

NOTES

Limnol. Oceanogr., 53(3), 2008, 1181–1185
© 2008, by the American Society of Limnology and Oceanography, Inc.

Growth rates of six coccolithophorid strains as a function of temperature

Abstract—We determined growth rates of six coccolithophorid strains (five species) as a function of temperature. We grew four strains (three species: *Emiliania huxleyi*, *Gephyrocapsa oceanica*, and two strains of *Calcidiscus leptoporus*) at six temperatures between 6°C and 25°C, *Coccolithus braarudii* at four temperatures, and *Syracosphaera pulchra* at two temperatures. The growth rates were to a large extent consistent with the biogeographical distributions of these species. *C. braarudii* grows relatively fast at low temperatures, the two strains of *C. leptoporus* have temperature optima of 12°C and 20°C, *E. huxleyi* has an optimum at 20°C, and the growth rate of *G. oceanica* (and *S. pulchra*) increases up to the highest tested temperature of 25°C. This shows that maximum growth rate is an important factor in controlling distribution in the ocean, but it is not the only one.

Laboratory experiments on coccolithophores are heavily biased toward *Emiliania huxleyi*. From a physiological research perspective, it makes sense to work with an organism about which a lot is known. However, when one wants to consider what happens in the ocean, for instance, by modelling the role of coccolithophores in biogeochemical cycles, this approach is not satisfactory (Le Quéré et al. 2005). Blooms of *E. huxleyi* dominate the signal visible from satellites (Brown and Yoder 1994; Iglesias-Rodríguez et al. 2002), but because of the small size of its coccoliths (Young and Ziveri 2000), they do not dominate the sedimentation fluxes of coccolithophores as determined with sediment traps (Broerse 2000).

Thus, most physiological data are based on a species that may be a minor player in terms of carbonate flux. In an initial attempt to address this skewed situation, we determined the growth rate of six strains from five species of coccolithophore at six temperatures, although we did not test all strains at all temperatures.

Materials and methods—We used the following strains of coccolithophores because they represent some of the most abundant species, where abundance maxima range from high to low latitudes (McIntyre and Bé 1967): *Coccolithus braarudii* N476-2, *Calcidiscus leptoporus* NS10-2, *Calcidiscus leptoporus* N482-1, *Emiliania huxleyi* TQ26DIP (diploid, naked), *Gephyrocapsa oceanica* NS6-2, and *Syracosphaera pulchra* N10. These strains were provided by the Algbank culture collection, Caen, France (http://www.nhm.ac.uk/hosted_sites/ina/CODENET/caencultures.htm). All strains were unialgal but not axenic. *Coccolithus braarudii* N476-2 is identified as *Coccolithus pelagicus* in the Algbank collection, but here we follow the new

taxonomy of Sáez et al. (2003). We measured the coccoliths of *Calcidiscus leptoporus* N482-1 and *Calcidiscus leptoporus* NS10-2. They were both the intermediate size (6 µm) variety, and we will therefore refer to both strains as the same species (Sáez et al. 2003).

Cultures were grown in 250 mL of medium in 500-mL Erlenmeyer flasks. The medium used was K/5, based on filtered natural seawater (Keller et al. 1987; http://ccmp.bigelow.org/CI/K_family.html), but the beta-glycerophosphate was replaced with 7.2 µmol L⁻¹ NaH₂PO₄. Cultures were grown in a 14 : 10 light : dark (LD) cycle. Light intensities were measured with a Biospherical Instruments QSL 2101. Light intensity was 300 µmol photons m⁻² s⁻¹ at 6°C, and 180 µmol photons m⁻² s⁻¹ in the experiments at 9°C, 12°C, 15°C, 20°C, and 25°C. Stock cultures were maintained at 15 °C. Experimental cultures were inoculated at about 1,000 cells mL⁻¹.

Samples were taken daily about an hour after the light came on directly after gently swirling the cultures by hand. In one experiment at 20°C, cultures were swirled continuously on a rotary shaker at 60 rpm. Samples were acidified (except for the naked strain of *E. huxleyi*) by addition of 3.6 mmol L⁻¹ HCl final concentration to dissolve coccoliths, in order to accurately measure cell volume. Samples were counted in triplicate on a MultiSizer 3 Coulter Counter with an aperture tube orifice diameter of 100 µm, generally within three hours of sampling.

