994.

Yoder, J.A., C.R. McClain, G.C. Feldman, and W.E. Esaias, Annual cycles of phytoplankton chlorophyll concentrations in the global ocean: A satellite view, *Global Biogeochem. Cycles, 7*, 181-194, 1993.

Young, R.W., K.L. Carder, P.R. Betzer, D.K. Costello, G.R. DiTullio, N.W. Tindale, E.A. Laws, M. Uematsu, J.T. Merril and R.A. Feely, Atmospheric iron inputs and primary productivity: Phytoplankton responses in the North Pacific, *Global Biogeochem. Cycles, 5*, 119-134, 1991.

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