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Diel cycles in surface waters of the equatorial Pacific

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Abstract

Diel variations in the abundance, cellular light scattering, and fluorescence of picophytoplankton (*Prochlorococcus* and *Synechococcus*) and ultraphytoplankton in the equatorial Pacific are well-documented. Cycles of abundance are driven primarily by cell division, which in these groups generally occurs in the late afternoon or at night. Cell division is well-phased within each group, but its timing varies to some extent among groups and between different depths and locations. Grazing mortality acts to reduce abundance over the course of the day and can itself vary over the diel cycle, although a consistent picture of this variation has not yet emerged. Diel cycles in cellular light scattering appear to be largely driven by cell size changes, which reflect the balance between cell growth and division. Decreases in light scattering generally coincide with increases in cell number, and (in the case of *Prochlorococcus*) with periods of active cell division as determined by cell cycle analysis. Diel variability in cellular fluorescence is most apparent in *Prochlorococcus* and *Synechococcus*, and appears to be driven by changes in both pigment content (mediated by pigment synthesis and cell division) and light-dependent non-photochemical fluorescence quenching.

Diel variations in bulk fluorescence and beam attenuation are consistent (at least qualitatively) with the observed cycles of cellular abundance, light scattering, and fluorescence. Direct and quantitative assessments of the relative contributions of variations in cell abundance, pigment quota, and fluorescence yield among phytoplankton groups to diel cycles in bulk fluorescence are not yet available. In the case of beam attenuation a few such quantitative assessments have been made, and although the results are not all in agreement, diel changes in the size and abundance of *Prochlorococcus* and ultraphytoplankton appear to explain the observed diel variation in beam attenuation to a large degree. Estimates of primary production and phytoplankton growth rate can be derived from this diel variation, but owing to the assumptions they incorporate, these estimates should be interpreted with caution. © 2002 Elsevier Science Ltd. All rights reserved.

1. Introduction

The daily light/dark cycle is pervasive in all temperate and tropical near-surface marine ecosystems. It influences these ecosystems in a great

variety of ways, ranging from direct control of primary production to indirect effects on water-column mixing to subtle influences on the behavior of organisms. In the equatorial Pacific and other tropical areas, where seasonal variations in biological and physical characteristics are relatively small, diel cycles may represent a particularly important source of variability in many ecosystem properties (e.g., Chung et al., 1996). This fact has

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practical implications (e.g., for designing sampling strategies), but more importantly it has fundamental implications for our understanding of ecosystem processes in the equatorial Pacific and elsewhere (Vaulot and Marie, 1999). We may be interested, for example, in the dynamics of carbon export over the course of an ENSO cycle, but these dynamics will be controlled ultimately by interactions among biological and physical processes that have strong diel components. A full understanding of the factors regulating larger scale variability in the equatorial Pacific therefore will require a meaningful understanding of diel variability in this environment. At the same time, observations of such diel variability can be used to develop a more thorough understanding of many of the processes that contribute to the larger scale patterns.

The intent of this paper is to examine the nature and extent of diel cycles in the biological characteristics of the surface waters of the equatorial Pacific, particularly with respect to the influence of such characteristics on “bulk” water properties. As will be discussed herein, the smallest biological components (picophytoplankton, ultraphytoplankton, and heterotrophic bacteria) are generally assumed to contribute most to these bulk properties. Therefore, in this review we concentrate on these smaller components. Diel cycles are no less important among larger organisms in the equatorial Pacific (e.g., Zhang and Dam, 1997), but these organisms are not believed to influence directly bulk water properties to the same extent as microbial populations. For the purposes of the discussion that follows, we will use the terms picophytoplankton and ultraphytoplankton to refer to photosynthetic prokaryotes (*Synechococcus* and *Prochlorococcus*) with cell diameters $< \sim 2 \mu\text{m}$, and photosynthetic eukaryotes with cell diameters on the order of $3 \mu\text{m}$ or less, respectively (Binder et al., 1996). In those cases where authors have used different terminology (particularly with respect to the small photosynthetic eukaryotes), we use the authors’ terminology when referring specifically to their study.

It is worth noting that most of the published studies relevant to this review concern upwelling “HNLC” waters of the eastern equatorial Pacific.

The discussion and conclusions that follow should therefore be considered to apply most directly to this area. Data from the more oligotrophic “warm pool” waters of the western side of the Pacific basin are included when available, and are noted as such.

The forces underlying diel cycles in biological properties can be numerous and closely intertwined. Diel cycles in the activities or characteristics of organisms can be driven by extrinsic periodicity in the physical environment (e.g., light, temperature) or the biological environment (e.g., grazing pressure, prey abundance), and by intrinsic biological cycles (e.g., circadian cycles, cell division cycles), which can themselves be entrained by environmental cues (Prézelin, 1992). These diel cycles in biological activity in turn can influence the physical and biological environment. Teasing apart the specific forcing functions that act to shape observable diel behavior is beyond the scope of this review. We will instead concentrate on characterizing such behavior in pico- and ultraphytoplankton, and on examining the extent to which this behavior can explain diel variability in bulk water properties.

2. Biological characteristics

2.1. Pico- and ultraphytoplankton abundance

In the absence of significant import or export of cells, diel variations in the abundance of a phytoplankton population are driven primarily by two processes: cell division, which increases cell number, and cell mortality (e.g., grazing, viral lysis), which decreases it. Because these two processes affect abundance in opposite directions, changes in cell number often can be used to establish the timing of cell division and/or grazing, in at least a qualitative way. However, because both of these processes can occur simultaneously, cell abundance data alone can never unequivocally establish the *absence* of either at a given time. Thus, while increases in cell number can only result from cell division, grazing mortality may be occurring at the same time; likewise, while decreases can only result from cell mortality, this

does not exclude the possibility that cell division is occurring as well.

It should be noted that diel changes in mixed-layer depth (see discussion below) may also influence diel patterns in cell abundance (as well as in mean cellular properties) at a given depth, particularly if there exists a strong vertical gradient in abundance (or in the properties of interest). Although such gradients do exist in at least some of the relevant studies (e.g., Vaultot and Marie, 1999), the consistency of the observed diel patterns, and their occurrence in the absence of vertical gradients in at least some cases (e.g., Blanchot et al., 1997) argues against diel mixing as a major forcing function of these patterns. Nevertheless, it is possible that subtle differences in diel patterns of cellular abundance or properties could result from this sort of forcing (André et al., 1999).