All particles within the diameter range of living cells were counted as cells (with a different diameter range for each strain). In all but the two slowest growing experiments, cells were counted until they visibly started deviating from logarithmic growth. Growth rates were calculated as the slope of a linear regression of ln(cell counts) against time over the logarithmic nutrient-saturated growth phase. After trying out threshold residual sum of squares (RSS) as a criterion for determining the logarithmic growth phase, we found no consistency in the increase of RSS at the start of the stationary phase and instead determined the logarithmic growth phase by eye, which in most cases was easy, and in almost all cases gave very similar results when one measurement more or less was included. In most cultures, the logarithmic phase started on day 3. At this point, in most cultures, the cell concentration was at least 2,000 cell mL⁻¹, which gave a typical standard deviation of 2% in triplicate countings of the same sample. The growth rates were determined from on average 11 samples at 4°C, 8 at 15°C, and 5 samples at 25°C. The standard error of growth rate was estimated from the linear regression

statistics. Most growth rates were determined from a single culture at each temperature. The experiment at 15°C was the only one that was repeated. When cell counts declined, the growth rate could not be determined and is shown as 0.

We compared three equations for growth rate (μ) as a function of temperature (T), linear:

$$\mu_{\max} = \mu_{\max,0^\circ\text{C}} + \text{slope} \times T \quad (1)$$

exponential:

$$\mu_{\max} = \mu_{\max,0^\circ\text{C}} \times Q_{10}^{(T/10)} \quad (2)$$

and optimal (Schoemann et al. 2005):

$$\mu_{\max} = \mu_{\text{opt}} \exp\left\{-\left[\frac{(T - T_{\text{opt}})^2}{dT^2}\right]\right\} \quad (3)$$

Parameters and their standard errors were estimated with a nonlinear model fitting function. To compare how well the equations fit the measured growth rates, we calculated Akaike's Information Criterion (Burnham and Anderson 1998):

$$\text{AIC} = n_{\text{obs}} \log(\sigma^2) + 2n_{\text{param}} \quad (4)$$

in which n_{obs} is the number of observations, n_{param} is the number of parameters, and

$$\sigma^2 = 1/(n_{\text{obs}} - n_{\text{param}}) \times \sum (\mu_{\text{obs}} - \mu_{\text{fit}})^2 \quad (5)$$

in which μ_{obs} is the growth rate of each species at each temperature, and μ_{fit} is the fit to previous equation 1, 2, or 3.

The μ_{\max} values at the optimum temperature of the five strains for which an optimum temperature could be determined were fit to the equation (Sarhou et al. 2005):

$$\mu_{\max} = x \times V^y \quad (6)$$

in which V is the cell volume of each species.

Results—The growth rate of *Coccolithus braarudii* showed the smallest decrease with decreasing temperature and was optimal at 15°C (Fig. 1A). The temperature optima of the two *Calcidiscus leptoporus* strains were 12°C and 20°C, respectively. *Emiliania huxleyi* grew faster than the other coccolithophores at all temperatures up to 20°C, at which temperature its growth was optimal. Of the four strains that were measured at all six temperatures, the growth rate of *Gephyrocapsa oceanica* was the only one to still increase with temperature at 25°C. The growth rate of *Syracosphaera pulchra* was only measured at 9°C and 25°C, and there was no growth at 9°C.

At 20°C, growth rates were measured both in cultures that were swirled daily and in cultures that were swirled continuously at 60 rpm. *G. oceanica*, both strains of *C. leptoporus*, and *C. braarudii* grew more slowly when swirled continuously. Only *E. huxleyi* grew faster when swirled continuously. Because most cultures grew more slowly when swirled continuously, cultures were gently swirled only once a day in the experiments at the other temperatures.

