



**HAL**  
open science

## A coccolithophore concept for constraining the Cenozoic carbon cycle

J. Henderiks, R. E. M. Rickaby

► **To cite this version:**

J. Henderiks, R. E. M. Rickaby. A coccolithophore concept for constraining the Cenozoic carbon cycle. *Biogeosciences*, 2007, 4 (3), pp.323-329. hal-00297616

**HAL Id: hal-00297616**

**<https://hal.science/hal-00297616>**

Submitted on 18 Jun 2008

**HAL** is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers.

L'archive ouverte pluridisciplinaire **HAL**, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d'enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.

# A coccolithophore concept for constraining the Cenozoic carbon cycle

J. Henderiks<sup>1</sup> and R. E. M. Rickaby<sup>2</sup>

<sup>1</sup>Stockholm University, Department of Geology and Geochemistry, Svante Arrhenius väg 8C, SE-106 91 Stockholm, Sweden

<sup>2</sup>Department of Earth Sciences, Oxford University, Parks Road, Oxford OX1 3PR, UK

Received: 7 December 2006 – Published in Biogeosciences Discuss.: 16 January 2007

Revised: 23 May 2007 – Accepted: 6 June 2007 – Published: 19 June 2007

**Abstract.** An urgent question for future climate, in light of increased burning of fossil fuels, is the temperature sensitivity of the climate system to atmospheric carbon dioxide (pCO<sub>2</sub>). To date, no direct proxy for past levels of pCO<sub>2</sub> exists beyond the reach of the polar ice core records. We propose a new methodology for placing a constraint on pCO<sub>2</sub> over the Cenozoic based on the physiological plasticity of extant coccolithophores. Specifically, our premise is that the contrasting calcification tolerance<sup>1</sup> of various extant species of coccolithophore to raised pCO<sub>2</sub> reflects an “evolutionary memory” of past atmospheric composition. The different times of evolution of certain morphospecies allows an upper constraint of past pCO<sub>2</sub> to be placed on Cenozoic timeslices. Further, our hypothesis has implications for the response of marine calcifiers to ocean acidification. Geologically “ancient” species, which have survived large changes in ocean chemistry, are likely more resilient to predicted acidification.

## 1 Introduction

Anthropogenic pCO<sub>2</sub> is absorbed largely by the oceans, causing acidification of the biologically productive surface waters (The Royal Society, 2005; Kleypas et al., 2006). Corals, foraminifera and pteropods decrease their calcification in response to increased acidification (Gattuso et al., 1999; Kleypas et al., 1999; Bijma et al., 1999; Orr et al., 2005). Despite the intracellular nature of coccolithophore calcification, culture experiments confirm that *Emiliana huxleyi* and its close relative *Gephyrocapsa oceanica* also decrease calcification as pCO<sub>2</sub> increases (Riebesell et al., 2000;

Correspondence to: J. Henderiks  
(jorijntje.henderiks@geo.su.se)

<sup>1</sup>We define “calcification tolerance” as the tolerance to variable pCO<sub>2</sub> of coccolithophores with respect to calcify normally (with no malformation/incomplete coccolithogenesis).