Diel variations in the abundance of *Prochlorococcus*, *Synechococcus*, and ultraphytoplankton are ubiquitous and obvious in the equatorial Pacific (Binder et al., 1996; DuRand and Olson, 1996; Landry et al., 1996; Blanchot et al., 1997; Liu et al., 1997; André et al., 1999; Vaultot and Marie, 1999; Mann and Chisholm, 2000). In general, these groups increase in abundance during the latter half of the day and/or during the night and decrease in abundance at other times (Fig. 1, Table 1). The precise timing of daily cell number increases and decreases is not identical among these three groups, nor among populations at different times and in different places, however. Generally speaking, the daily increase in surface *Synechococcus* cell number occurs in the afternoon or early evening, and precedes that in *Prochlorococcus* (Fig. 1); in the single case where *Synechococcus* and *Prochlorococcus* increases began at the same time, the perceived timing of the *Prochlorococcus* increase may have been affected by undercounts of this group in the afternoon (owing to very low midday fluorescence) (Vaultot and Marie, 1999). This is supported by the cellular light scattering data from this same study, which shows that the decrease in *Prochlorococcus* cell size lags that in *Synechococcus* (see Section 2.2, below). Except as discussed above, the daily increase in surface *Prochlorococcus* numbers is generally observed to occur in the early evening (sunset–

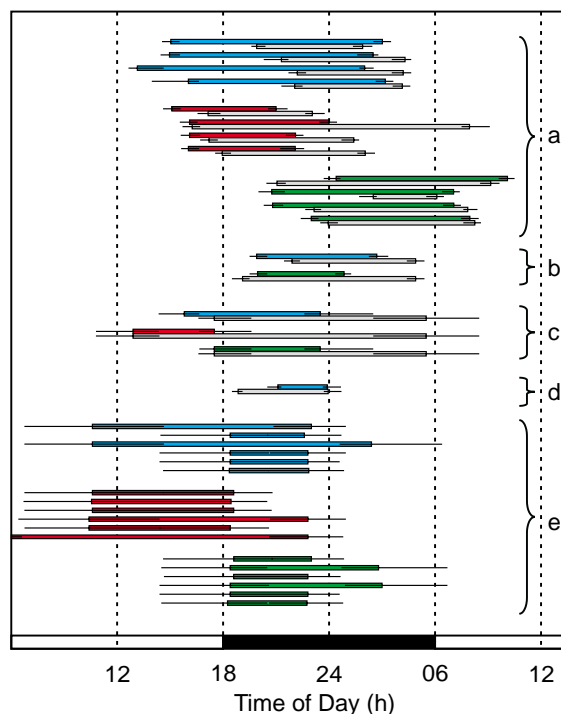


Fig. 1. The apparent diel timing of cell division, as reflected in increasing cell abundance (colored bars) or decreasing cellular light scattering (shaded bars) in surface populations (depth $\leq \sim 5$ m) of *Prochlorococcus* (blue bars), *Synechococcus* (red bars), and ultraphytoplankton (green bars) in the equatorial Pacific. Bars represent the period of time over which abundance is increasing or light scattering is decreasing on a given day, as picked by eye from references (indicated by letters) listed in Table 1. Error lines are intended to indicate the time resolution of each data set, and for each starting or ending point show half the distance between that point and adjacent time points. This error may be less than the actual error associated with determining the starting and ending points (owing to random variability in the data). Data are normalized to sunset at 18:00. In the case of (c), the end of the light scatter decrease is taken as the last time point in the study, and the corresponding time resolution is approximated by the preceding sampling interval.

midnight) (Fig. 1). Picoeukaryotes increase over about the same time period, or slightly later than *Prochlorococcus* (Fig. 1). Given the large range in time and space that these observations subtend (Table 1), the eulerian nature of the sampling in most cases, and the sensitivity of cell cycle timing to environmental conditions (Mann and Chisholm, 2000), the consistency of these diel patterns is notable.

Table 1

Studies of diel variation in surface pico- and ultraphytoplankton abundance and/or cellular characteristics in the equatorial Pacific, used for Fig. 1

Reference ^a	Location (cruise name)	Dates	Sampling interval (h) ^b
(a) Vaultot and Marie (1999)	5°S, 150°W (OLIPAC)	Nov. 18–22, 1994	1.0
(b) Blanchot et al. (1997) [Fig. 3]	0°, 150°W (FLUPAC)	Oct. 23, 1994	1.0
(c) DuRand and Olson (1996)	0°, 140°W (EQPAC-TT12)	Oct. 11, 1992	3.0
(d) Mann and Chisholm (2000) [Fig. 1]	4°S, 105°W (IronEx II)	May 1995	1.2 (0.7)
(e) André et al. (1999)	0°, 150°W (FLUPAC)	Oct. 19–25, 1994	6.0 (4.1)

^a Letter in parentheses refers to designation in Fig. 1. Source of data is indicated in brackets when necessary to avoid ambiguity.

^b Average sampling interval over the entire day; values in parentheses are intervals during the first half of the dark period, when this is different than the 24 h average.

Similar diel abundance patterns are observed in sub-surface (> ~5 m depth) populations (André et al., 1999; Vaultot and Marie, 1999), although the data for these populations are necessarily less comprehensive than for the more easily accessible surface populations (which can be sampled by pump at high frequency under all conditions). Vaultot and Marie (1999) observed that the daily increase in *Synechococcus* abundance at 25 and 50 m occurs 2 h earlier than at the surface. The data of André et al. (1999), though of lower frequency, suggest that a similar advance in timing at depth occurs among *Prochlorococcus* and picoeukaryotes. The sampling regime in this case is such that although no advance in *Synechococcus* timing was evident, the data are not inconsistent with the advance in *Synechococcus* observed by Vaultot and Marie (1999). These depth-dependent differences in the timing of cell abundance increases are paralleled by differences in the timing of light scattering decreases and cell cycle progression (in the case of *Prochlorococcus*), and have been suggested to be a consequence of UV exposure of surface populations (see discussion below) (Vaultot et al., 1995; Vaultot and Marie, 1999).

2.2. Cell size

Diel changes in cell size or carbon content result primarily from two opposing processes: cell growth via carbon fixation (which increases cell size) and cell division (which decreases it). To a lesser degree, size-preferential grazing also may influence mean cell size for a given population

(e.g., González et al., 1990), but for the present we assume that this effect is minor. A decrease in mean cell size over a certain time period can therefore be taken as an indication of cell division during that period, although a lack of such a decrease (at least during the light period) cannot be taken to indicate an absence of cell division.

Virtually all the available data concerning diel changes in phytoplankton cell size in natural populations are based on flow cytometrically measured light scattering. The quantitative relationship between cell size and cellular light scattering is complex, and depends on both the inherent properties of the cell (particularly its refractive index) and the angle over which light scattering is measured (Jerlov, 1976). Thus, the value of light scattering data for assessing diel variations in cell size depends in large part on the relative strength of cell size and refractive index in driving such variations. Although there is evidence from studies on phytoplankton cultures that changes in refractive index can be important (Ackleson et al., 1993; Stramski and Reynolds, 1993; Stramski et al., 1995), changes in cell size and carbon content have clearly been shown to strongly influence, and in a number of cases to be the major determinants for, light scattering changes for a given phytoplankton species (Stramski and Reynolds, 1993; Stramski et al., 1995; DuRand and Olson, 1998; DuRand et al., submitted.). The correspondence between light scattering decreases and other indicators of cell division among natural picoplankton populations support this latter contention (see below).