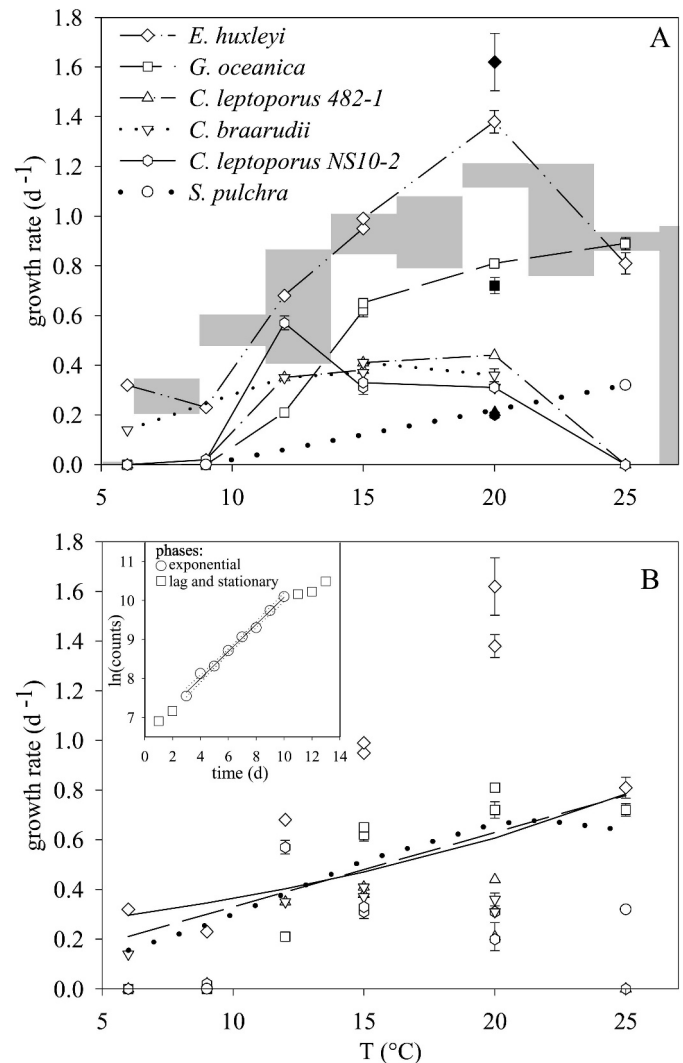


Fig. 1. Growth rates of coccolithophores as a function of temperature. (A) Errors smaller than 0.025 d^{-1} are not shown. Open symbols: swirled daily; closed symbols: swirled continuously at 60 rpm. Lines connect the cultures that were swirled daily. Gray bars: literature values for *E. huxleyi*. (B) Dashed line: linear growth increase; solid line: exponential growth increase; dotted line: optimal growth function. (B, inset) $\ln(\text{cell counts})$ versus time after inoculation (in days) for *C. braarudii* at 12°C; growth rate and SE are based on the circles.

We fitted the measured growth rates to linear, exponential, and optimal functions of temperature (Fig. 1B). We tested the fits to all positive growth rates, to the growth rates excluding the shaken cultures, and to the growth rates excluding both the shaken cultures and the growth rates of *C. leptoporus* NS10-2 and *C. leptoporus* N482-1 at 9°C, which were close to, though significantly different from, 0 (Table 1). The best fits were obtained using all growth rates (excluding nongrowth results). Based on the AIC value, there is substantial support for both the linear and the exponential equation, and although the optimal function had a slightly lower sum of squared residuals, there was not enough information to justify the extra parameter in this equation.

Table 1. Fits of the measured growth rates to linear, exponential, and optimal functions of temperature (n is the number of cultures: six strains at six temperatures, including five duplicates at 15°C. Not all strains were grown at all temperatures.).

	Parameter*	All data ($n=33$)	Nonshaken ($n=28$)	Nonshaken excluding <i>Calcidiscus</i> at 9°C ($n=26$)
Linear	$\mu_{\max,0^\circ\text{C}}$ (d^{-1})	0.03 ± 0.19	0.14 ± 0.17	0.14 ± 0.17
	slope	0.030 ± 0.012	0.025 ± 0.011	0.025 ± 0.011
	ΔAIC	0	0.2	2.2
Exponential	$\mu_{\max,0^\circ\text{C}}$ (d^{-1})	0.22 ± 0.09	0.22 ± 0.08	0.27 ± 0.09
	Q_{10}	1.7 ± 0.4	1.7 ± 0.3	1.5 ± 0.3
	ΔAIC	0.4	0.8	2.5
Optimum	μ_{opt} (d^{-1})	0.68 ± 0.10	0.71 ± 0.10	0.69 ± 0.10
	T_{opt} ($^\circ\text{C}$)	21 ± 6	22 ± 3	23 ± 5
	dT ($^\circ\text{C}$)	13 ± 4	12 ± 4	15 ± 8
	ΔAIC	2.1	2.0	4.4

* See text for definitions.