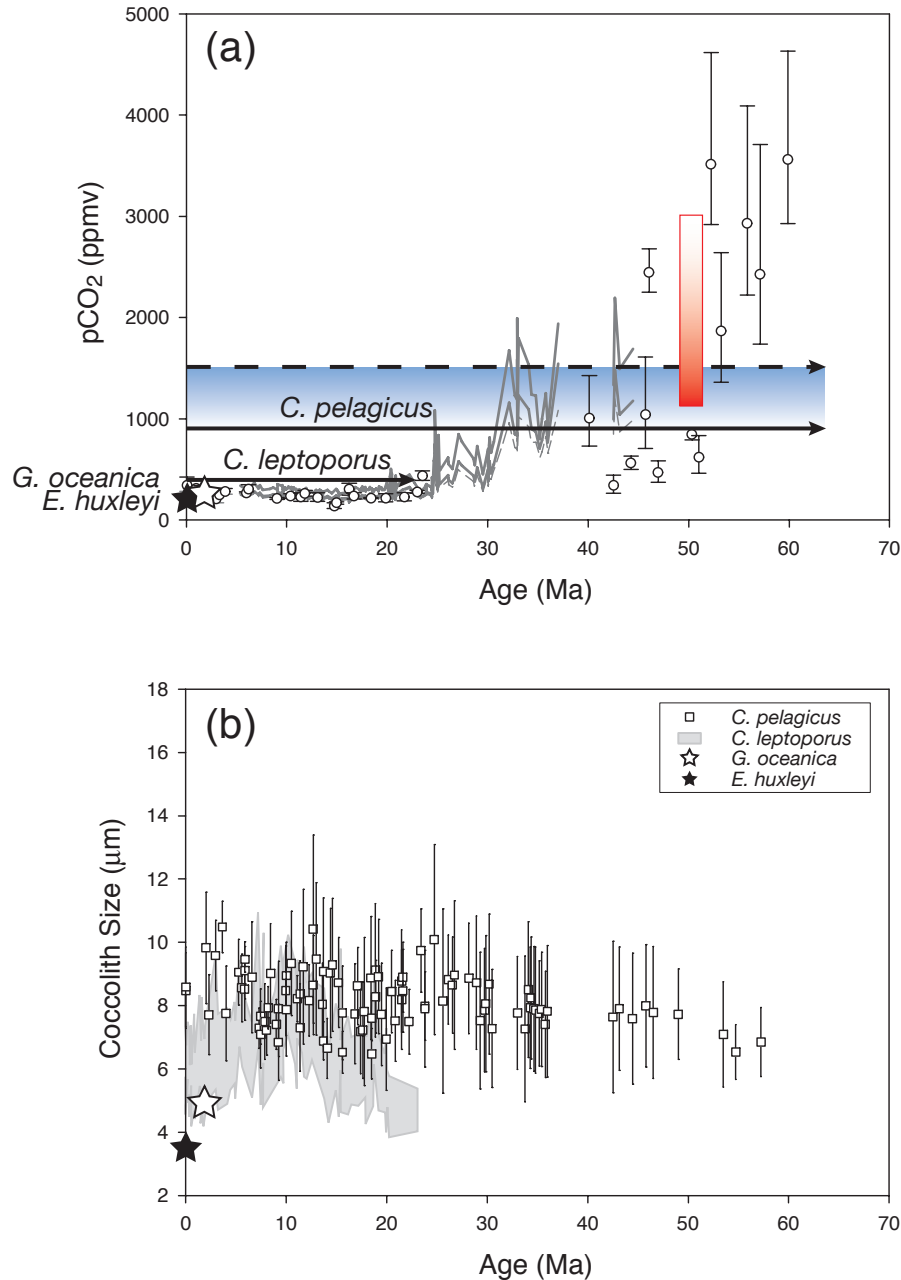
Zondervan et al., 2001). Decreased calcification is a negative feedback to acidification on the short term (Zondervan et al., 2001) but poses a serious threat to the sensitive ecologic balance. Ultimately, the rate at which populations acclimatise or adapt to changing ocean chemistry will determine the operation of feedback mechanisms on future climate.

Long-term patterns in coccolithophorid evolution and success seem, however, counterintuitive to this paradigm of low calcification under high pCO<sub>2</sub>. Coccolithophore diversity and abundance peaked during the greenhouse Cretaceous world and have since declined (Bown, 2005). In addition, and by contrast to the tests on *E. huxleyi* and *G. oceanica* (Riebesell et al., 2000; Zondervan et al., 2001), recent acidification experiments testing the environmental tolerance of different extant coccolithophore species reveal that the calcification response is species-specific (Langer et al., 2006). Calcification ability<sup>2</sup> in the species *Coccolithus pelagicus* is unaffected by values of pCO<sub>2</sub> up to at least 920 ppmV, and even though calcification ability declines, coccoliths are still produced up to much higher levels (S. Krug, personal communication). By contrast, *Calcidiscus leptoporus* shows an “optimum” calcification ability close to modern day pCO<sub>2</sub> (Langer et al., 2006). Thus, species-specific effects must be considered when evaluating whole ecosystem response to elevated pCO<sub>2</sub>. Although representative of few algal strains, these culture results raise a major question: why do different species respond differently to pCO<sub>2</sub>?