Diel patterns in cellular light scattering were first noted by Olson et al. (1990) for *Synechococcus* in the subtropical and temperate N. Atlantic. In that study, consistent increases in mean *Synechococcus* forward-angle light scatter (FALS) during the light period and decreases during the dark period were noted, despite considerable variability in the location of sampling. This pattern was taken to reflect cell growth throughout the light, and cell division in the early evening. In the equatorial Pacific, similar diel patterns in FALS or right-angle light scatter (RALS) have been observed among surface populations of *Prochlorococcus*, *Synechococcus*, and ultraphytoplankton, although again, the detailed behavior of these patterns sometimes varies (Binder et al., 1996; DuRand and Olson, 1996; Blanchot et al., 1997; Vaultot and Marie, 1999; Mann and Chisholm, 2000). In general, there is a good correspondence between times of decreasing FALS or RALS and times of increasing cell numbers, as would be expected if both trends reflected cell division (Fig. 1). In only one case did the decrease in light scattering lag significantly behind the increase in cell numbers (Fig. 1, series a), and in that case undercounts of *Prochlorococcus* in the afternoon may have resulted in an inaccurate assessment of the start of the daily abundance increase, as discussed above (Vaultot and Marie, 1999). As was observed for the daily increase in cell numbers, the daily decrease in light scattering is shifted toward earlier times in sub-surface populations, again consistent with the idea that decreases in scattering and increases in cell numbers are both reflective of cell division. (DuRand and Olson, 1996; Vaultot and Marie, 1999).

Cellular light scattering often is observed to remain constant during the dark period once cell division is completed, presumably reflecting unchanged cell size at these times. With the onset of light, daily increases in light scattering commence. Vaultot and Marie (1999) suggest that these increases can be delayed by a few hours after sunrise in *Synechococcus* and picoeukaryotes, but the significance of any such delay is difficult to judge at present. DuRand and Olson (1996) observed a similar delay among “nanophytoplankton”, but not among “ultraphytoplankton” or

Synechococcus; Blanchot et al. (1997) observed no such delay among picoeukaryotes.

Vaultot and Marie (1999) observed that at the end of the day RALS of *Prochlorococcus* and ultraphytoplankton (but not *Synechococcus*) continues to increase for ~2–4 and ~4–6 h, respectively, after the onset of darkness. Blanchot et al. (1997) observed a similar post-sunset increase in FALS among surface populations of *Prochlorococcus*, but not among ultraphytoplankton. In contrast, Mann and Chisholm (2000) observed only a very subtle peak in *Prochlorococcus* FALS 1 h after sunset (in unenriched waters), and DuRand and Olson (1996) observed no such increase in any picoplankton group, although in the latter study lower sampling frequency could have obscured an increase in light scattering within the first few hours after sunset. Thus, although early evening increases in light scattering do appear to occur at certain times among *Prochlorococcus* and picoeukaryote populations, the extent of this phenomenon remains to be determined.

The physical basis for increases in cellular light scattering during the dark period is not known. In those cases where the increase is small and occurs within an hour of sunset, it does not seem unreasonable to ascribe this increase to net cellular growth occurring during twilight. In the complete absence of light there can be no net increase in carbon biomass, but there can certainly be a redistribution of carbon among different cellular constituents (e.g., from low molecular weight components to macromolecules). Furthermore, cell shape, cell volume (e.g., reflecting increased or decreased water content), and refractive index may change. As discussed above, all of these factors can influence cellular light scattering properties.

The magnitude of change in mean cell size (and presumably in cellular light scattering) over the course of the diel cycle is related to the extent to which division is phased, and to the population division rate (Olson et al., 1990). For well-phased populations growing at one doubling per day, mean cellular biomass should double over the course of the day; at slower division rates a smaller fraction of the population divides on any day, and the mean diel increase in biomass is reduced; at

higher division rates, a fraction of the population will divide more than once, implying a greater increase in mean cellular biomass over the course of the day (assuming that all division is restricted to a small part of the L:D cycle, as appears to be the case at least for *Prochlorococcus* (Shalapyonok et al., 1998)). In *Prochlorococcus*, for which independent measures of division rate are possible (see below), the daily ratio of maximum to minimum RALS or FALS is indeed positively correlated with division rate (Binder et al., 1996; Vaultot and Marie, 1999). To a first approximation, then, the magnitude of diel variation in cellular light scattering (and biomass) is expected to decrease with depth in all three picoplankton groups (DuRand and Olson, 1996).

2.3. Cell division

As discussed above, cell division can be taken as the primary process responsible for diel declines in mean cell size and for diel increases in cell abundance. Because cell growth and mortality act in opposition to these trends in cell size and abundance, respectively, neither of these characteristics is a perfect proxy for cell division. Taken together, however, they can certainly provide a reliable qualitative picture of the timing of cell division in pico- and ultraphytoplankton.

An alternative, independent, and quantitative approach for assessing cell division involves DNA-based cell cycle analysis (Carpenter and Chang, 1988). This approach requires that the target population is well-phased with respect to cell division, is relatively homogeneous with respect to cell cycle behavior, is amenable to uniform and quantitative DNA staining, and is abundant enough to make possible the collection of statistically meaningful DNA frequency distributions. In the equatorial Pacific, *Prochlorococcus* is the single picophytoplankton group that satisfies these requirements. For this group, cell cycle analysis confirms the general conclusions about cell division timing based on patterns of cell abundance and light scattering, i.e. that division is well-phased, is concentrated in the late afternoon or early evening, and occurs later at the surface than at depth (Vaultot et al., 1995; Binder et al., 1996;

Liu et al., 1997; Vaultot and Marie, 1999; Mann and Chisholm, 2000).

The biological underpinnings of cell cycle regulation in *Prochlorococcus* are not well-established. It has been suggested that the delay in cell division among *Prochlorococcus* (and other picophytoplankton) at the surface is the result of UV exposure or represents an adaptive response to reduce UV damage during DNA synthesis, but this idea remains to be tested (Vaultot et al., 1995; Vaultot and Marie, 1999). Given the extraordinarily tight cell cycle phasing observed in *Prochlorococcus* populations, the recent discovery of the existence of circadian rhythms in cyanobacteria (Golden et al., 1997), and the recognition of the importance of these rhythms in controlling many cellular processes in these organisms (including DNA replication) (Liu et al., 1995; Mori et al., 1996), it is reasonable to expect that circadian rhythms may play a role in controlling the cell cycle in *Prochlorococcus*. Again, this hypothesis has not yet been tested. The observation that cell cycle timing varies with depth (Vaultot et al., 1995) is not in and of itself a strong argument against the involvement of circadian control; although the period of events under circadian control should be relatively insensitive to environmental and/or physiological conditions, the absolute timing of those events need not be so.