The maximum growth rate is a function of cell volume: $\mu_{\text{opt}} = 3.5V^{-0.32}$, $r^2 = 0.86$ (Fig. 2).

Discussion—Most growth rates were measured on only one culture at each temperature. However, the reproducibility of the measured growth rates at 15°C was good (average absolute standard deviation [SD] = 0.02 d^{-1} , relative SD = 5%, $n = 5$). In addition, the errors calculated from the logarithmic growth phases were similarly low (see Fig. 1B inset, average absolute standard error [SE] = 0.02 d^{-1} , relative SE = 6%, $n = 33$) when the algae grew at that temperature. This suggests that the maximum growth rates are reliable.

Between 9°C and 25°C, cultures were grown in the same incubator with a light intensity of 180 $\mu\text{mol photons m}^{-2} \text{s}^{-1}$. The experiment at 6°C was done in a different incubator, in which light intensity was higher at 300 $\mu\text{mol photons m}^{-2} \text{s}^{-1}$. This is probably the reason why growth rate in *E. huxleyi* was higher at 6°C than at 9°C. Of the 13 studies that were compared by Nanninga and Tyrrell (1996), eight reported light saturation of *E. huxleyi* growth occurring above 180 $\mu\text{mol photons m}^{-2} \text{s}^{-1}$. However, the synthesis of

data by Le Vu (2005) for *E. huxleyi* estimated a half saturation constant ($K_{1/2}$) of 3.86 $\text{mol photons m}^{-2} \text{d}^{-1}$, which in our case translates to a $K_{1/2}$ value of 77 $\mu\text{mol photons m}^{-2} \text{s}^{-1}$ or light saturation at around 150 $\mu\text{mol photons m}^{-2} \text{s}^{-1}$. The growth rate measured by Houdan et al. (2006) for *C. leptoporus* NS10-2 was 0.41 d^{-1} , and for *C. braarudii* (a different strain from ours), it was 0.48 d^{-1} , both at 17°C and 25 $\mu\text{mol photons m}^{-2} \text{s}^{-1}$, which is higher than our growth rates interpolated to that temperature. On the other hand, at 300 $\mu\text{mol photons m}^{-2} \text{s}^{-1}$ and 35 psu, Schouten (2006) found a similar growth rate at 10°C and lower growth rates at 15°C and 21°C for *E. huxleyi* and similar growth rates at 15°C and 21°C for *G. oceanica* (both different strains from ours). This suggests that these four species were not light limited at 180 $\mu\text{mol photons m}^{-2} \text{s}^{-1}$.

We used a naked strain of *E. huxleyi*. Paasche and Klaveness (1970) found that a naked strain grew 15% slower than a calcifying strain, while Lecourt et al. (1996) found that a naked strain grew ~15% faster than a calcifying strain. In both cases, the naked strain was derived from the calcifying strain, and we compared the nitrate-grown, light-saturated cultures. These are relatively small differences in growth rates relative to the differences with temperature and among strains. The growth rates of our naked strain are quite similar to published growth rates of calcifying strains of *E. huxleyi* grown under at least 100 $\mu\text{mol photons m}^{-2} \text{s}^{-1}$ (gray bars in Fig. 1A; Le Vu 2005).

In the dinoflagellate *Gymnodinium nelsonii*, cell division was completely suppressed when swirled continuously at 100 rpm. Berdalet (1992) suggested that swirling might cause disturbance of the microtubule structure that divides the nucleus during mitosis. This could also have caused the reduced growth rates in our experiment when swirled continuously at 60 rpm.

The exponential fit and the linear fit to the data were very similar; so, not surprisingly, the AIC values were also very similar (Table 1), but extrapolation of these fits to the full range of temperatures in the ocean would lead to rather different predictions. The extrapolated growth rate at 0°C for the exponential fit is seven times that for the linear fit. A Q_{10} value of 1.7 ± 0.4 is slightly lower than the widely used results of Eppley (1972), who estimated $Q_{10} = 1.9$ for all phytoplankton using the largest measured growth rate at each temperature only.