## 2 Methodology and hypothesis

With a geological perspective, the ability of different coccolithophore species to calcify in culture under a range of ocean acidity correlates with likely levels of atmospheric

<sup>2</sup>We define “calcification ability” as the ability to produce coccoliths with no, or only minor, decrease in PIC/POC ratio from those during optimum conditions.



**Fig. 1.** (a) At face value, the levels of pCO<sub>2</sub> tolerated by different coccolithophore species for calcification in culture experiments (Riebesell et al., 2000; Langer et al., 2006) coincide with levels of pCO<sub>2</sub> from marine proxy reconstructions, based on alkenones (solid and dashed lines; Pagani et al., 2005) and boron isotopes in planktic foraminifera (open circles; Pearson and Palmer, 2000), at the time of emergence of each respective morphospecies; marked by a star for *E. huxleyi* (291–268 ka; closed star) and *G. oceanica* (1.7 Ma; open star), and solid arrows for *C. leptoporus* (23 Ma) and *C. pelagicus* (~63 Ma). The upper pCO<sub>2</sub> tolerance range for *C. pelagicus* is indicated by the blue box with a solid arrow at the maximum pCO<sub>2</sub>, thus far tested, at which its calcification remains unaffected (Langer et al., 2006), and dashed arrow showing the potential for this species, with further experiments, to determine the Palaeogene maximum pCO<sub>2</sub>. The red box depicts the pCO<sub>2</sub> range in which the sodium carbonate nahcolite precipitates (Lowenstein and Demicco, 2006), fixing minimum early Eocene pCO<sub>2</sub> at 1125 ppmV. (b) Mean coccolith size, a proxy for coccolithophore cell size, has remained largely constant for *Coccolithus* during the Cenozoic decline of pCO<sub>2</sub> (open squares ±1 stdev; this study), and for *Calcidiscus* (grey envelope ±1 stdev; Knappertsbusch, 2000) since the Miocene. Both genera are significantly larger than *E. huxleyi* (closed star), which stands out as one of the smallest, but most prolific calcifying coccolithophore alive today and represents the youngest evolutionary addition (291–268 ka) to the fossil record of calcifying haptophytes.

pCO<sub>2</sub> since the emergence of their corresponding morpho-species during the Cenozoic (Fig. 1a). Modern *E. huxleyi* and *G. oceanica* are distinct genetic outliers to most other extant coccolithophore (Fujiwara et al., 1994; Sáez et al., 2003), and group together with other alkenone-producing haptophytes such as the non-coccolith bearing species *Isochrysis galbana*. The morphospecies *E. huxleyi* had a first occurrence ~291–268 ka and has risen to global dominance since 85 ka, while *G. oceanica* evolved at 1.7 Ma (Thierstein et al., 1977; Raffi et al., 2006). In contrast, the morphospecies *Calcidiscus leptoporus* originated at 23 Ma (Knappertsbusch, 2000) and *Coccolithus pelagicus* in the Palaeocene, ~63 Ma (Haq and Lohmann, 1976). At face value, the levels of pCO<sub>2</sub> tolerated in cultures of the different coccolithophorid morphospecies coincide with the levels of pCO<sub>2</sub> from proxy reconstructions at the time of their diversification (Pagani et al., 2005; Pearson and Palmer, 2000). This evolutionary view of the culture results holds the potential to place an upper constraint on Cenozoic palaeo-pCO<sub>2</sub>, which must have been at least 920 ppmV at the time of evolution of *C. pelagicus*, 63 Ma. With further experiments to push the calcification tolerance of *C. pelagicus* and additional modern representatives of ancient morphospecies, we predict a tight bracketing of palaeo-pCO<sub>2</sub> estimates for the Palaeogene, especially in combination with a fixed minimum early Eocene pCO<sub>2</sub> of 1125 ppmV, as reconstructed by stability fields of sodium carbonates (Fig. 1a; Lowenstein and Demicco, 2006). Our proposed proxy appears to concur with an emerging view of different climate sensitivities before and after the marked draw-down of pCO<sub>2</sub> at the early Oligocene from the greenhouse of the early Cenozoic (Pagani et al., 2005; Lowenstein and Demicco, 2006).