2.4. Grazing activity

By combining cell cycle-based estimates of the timing and rate of *Prochlorococcus* cell division with observed diel changes in *Prochlorococcus* abundance, and assuming that viral lysis does not account for a large fraction of total daily mortality (e.g., Waterbury and Valois, 1993), diel variability in *Prochlorococcus* grazing mortality can be assessed. While the results of this sort of analysis strongly suggest that grazing mortality does indeed vary over the course of the day, the observed patterns of this variation have not been consistent. Vaultot and Marie (1999) report that *Prochlorococcus* cell numbers remained approximately constant over the second half of the dark period, when according to cell cycle analysis cell division had essentially stopped. This implies that grazing

mortality is much weaker during this period than during the subsequent light period, when cell numbers dropped substantially. However, these observations are in direct contrast to those of Mann and Chisholm (2000), who observed relatively rapid cell loss in the dark period just after division, followed by much slower loss after sunrise. Liu et al. (1997) calculated the grazing mortality for *Prochlorococcus* during times of cell division (~16:00–24:00) based on the cell cycle-derived division rate and the change in cell numbers over this time period, and during times without division (~00:00–16:00) based on changes in cell number alone. These calculations indicate that at a western equatorial Pacific station (167°E), grazing mortality was higher during division than at other times of the day by a factor of 2, whereas at an eastern station (158°W) the trend was reversed, with mortality during division lower than during the rest of the day by a factor of 2. These various observations were made at different times and/or different locations, and may well reflect variation in the behavior of grazer populations under these different circumstances. It should be noted, however, that variations in other factors (such as diel mixing) also could play a role in the relatively subtle variations in cell abundance decrease observed in these studies. It is therefore difficult to draw firm conclusions at present regarding diel patterns of grazing mortality among *Prochlorococcus* (or other pico- or ultraphytoplankton) in the equatorial Pacific.

2.5. Cellular fluorescence

Diel variation in cellular fluorescence from photosynthetic pigments reflects the combined influence of pigment synthesis, cell division, and photophysiology (as it pertains to fluorescence quenching). The first two of these processes result in increases and decreases in per-cell pigment content, respectively, while the third determines the fluorescence yield of that pigment. This situation is analogous to that for cellular light scattering, in which growth and division determine cell size or biomass, and cellular properties such as refractive index determine the relationship

between size and scattering. However, whereas diel variation in light scattering appears to be primarily driven by variation in cell size (see above), diel variation in fluorescence is strongly influenced by changes in both photophysiology and cellular pigment quotas.

Vaulot and Marie (1999) present the only detailed examination to date of diel patterns in the cellular fluorescence characteristics of pico- and ultraphytoplankton in the equatorial Pacific. These authors report dramatic diel variations in cellular chlorophyll fluorescence among surface populations of both *Prochlorococcus* and *Synechococcus*. In the case of *Prochlorococcus*, cellular fluorescence decreases sharply (by a factor of 3, overall) from sunrise through noon and increases thereafter until early evening. Because cell division in this group is insignificant before noon (see above), and because directly measured (divinyl) chlorophyll-*a* per *Prochlorococcus* cell was found to be invariant during the light period, this pattern can be assumed to reflect the development and subsequent reversal of non-photochemical quenching in this surface population (Claustre et al., 1999; Vaulot and Marie, 1999). *Prochlorococcus* fluorescence shows a secondary decrease in the middle of the dark period, likely resulting from cell division (and its associated reduction in cellular chlorophyll content).

The diel cycle in *Synechococcus* chlorophyll fluorescence is similar to that in *Prochlorococcus*, except that the midday minimum is shifted later, reflecting the additional effect of decreased cellular chlorophyll content associated with *Synechococcus* cell division in the afternoon (Vaulot and Marie, 1999). Consistent with this idea, the recovery of fluorescence occurs largely during the first half of the night, and no midnight depression in fluorescence is observed. *Synechococcus* phycoerythrin fluorescence follows a similar pattern, although the magnitude of the midday decrease is considerably reduced, perhaps reflecting reduced non-photochemical quenching in the case of phycoerythrin. The nighttime increase in phycoerythrin fluorescence also appears to lag that of chlorophyll fluorescence in *Synechococcus*, suggesting perhaps an ordered sequence of pigment synthesis during the night.

The obvious midday decreases in both *Prochlorococcus* and *Synechococcus* chlorophyll fluorescence at the surface are absent at depth, as might be expected if these decreases were the result of light-dependent non-photochemical fluorescence quenching (Vaulot and Marie, 1999). In the case of *Prochlorococcus*, fluorescence in sub-surface populations increases during the day, and this increase is stronger at deeper depths, presumably reflecting photoacclimation and chlorophyll synthesis in these low-light populations.

In contrast to *Prochlorococcus* and *Synechococcus*, reproducible trends in ultraphytoplankton chlorophyll fluorescence over the course of the light period were not observed. Cellular fluorescence does appear to decrease in the second half of the dark period, coincident with cell division in this group (Vaulot and Marie, 1999).

3. Bulk water properties

3.1. Bulk fluorescence

Bulk in vivo chlorophyll fluorescence is widely used as a proxy for chlorophyll concentration, although variability in quantum yield for fluorescence is significant and complicates the interpretation of such measurements considerably (Falkowski and Kolber, 1995). As described above, cellular in vivo fluorescence is influenced by both per-cell chlorophyll content and photo-physiological factors that influence fluorescence quenching. Bulk fluorescence reflects the sum of all cellular fluorescence in a parcel of water, and in theory can therefore be decomposed into contributions from individual phytoplankton groups, each of which is the product of mean cellular in vivo fluorescence and cell concentration. Although cellular fluorescence as measured by flow cytometry (involving very high excitation energy and very short time scales) is by no means the same as that measured by standard “bulk” fluorescence instruments (involving relatively low excitation energy and long time scales), the general correspondence between the two measurements suggests that flow cytometer-derived fluorescence measurements can be used to explore the relative contributions of diel

variations in cellular fluorescence and cell abundance to diel variations in bulk fluorescence (Claustre et al., 1999).