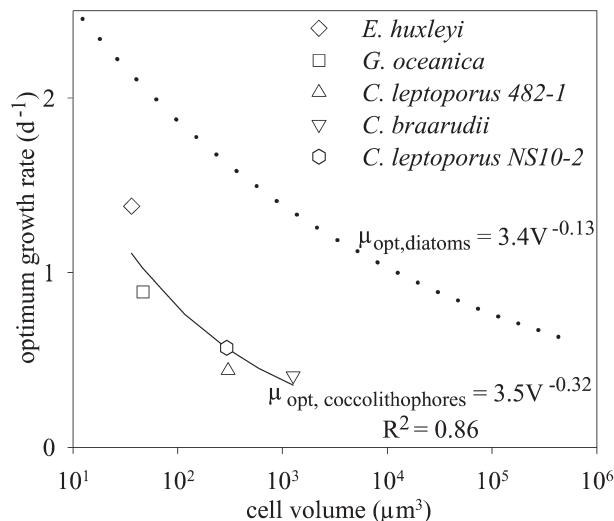


Fig. 2. Maximum growth rate at the optimum temperatures as a function of cell volume for cultures that were swirled daily. Symbols are as in Fig. 1. For reference, the function for diatoms (Sarthou et al. 2005) is plotted as the dotted line.

To a first approximation, the observed temperature minima for growth are consistent with the biogeography of these coccolithophores as determined from coccolith abundance in surface samples and surface sediments (McIntyre and Bé 1967). *Coccolithus braarudii* grew relatively best at low temperatures. That is, though growth rate decreased with temperature, it decreased much more slowly than the growth rate of *Emiliania huxleyi*, while the other species did not grow at all at 6°C. This is also consistent with its abundance in sediment trap samples from the northern North Atlantic (Knappertsbusch and Brummer 1995). The two strains of *Calcidiscus leptoporus* showed somewhat different temperature optima at 12°C and 20°C. *C. leptoporus* is a rather cosmopolitan species; its distribution is bounded by the 8°C isotherm (McIntyre and Bé 1967). Indeed, neither strain grew at 6°C and grew at only 0.02 d⁻¹ at 9°C. However, they did not grow at 25°C either, suggesting that there is a tropical strain that completely replaces these more temperate strains. *E. huxleyi* has an even more cosmopolitan distribution pattern and is found everywhere except in polar regions with a summer temperature of less than 2°C. Consistent with its documented distribution, it was the only one of the four strains tested at all six temperatures that grew at all temperatures. An optimum growth rate of 1.4–1.6 d⁻¹ at a temperature of 20°C is fairly typical when compared to other strains of *E. huxleyi* as reviewed by Le Vu (2005). The growth rate of *Gephyrocapsa oceanica* was the only one to still increase at 25°C, while it did not grow at 6°C and 9°C. Again, this is consistent with its biogeographical distribution equatorward of the 14°C or 15°C isotherm (McIntyre and Bé 1967; Ziveri et al. 2004). The growth rate of *Syracosphaera pulchra* was only measured at two temperatures, but despite the limitations of these data, they are still consistent with the subtropical abundance maximum of *S. pulchra* (Ziveri et al. 2004).

Coccolithophores are considered to be K-strategists that specialize in efficient nutrient uptake at the expense of a high maximum growth rate, which characterizes r-strategists. Our finding, that the minimum growth temperature defines biogeography, suggests a refinement of this ecological rule: that the difference in growth rate between K- and r-strategists should be considered at the optimum growth temperature, while the minimum growth temperature is shown to be important for K-strategists as well. Indeed, our results at the optimum temperatures confirm that coccolithophores grow considerably slower than diatoms of the same size (Fig. 2).

Unfortunately, of the four species that have been found to dominate in the subtropics, both at Bermuda in the Atlantic Ocean (Haidar and Thierstein 2001) and HOT in the Pacific Ocean (Cortes et al. 2001)—*Umbellosphaera irregularis*, *U. tenuis*, *Florisphaera profunda*, and *E. huxleyi*—only the latter is available in culture collections. In low latitudes, the contribution of CaCO₃ to surface sediments is higher than at high latitudes, and, given that half of the surface ocean is above 22°C (Locarnini et al. 2006; <http://www.nodc.noaa.gov/OC5/WOA05/wao5data.html>), this lack of testable strains limits what can be said about coccolithophorid physiology on a global scale. That

is not to say that further progress beyond this study is not possible. Other less abundant coccolithophorid species that have been isolated from the tropics may be used.