Dandonneau and Neveux (1997) observed a dramatic and robust diel cycle of fluorescence in the surface waters of the equatorial Pacific. When normalized against 24-h fluorescence mean and standard deviation, this cycle is remarkably reproducible over seasons, years, and locations. It involves a period of fluorescence decrease from midnight until midday (~12:00–13:00), a period of increase thereafter until early evening (2–3 h after sunset), and finally a short period of constant fluorescence until midnight again. This pattern is very similar to that reported by Claustre et al. (1999) for depth-integrated fluorescence at a 5°S 150°W station. The decrease in bulk fluorescence over the first half of the day in both of these studies is strikingly similar to the pattern observed in flow cytometrically measured per-cell fluorescence in *Prochlorococcus* and *Synechococcus* (see *Cellular fluorescence*, above), suggesting that this is the result of fluorescence quenching rather than a decrease in chlorophyll standing stock (Dandonneau and Neveux, 1997; Claustre et al., 1999). This interpretation is supported by the observation that directly measured total chlorophyll-*a* (and divinyl chl-*a*) remains approximately constant in the face of strongly varying bulk fluorescence (Blanchot et al., 1997; Dandonneau and Neveux, 1997). Interestingly, unchanging total chlorophyll also implies active chlorophyll synthesis and increasing per-cell chlorophyll during this time period, since total pico- and ultraphytoplankton cell numbers are decreasing. That no such increase in cellular chlorophyll content is evident from flow cytometrically measured fluorescence could be explained by decreasing fluorescent yields at this time of day. However, as mentioned above, Vaulot and Marie (1999) report that at least for *Prochlorococcus*, directly measured per-cell chlorophyll-*a* remains invariant throughout the light period.

Diel cycles in bulk fluorescence are apparent in sub-surface waters as well, although their amplitude is diminished considerably as compared with surface waters, and the timing of the fluorescence minimum is shifted to earlier in the day (Dandonneau and Neveux, 1997; Claustre et al., 1999).

These cycles are present, although of reduced amplitude, in “warm pool” western equatorial waters as well as in upwelling eastern equatorial waters (Dandonneau and Neveux, 1997). In contrast to surface water, light-dependent non-photochemical quenching is unlikely to be involved in the diel fluorescence variation at depth. Dandonneau and Neveux (1997) argue that intrinsic physiological cycles in fluorescence yield may be responsible for diel changes in sub-surface bulk fluorescence, whereas Claustre et al. (1999) suggest that variations in cell number and/or chlorophyll cell quota may be important.

Thus, while diel cycles in bulk fluorescence are very clear in the equatorial Pacific, the competing effects on these cycles of changes in fluorescence quenching, cellular chlorophyll content, and cell abundance have yet to be fully disentangled.

3.2. *Inherent optical properties*

The inherent optical properties, those which are not influenced by the radiative light field, are comprised of absorption (a), scattering (b), and attenuation (c), where

$$c(\lambda) = a(\lambda) + b(\lambda).$$

The attenuation coefficient, c , in units of m^{-1} , is easily measured using a beam transmissometer at 660 nm or an absorption–attenuation meter (ac-9) at nine wavelengths (412, 440, 488, 510, 532, 555, 650, 676, and 715 nm). Either instrument is commonly attached to CTD-rosettes or deployed on moorings. The resulting attenuation data have been compared with particle mass concentration and significant correlations have been found (Baker and Lavelle, 1984). Frequent measurements made over the daily light cycle have yielded observations of diel variations in beam attenuation in many different areas of the world’s oceans (Siegel et al., 1989; Hamilton et al., 1990; Gardner et al., 1993, 1995), with a minimum near dawn and a maximum near dusk. This pattern is repeated on a daily basis, although the magnitude of the diel changes may vary from day to day. These diel variations have been attributed to accumulation of particles due to primary production during the day and losses of particles due to grazing at night, and

have been used to estimate particle production rates (Siegel et al., 1989).

The first observations of diel variations in beam attenuation in the equatorial Pacific were made by Cullen et al. (1992) in February–March 1988 on a cruise along 150°W. Beam attenuation increased between morning (08:30) and afternoon profiles (13:30) in the upper ~70 m with increases as great as ~50% at certain depths. These variations were found to reflect primary production quite accurately, though numerous caveats about this interpretation were presented. During this same cruise, Carr et al. (1992) found that nocturnal mixing homogenized the phytoplankton in the upper 20 m, thus eliminating some of the changes in the upper part of the water column that developed during the day.

Gardner et al. (1995) made measurements of beam attenuation in the equatorial Pacific during El Niño conditions in March–April 1992 and during non-El Niño conditions in October 1992 as part of the US JGOFS time-series cruises at 140°W. They observed strong diel variations in beam attenuation due to particles (c_p) to a depth of 60 m (Fig. 2). Increases of up to 70% occurred during the day in surface waters, with corresponding decreases at night. Gardner et al. attribute the daytime increases to primary production and the nighttime decreases to the combined effects of grazing, remineralization, production of large particles (such as aggregates), and upward mixing of particle-depleted waters from below the daytime mixed layer. The concentration of aggregates was lower in morning and noon profiles than in dusk and midnight profiles, indicating production of aggregates during the day (Walsh et al., 1997). Gardner et al. (1995) describe the physical process by which a thin daytime mixed layer caused by solar heating is deepened at night by wind mixing, upwelling and convective cooling (Fig. 2) (see also data in André et al., 1999; Vaultot and Marie, 1999). They argue that this alternate shoaling and deepening of the mixed layer operates as a “mixed-layer pump” that could have important consequences, including the downward mixing of plankton and detritus. These authors point out that the pump effect would be particularly important if the particles that are produced in

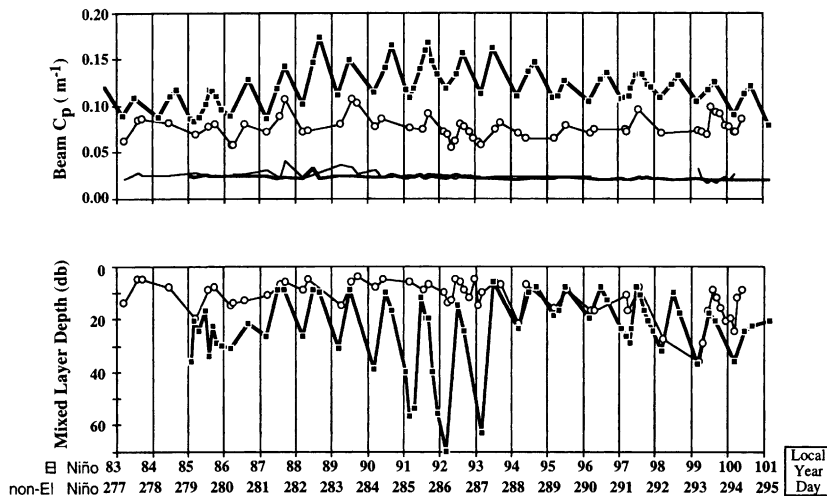


Fig. 2. Time-series data showing diel variations in beam attenuation due to particles (c_p) (top panel) and mixed-layer depth (bottom panel) during El Niño (open circles, light lines) and non-El Niño (filled squares and heavy lines) conditions at 0° 140° W in 1992. The diel variations in c_p , shown here at 15 m, were strong down to 60 m. Lines near the baseline in the upper panel are c_p at 150 m, at which depth c_p values were similar for both cruises. Vertical lines indicate midnight. Figure is from Gardner et al. (1995).

the upper part of the water column during the day and mixed downward at night can then sink below the following day's mixing depth. This would probably not be the case for pico- or ultraphytoplankton.