E. huxleyi is the fastest growing coccolithophore of those we tested up to 20°C, and, numerically, its coccoliths are the most abundant in the surface ocean (McIntyre and Bé 1967), although this is complicated by the fact that *E. huxleyi* is the only coccolithophore that continues coccolith production beyond a complete coccosphere. However, its coccoliths are rather small (Young and Ziveri 2000), and Broerse (2000) calculated from sediment trap data and CaCO₃ content per coccolith that *G. oceanica* and *C. pelagicus/braarudii* are the two coccolithophores that contribute most to the global sedimentation flux of CaCO₃. Therefore, maximum growth rate cannot be the only factor that controls the productivity of these species. Indeed, productivity is the product of growth rate and biomass, and while growth rate is controlled bottom-up, biomass is controlled both bottom-up and top-down, primarily by grazing (Holligan et al. 1993). Grazing control is a function of size, since both phytoplankton and zooplankton growth rates decrease with size. The maximum growth rate at the optimum temperature is a function of cell volume (Fig. 2), as was found for diatoms (Sarhou et al. 2005). Cell volume decreased with temperature, but this was a small effect relative to the differences in cell volume between species.

In conclusion, we have shown here that there is a direct link between minimum growth temperature and biogeography. However, the physiological characteristics of coccolithophores as a biogeochemically important phytoplankton group are very poorly characterized, and we hope that this paper will inspire further work in this vein.

Erik T. Buitenhuis^{1,2}

Tanja Pangerc¹

Daniel J. Franklin

Corinne Le Quéré¹

Gill Malin

Laboratory for Global Marine and Atmospheric Chemistry
School of Environmental Sciences
University of East Anglia
Norwich NR4 7TJ, United Kingdom

¹ Also at: British Antarctic Survey, High Cross, Madingley Road, Cambridge CB3 0ET, United Kingdom.

² Corresponding author (martinburo@email.com).

Acknowledgments

We thank Ian Probert for providing the cultures from the CODENET/AlgoBank culture collection, and two reviewers for their helpful criticism of a draft of this paper. This study was supported by the European Union (CarboOcean 511176[GOCE] to E.T.B., FAASIS MEST-CT-2004-514159 to T.P.) and the U.K. Natural Environment Research Council (MarQUEST NE/C516079/1 to E.T.B., NER/A/S/2002/00917 to G.M., Advanced Research Fellowship NE/B501039/1 to G.M., and NER/H/S/1999/00176 to the Laboratory for Global Marine and Atmospheric Chemistry).