3.2.1. Decomposing beam attenuation

Since beam attenuation is an additive property, it can be deconstructed into its constituent elements:

$$c(\lambda) = c_w(\lambda) + c_y(\lambda) + c_p(\lambda),$$

where c_w (the contribution of pure seawater) is a constant, c_y (the contribution of dissolved organic material) is negligible at the red wavelength (660 nm) of the transmissometer in the open ocean, and c_p (the contribution due to particles) is the determining factor in variations in c . C_p can be broken down further into the contributions of different particle types, such as bacteria and different groups of phytoplankton.

DuRand and Olson (1996) used flow cytometric measurements of concentration and forward-angle light scattering to estimate the contribution of different phytoplankton groups to beam attenuation due to particles. The volume of sample analyzed by flow cytometry in this study was

similar to that measured with a 25-cm transmissometer, so the particle assemblage evaluated in both cases should be comparable. They found that usually nanophytoplankton (2–3 μ m coccoid cells), but sometimes ultraphytoplankton (1–2 μ m coccoid cells), were responsible for the greatest portion of c_p (28–46% for the two groups of autotrophic eukaryotes combined) during the two 1992 JGOFS times-series cruises, with a smaller portion due to *Prochlorococcus* (4–12%) and a negligible amount due to *Synechococcus* (<2%) (heterotrophic bacteria were not measured). These four phytoplankton groups accounted for an average of 44% (range of 32–59%) of total c_p during the near-noon sampling times of the intensive diel samplings in April and October 1992. Applying a method similar to (though with important differences from) that of DuRand and Olson, Chung et al. (1998) used flow cytometric data to determine the contribution of heterotrophic bacteria, *Prochlorococcus*, *Synechococcus*, and small autotrophic eukaryotes to beam attenuation during the two JGOFS transect cruises in 1992 (February–March and August–September). They determined that about 30% of total c_p could be explained by these groups of microorganisms, with heterotrophic bacteria (16%) and

Prochlorococcus (7%) as the major contributors, and autotrophic eukaryotes (2%) and *Synechococcus* (1%) as only minor contributors. According to this analysis, phytoplankton (i.e. *Prochlorococcus*, *Synechococcus*, and picoeukaryotes) were responsible for only ~10% of the total c_p .

The discrepancies between these two studies may reflect methodological differences in data collection and analysis. DuRand and Olson (1996) used two flow cytometric configurations to obtain their data: a high-sensitivity configuration for picophytoplankton (*Prochlorococcus* and *Synechococcus*) and a large-volume (lower sensitivity) configuration for eukaryotic phytoplankton. In this latter configuration a 50-ml sample was run and larger cells (~1–20 μm in diameter) were counted. In contrast, Chung et al. (1998) utilized a high-sensitivity configuration alone, allowing only ~100 μl of sample to be analyzed (Landry et al., 1996). This small sample size could have resulted in an undersampling of rarer (and larger) ultraphytoplankton. Furthermore, owing to the high FALS sensitivity necessary for analyzing *Prochlorococcus* populations, many of the ultraphytoplankton cells were likely “off-scale”, which would result in an underestimate of their FALS. In addition, DuRand and Olson (1996) and Chung et al. (1998) used different assumptions in their calculation of the contribution to c_p of each group. DuRand and Olson used empirical calibrations relating beam attenuation to forward light scattering in laboratory cultures of phytoplankton combined with Mie scattering theory calibrations to convert flow cytometrically measured forward scattering cross section (3–19°) to total attenuation cross section (μm^2). In contrast, Chung et al. made the assumption that the ratio between the forward light scattering (1–19°) of a cell and that of a polystyrene bead is equivalent to the ratio between the total light scattering of a cell and that of a bead. Since the angular distribution of scattering is different at different particle sizes, this assumption may be problematic.

The analysis by DuRand and Olson (1996) indicates that the observed diel variations in beam attenuation in the equatorial Pacific can be almost completely accounted for by variations in the estimated beam attenuation due to phytoplankton

(with the eukaryotic ultraplankton being the most important group). The beam attenuation due to phytoplankton increases during the day as cell size increases (offset to some extent by concurrent cell abundance decreases), and decreases at night due to factors such as cell division, respiration, and losses from grazing and mixing.

In direct contrast to the findings of DuRand and Olson, Chung et al. (1998) came to the “uncomfortable conclusion” that scattering of the four picoplankton groups they analyzed does not appear to be responsible for diel variations in beam attenuation due to particles. According to their analysis, although the calculated scattering contributions of *Prochlorococcus*, *Synechococcus*, and autotrophic eukaryote populations were generally higher at dusk than at dawn, heterotrophic bacterial scattering decreased during the day (due to decreasing cell size). As a result, the scattering coefficient (b) of the four populations combined did not show the same diel trend as that of c_p (which is higher at dusk than at dawn). Chung et al. suggest that there must be another component of the particle assemblage that was not measured in their study causing the observed diel variations in c_p .

One important potential cause for the different conclusions reached in these two studies regarding diel variations in c_p is the lack of consideration of heterotrophic bacterial light scattering by DuRand and Olson (1996). In order to estimate the contribution of heterotrophic bacteria to c_p for the JGOFS time-series stations, we have combined the flow cytometric measurements of abundance and FALS of bacteria from Binder et al. (1996) with those of phytoplankton from DuRand and Olson (1996). The two flow cytometers used for these studies were cross-calibrated with a number of different sized polystyrene microspheres and also by comparing FALS means of *Prochlorococcus* and *Synechococcus* populations from each study on approximately consecutive days at dawn and dusk in the upper 100 m of the water column ($r^2 = 0.80$, $n = 80$, for power fit to data). The data for forward scattering cross section were converted to total scattering cross section through a power fit of Mie theory calculations using a refractive index ($n = 1.05$) and absorption by cellular material

($a_{\text{cm}} = 0.1 \mu\text{m}^{-1}$; thus n' [the part of the refractive index attributed to absorption] = 0.004 at 488 nm) that are reasonable for phytoplankton (Morel and Bricaud, 1986; Aas, 1996). For each cell type, a different range of diameters was used to correspond to the ranges of forward light scattering measured on the cells in the field.

The results of these calculations indicate that for this data set, heterotrophic bacteria make a contribution to the scattering coefficient that is similar to that of *Prochlorococcus*, and that this contribution does not appear to exhibit significant variation from dawn to dusk (Fig. 3). Thus, the contribution of heterotrophic bacteria to c_p cannot account for the discrepancies between the conclusions of DuRand and Olson (1996) and Chung et al. (1998).

As discussed by Claustre et al. (1999), assumptions made when using flow cytometry data to decompose c_p are inevitable, but imperfect. These authors used the anomalous diffraction approx-

imation (with a typical diameter value for each cell type and a refractive index of 1.05 for all) to calculate the contribution of different plankton types to c_p in the tropical Pacific in November 1994 (a method that differs from that used by either DuRand and Olson (1996) or Chung et al. (1998)). Similar to the results of DuRand and Olson, they found that at the equator the picoeukaryotes are the largest contributor to c due to phytoplankton, with *Prochlorococcus* and *Synechococcus* contributing less. Claustre et al. contrast their estimates with those of Chung et al. (who found that *Prochlorococcus* contributed significantly more than the picoeukaryotes) and point out that the mean size estimate for picoeukaryotes used by Chung et al. (1.26 μm diameter) leads to a ten-fold difference in calculated scattering cross section compared to the size they used (2.28 μm), based on microscopic observations.

3.2.2. Estimates of primary productivity from beam attenuation

If diel variations in beam attenuation can be related to cell growth (addition of carbon) and thus primary production, then estimates of primary productivity (PP) could be made from this bulk optical property. Walsh et al. (1995) use the diel cycle in the integrated particle load (IPL) to estimate primary production in the equatorial Pacific. They convert beam attenuation data to particulate matter concentration (PMC) using calibrations derived from measurements in these waters of c_p and filtered particulate matter (PMC = $451 \times c_p$ for a March/April 1992 cruise, and = $642 \times c_p$ for an October 1992 cruise). The PMC estimates are then integrated to the depth of the euphotic zone to calculate the IPL. The increase in the IPL during the day is converted to carbon units by assuming that 40% of PMC is due to particulate organic carbon (POC), a conservative estimate based on data from the North Atlantic Bloom Experiment (Gardner et al., 1993). These conversions are equivalent to assuming a carbon-specific beam attenuation (c_c^* , in $\text{m}^2[\text{gC}]^{-1}$) of 5.5 for the first cruise and 3.9 for the second cruise (see further discussion, below). Walsh et al. obtained PP estimates that were about one-third those obtained by Barber et al.

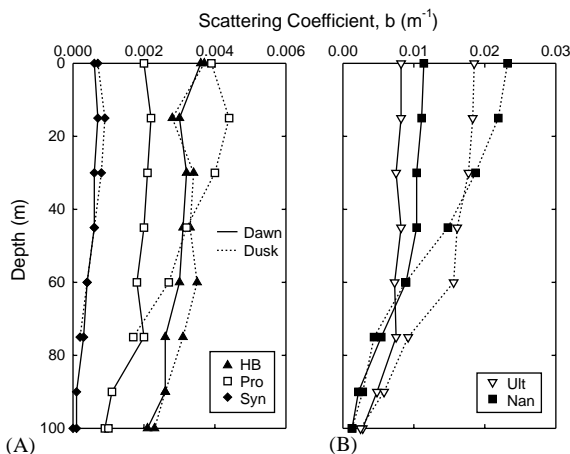


Fig. 3. Depth profiles of the estimated scattering coefficients (b , m^{-1}) for (A) heterotrophic bacteria (HB), *Prochlorococcus* (Pro), and *Synechococcus* (Syn), and (B) eukaryotic ultraphytoplankton (Ult) and nanophytoplankton (Nan) at dawn (solid lines) and dusk (broken lines) at 0° 140°W on 2 October (HB) and 5 October (all other populations) 1992. The scattering coefficients for Pro, Ult, and Nan all increase substantially (\sim doubling) from dawn to dusk in the upper part of the water column. Syn shows small increases at the upper depths and HB shows either small increases or decreases, depending on the depth. Note that the two panels are scaled differently. Calculations based on data from Binder et al. (1996) and DuRand and Olson (1996) (see text for details).

(1994) using the ^{14}C method. This difference is perhaps not surprising, given that the IPL approach would be expected to reflect a sort of “community net production” that would not include primary production that was removed by grazing, respiration, or aggregation.

Claustre et al. (1999) estimated net community production rates in the equatorial Pacific using a direct conversion between POC and c_p from samples at 5°S , 150°W . The conversion used is equivalent to a carbon-specific beam attenuation (c_c^*) of 2.0, considerably lower (thus leading to higher estimates of production) than that used by Walsh et al. (1995). Their resulting estimates of net community production rates were similar (within one sd) to those from ^{14}C -based estimates.

A potential complication of this approach for estimating primary production is the possibility of variable carbon-specific beam attenuation. In laboratory studies with both eukaryotic and cyanobacterial phytoplankton, c_c^* has been found to vary over the diel cycle by 25–30% within a given species, and by $\pm 21\%$ of the mean among different species (mean c_c^* ranging from $2.5\text{--}3.8\text{ m}^2[\text{gC}]^{-1}$) (Stramski and Reynolds, 1993; Stramski et al., 1995; DuRand and Olson, 1998; DuRand et al., submitted.). Therefore, assuming a constant c_c^* in the field introduces significant uncertainty in the primary production estimate. It may be necessary to consider the composition of the phytoplankton community in a given body of water and at a given time in order to interpret diel variations in beam attenuation in terms of production with any confidence (DuRand and Olson, 1996).

3.2.3. Estimates of phytoplankton growth rate from beam attenuation

Diel variability in c_p can be used to estimate phytoplankton growth rate as well as daily production rate. Net *specific* particle production rate (r , time^{-1}) over a given period of observation (t) can be calculated as

$$r = \frac{1}{t} \ln \frac{c_p[t]}{c_p[0]}$$

where $c_p[t]$ and $c_p[0]$ are the observed c_p at the end and the beginning of the period, respectively

(Siegel et al., 1989). Unlike c_p -based estimates of absolute production (units, C time^{-1}) such estimates of specific particle production rates are independent of the absolute value of the carbon-specific beam attenuation (c_c^*). However, like c_p -based estimates of production they do assume that c_c^* is constant over the observation period, an assumption that is unlikely to be true (see discussion above), and that can have a significant impact on the growth rate estimate (Cullen and Lewis, 1995). In the context of phytoplankton growth, r represents a net increase during some portion of the day, which (under steady state) must be balanced over the course of the entire day by grazing or other loss terms. Estimation of the specific growth rate of phytoplankton (μ) therefore requires an assumption about the diel variation in grazing pressure (or lack thereof). More significantly, these calculations assume that phytoplankton are the sole contributors to c_p , an assertion that is clearly false (see discussion above), and one that can strongly bias the growth rate estimates (Siegel et al., 1989; Cullen et al., 1992). To the extent that phytoplankton are responsible for a large part of the diel variation in c_p (DuRand and Olson, 1996), but contribute considerably $< 100\%$ of the absolute magnitude of c_p (DuRand and Olson, 1996; Chung et al., 1998; Claustre et al., 1999), true phytoplankton growth rate is expected to be higher than that calculated as above (Siegel et al., 1989; Cullen et al., 1992). It is worth noting that the observed daytime increase in c_p , and therefore the calculated net and gross production growth rates (r and μ), largely reflect cell size increases rather than increases in cell number, as is often implicitly assumed. Whereas increases in c_p occur during the light period, and are perhaps highest during the first half of that period (see below), cell division among pico- and ultraphytoplankton is restricted to the late afternoon and early evening (see Section 2.1, above). At steady state, biomass growth rate must equal cell number growth rate over the course of 24 h; nevertheless, as discussed previously these processes are temporally uncoupled, and the c_p -based approach for measuring growth rate is based on the former.