References

- BERDALET, E. 1992. Effects of turbulence on the marine dinoflagellate *Gymnodinium nelsonii*. *J. Phycol.* **28**: 267–272.
- BROERSE, A. 2000. Coccolithophore export production in selected ocean environments: Seasonality, biogeography and carbonate production. Ph.D. thesis, Vrije Universiteit Amsterdam, Netherlands.
- BROWN, C. W., AND J. A. YODER. 1994. Coccolithophorid blooms in the global ocean. *J. Geophys. Res.* **99**: 7467–7482.
- BURNHAM, K. P., AND D. R. ANDERSON. 1998. Model selection and inference, a practical information-theoretic approach. Springer.
- CORTES, M. Y., J. BOLLMANN, AND H. R. THIERSTEIN. 2001. Coccolithophore ecology at the HOT station ALOHA, Hawaii. *Deep-Sea Res. II* **48**: 1957–1981.
- EPPLEY, R. W. 1972. Temperature and phytoplankton growth in the sea. *Fish. Bull.* **70**: 1063–1085.
- HAIDAR, A. T., AND H. R. THIERSTEIN. 2001. Coccolithophore dynamics off Bermuda (N. Atlantic). *Deep-Sea Res. II* **48**: 1925–1956.
- HOLLIGAN, P. M., AND OTHERS. 1993. A biogeochemical study of the coccolithophore, *Emiliania huxleyi*, in the North Atlantic. *Glob. Biogeochem. Cy.* **7**: 879–900.
- HOUDAN, A., I. PROBERT, C. ZALYLYNY, B. VÉRON, AND C. BILLARD. 2006. Ecology of oceanic coccolithophores. I. Nutritional preferences of the two stages in the life cycle of *Coccolithus braarudii* and *Calcidiscus leptoporus*. *Aquat. Micr. Ecol.* **44**: 291–301.
- IGLESIAS-RODRÍGUEZ, M. D., C. W. BROWN, S. C. DONEY, J. KLEYPAS, D. KOLBER, Z. KOLBER, P. K. HAYES, AND P. G. FALKOWSKI. 2002. Representing key phytoplankton functional groups in ocean carbon cycle models: Coccolithophorids. *Global Biogeochem. Cy.* **16**: 1100, doi:10.1029/2001GB001454.
- KELLER, M. D., R. C. SELVIN, W. CLAUS, AND R. R. L. GUILLARD. 1987. Media for the culture of oceanic ultraphytoplankton. *J. Phycol.* **23**: 633–638.
- KNAPPERTSBUSCH, M., AND G.-J. A. BRUMMER. 1995. A sediment trap investigation of sinking coccolithophorids in the North Atlantic. *Deep-Sea Res. I* **42**: 1083–1109.
- LECOURT, M., D. L. MUGGLI, AND P. J. HARRISON. 1996. Comparison of growth and sinking rates of non-coccolith- and coccolith-forming strains of *Emiliania huxleyi* (Prymnesiophyceae) grown under different irradiances and nitrogen sources. *J. Phycol.* **32**: 17–21.
- LE QUÉRÉ, C., AND OTHERS. 2005. Ecosystem dynamics based on plankton functional types for global ocean biogeochemistry models. *Global Change Biol.* **11**: 2016–2040, doi:10.1111/j.1365-2486.2005.001004.x.
- LE VU, B. 2005. La biocalcification dans l'océan actuel à travers l'organisme modèle *Emiliania huxleyi*. Quand la mer devient blanche. Ph.D. thesis, Université Pierre et Marie Curie.
- LOCARNINI, R. A., A. V. MISHONOV, J. I. ANTONOV, T. P. BOYER, AND H. E. GARCIA. 2006. World Ocean Atlas 2005. Volume 1: Temperature. U.S. Government Printing Office.
- MCINTYRE, A., AND A. W. H. BÉ. 1967. Modern coccolithophorids of the Atlantic Ocean: I. Placoliths and cyrtoliths. *Deep-Sea Res.* **14**: 561–597.
- NANNINGA, H. J., AND T. TYRRELL. 1996. Importance of light for the formation of algal blooms by *Emiliania huxleyi*. *Mar. Ecol. Prog. Ser.* **136**: 195–203.
- PAASCHE, E., AND D. KLAVENESS. 1970. A physiological comparison of coccolith-forming and naked cells of *Coccolithus huxleyi*. *Arch. Microbiol.* **73**: 143–152.
- SÁEZ, A. G., I. PROBERT, M. GEISEN, P. QUINN, J. R. YOUNG, AND L. K. MEDLIN. 2003. Pseudo-cryptic speciation in coccolithophores. *Proc. Natl. Acad. Sci. USA* **100**: 7163–7168.
- SARTHOU, G., K. R. TIMMERMANS, S. BLAIN, AND P. TRÉGUER. 2005. Growth physiology and fate of diatoms in the ocean: A review. *J. Sea Res.* **53**: 25–42.
- SCHOEMANN, V., S. BECQUEVORT, J. STEFELS, V. ROUSSEAU, AND C. LANCELOT. 2005. *Phaeocystis* blooms in the global ocean and their controlling mechanisms: A review. *J. Sea Res.* **53**: 43–66.
- SCHOUTEN, S., J. OSSEBAAR, K. SCHREIBER, M. V. M. KIENHUIS, G. LANGER, A. BENTHIE, AND J. BIJMA. 2006. The effect of temperature, salinity and growth rate on the stable hydrogen isotopic composition of long chain alkenones produced by *Emiliania huxleyi* and *Gephyrocapsa oceanica*. *Biogeosciences* **3**: 113–119.
- YOUNG, J. R., AND P. ZIVERI. 2000. Calculation of coccolith volume and its use in calibration of carbonate flux estimates. *Deep-Sea Res. II* **47**: 1679–1700.
- ZIVERI, P., K.-H. BAUMANN, B. BÖCKEL, J. BOLLMANN, AND J. R. YOUNG. 2004. Biogeography of selected Holocene coccoliths in the Atlantic Ocean, p. 402–428. *In* H. R. Thierstein and J. R. Young [eds.], *Coccolithophores, from molecular processes to global impact*. Springer.

Received: 30 April 2007

Accepted: 19 November 2007

Amended: 10 December 2007