Cullen et al. (1992) were the first to apply this approach for estimating phytoplankton growth

rate in the equatorial Pacific. Using pairs of morning and early afternoon measurements of c_p at 150°W, and assuming constant grazing mortality over the day, these authors calculated the net growth rate (r) and specific phytoplankton growth rate (μ) to be 0.85 and 1.46 d⁻¹, respectively, in the euphotic zone at this location in March 1988. This calculated μ , corresponding to >2 doublings d⁻¹, is considerably higher than growth rates measured in the equatorial Pacific using diel changes in DNA distributions (Vaulot et al., 1995; Binder et al., 1996; Liu et al., 1997; Vaulot and Marie, 1999), diel changes in cell numbers (André et al., 1999), or dilution incubations (Landry et al., 1995). Furthermore, correction for the presence of non-phytoplankton contributors to c_p increases the estimated μ to 3.48 d⁻¹, a biologically unrealistic value (Cullen et al., 1992).

The high growth rate estimates of Cullen et al. (1992) could have resulted at least in part from the extrapolation of the observed rate of change of c_p between 08:30 and 13:30 to the entire light period. The data of Claustre et al. (1999) suggest that the rate of increase in c_p during the first half of the light period is often greater than during the second. Indeed, mean r (integrated over the euphotic zone) calculated from the directly measured dawn and dusk c_p values in this data set is only 0.24 d⁻¹ at 5°S 150°W (in November 1994). (Note that this rate is recalculated from that stated originally in Claustre et al. to reflect the fact that growth occurs only during the 12 h light period.) Walsh et al. (1995) calculated very similar r values from diel c_p measurements at the equator at 140°W in March and October 1992. Assuming that grazing in the dark period balances this net daytime production, and that grazing continues at the same rate throughout the day, these net rates translate into gross phytoplankton growth rates on the order of 0.7 d⁻¹, which is much more in keeping with the independent μ estimates cited above. Nevertheless, if we recognize that phytoplankton are responsible for perhaps only ~40% of the total c_p in these waters (Claustre et al., 1999), the c_p -based mean phytoplankton growth rate increases to ~1.77 d⁻¹, an estimate that again appears to be unrealistically high.

Cullen and Lewis (1995) suggest that the assumption of constant c_c^* over the course of the L:D cycle may result in overestimates of growth rate derived from c_p measurements. As these authors point out, similar assumptions involving constant grazing mortality could likewise result in overestimates of μ . Clearly, a better understanding of the diel behavior of both of these factors is required before phytoplankton growth rates can be derived with any confidence from diel variation in beam attenuation.

4. Conclusions

Diel cycles in the biological and optical characteristics of surface waters in the equatorial Pacific are significant and well-documented. Among the pico- and ultraphytoplankton, strong diel variations in cell number and cell size (as reflected by flow cytometrically measured light scattering) have been consistently observed. These variations clearly result to a large degree from the interplay between photosynthetic biomass increase on one hand and cell division on the other. Whereas photosynthesis is obviously restricted to the daylight hours, there is no clear a priori reason that cell division should occur at a particular time of the day, nor that that time should be the same among different phytoplankton groups. Yet, in the equatorial Pacific at least, cell division does indeed occur at similar times (afternoon–early evening) among *Prochlorococcus*, *Synechococcus*, and ultraphytoplankton populations. Therefore, although the details of the diel behavior of these groups can vary, a generalized cycle can be recognized, involving cell size increases over the course of the light period followed by size decreases concomitant with increases in cell abundance in the late afternoon/early evening. Despite the qualitative consistency of this cycle, a complete quantitative understanding of the factors underlying the cycle must also take into account changes in the optical properties of cells (e.g., refractive index), and must account for loss terms such as grazing, the diel behavior of which has not yet been well-characterized. It is worth noting that diel variability in grazing need not be predicated upon specific diel

behavior among grazers: rather, it could result from simple functional or numerical responses by these grazers to the diel changes in phytoplankton size and abundance described above.

As is the case for cell size and abundance, diel variation in phytoplankton cellular fluorescence (at least among *Prochlorococcus* and *Synechococcus*) is also strongly influenced by cell growth and division. However, other factors such as non-photochemical quenching and light acclimation appear to exert considerable influence as well. These factors are not well-characterized in equatorial Pacific waters. The relatively heterogeneous diel behavior of cellular fluorescence among pico- and ultraplankton groups (contrasting the consistent behavior with respect to cell growth and division) may reflect variability in physiological responses to light among these groups.

Diel variation in bulk optical properties of surface water in the equatorial Pacific stem largely from variation in the concentration and optical properties of the phytoplankton cells suspended within these waters. However, because individual cell properties and bulk properties are necessarily measured in different ways, translating between the two is not trivial. To our knowledge there has not yet been an attempt to quantitatively account for observed diel variations in bulk fluorescence based on (flow cytometrically) measured individual cellular fluorescence characteristics. Diel variation in beam attenuation and in individually measured cellular light scattering properties have been quantitatively related, and at least in some cases appear to be mutually consistent.

Interpretation of diel cycles in beam attenuation in terms of primary production and/or phytoplankton growth rate remains somewhat problematic. The same factors that cloud our understanding of variations in individual cellular light scatter also limit our understanding of the variation in this bulk property. Accurate estimates of the carbon-specific attenuation coefficient (and the diel variation thereof) are essential. Of particular value would be an understanding of how this coefficient is influenced by phytoplankton community structure. The relative contribution of non-phytoplankton particles to beam attenuation must also be more fully characterized before

phytoplankton growth rate estimates can be made with any precision. From an ecological point of view, our poor understanding of diel variability in phytoplankton grazing mortality strongly limits the utility of c_p -based estimates of productivity and growth rate. Without accounting for loss terms, these estimates represent a poorly defined net production (or growth rate) value that may reflect the degree of diel uncoupling between production and loss as much as it reflects production itself.

There is clearly much to be learned before observations of diel cycles in surface water biological and optical characteristics can be fully understood or interpreted. Nevertheless, as the issues discussed above are addressed, these observations will likely provide important insights into the biology and ecology of surface waters in the equatorial Pacific, and elsewhere.

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