### 3 Discussion

Our statement that genetically “older” extant species have an “evolutionary memory” of calcification under conditions of higher pCO<sub>2</sub> in these culture experiments, and that this envelope of ability may constrain past pCO<sub>2</sub> estimates, requires detailed consideration. Nonetheless, other evidence suggests that ancient biomineralisation pathways may be recalled by extant species grown in conditions which reflect ocean chemistry of the past. For example, relict modern populations of sublittoral siliceous demosponges (genus *Crambe*), which have Jurassic ancestry, modulate the phenotypic expression of their spicules depending on the experimental concentrations of silicic acid (Maldonado et al., 1999). These organisms only produce lightly silicified spicules in their natural environment, which is likely silicon limited. Under “ancient” chemical conditions of elevated silica concentrations, *C. crambe* proves genetically capable of producing spicule types that are not normally found in natural populations, but have high resemblance to fossil morphotypes (Maldonado et al., 1999). In some organisms or species, the genes that encode biomineralisation may be conservative over time but

their genetic expression is sensitive to the environment such that biomineralisation is phenotypically plastic.

Calcification has most likely evolved only once in haptophytes, in the late Triassic. The presence of non-calcifying taxa within the typical “coccolithophore” phylogenetic clades suggests that the biochemical pathways involved in coccolithogenesis have been shut down and possibly reactivated several times in the course of their evolution (deVargas and Probert, 2004). Still, a relatively simple phylogenetic pattern can be derived based on life-cycle observations and related modes of biomineralisation (Young and Bown, 1997). For example, *C. pelagicus* and *C. leptoporus* produce both heterococcoliths (in diploid phase) and holococcoliths (haploid phase), while *E. huxleyi* and *G. oceanica* appear to have lost the ability to produce holococcoliths (they only produce organic scales in haploid phase). The close phylogenetic affinity of *E. huxleyi* and *G. oceanica* to the non-calcifying haptophyte genus *Isochrysis* (Sáez et al., 2004) also suggests that the genetic encoding for calcification is subject to variable evolutionary selection within this organism group, and is susceptible to phenotypic plasticity.

For our proposed evolutionary perspective of constraints on atmospheric pCO<sub>2</sub>, the photosynthetic or calcification potential of a species must be optimised for an aspect of ocean carbonate chemistry related to pCO<sub>2</sub> at the time of origin and, become preserved within the genetic make-up since the ocean and atmosphere chemistry changed. Although debate persists regarding the exact interplay between calcification and photosynthesis, calcification requires energy, and in coccolithophores, the only source of this energy can be photosynthesis. Therefore the two processes must be coupled to some degree due to the energy requirement for the transport of Ca<sup>2+</sup> and CO<sub>3</sub><sup>2-</sup> into the coccolithophore cells, and control of intracellular pH. In the culture experiments, as in the modern ocean, the carbonate saturation state and pH are inversely proportional to pCO<sub>2</sub>, with coherence between lower energetic requirement for calcification at high carbonate saturation and pH, and lower photosynthetic energy available from low pCO<sub>2</sub>. In the geological past, carbonate saturation state and pCO<sub>2</sub> were likely decoupled (Tyrrell and Zeebe, 2004). Nevertheless, available long-term reconstructions of ocean pH consistently depict a secular increase in pH during the Cenozoic (Pearson and Palmer, 2000; Tyrrell and Zeebe, 2004). Without further culture experiments of decoupled carbonate system parameters, it is impossible to resolve which aspect of ocean chemistry, and which physiological pathway is fundamental to coccolithogenesis. Because of the intrinsic coupling between photosynthesis and calcification, and between ocean pH and pCO<sub>2</sub>, we shall relate the following discussion of which extant coccolithophore species possess an “evolutionary memory” and how such physiological pathways could be preserved, to pCO<sub>2</sub> as the variant parameter.

According to the culture experiments, the different species of coccolithophore fall into two groups: conservative species which have retained the genetic encoding

for biomineralisation at past levels of  $p\text{CO}_2$ , and adaptive species. In the marine realm, it would be expected that populations of planktonic micro-organisms could quickly adapt to their changing environment (e.g. deVargas et al., 2004), such that all extant coccolithophores are adapted to present-day values of  $p\text{CO}_2$ . As discussed by Langer et al. (2006), the bell-shaped calcification response by *C. leptoporus*, centred around present-day values of  $p\text{CO}_2$ , is consistent with such adaptation to variable  $p\text{CO}_2$  on glacial-interglacial time scales and therefore this species is unsuitable to use as a palaeo-proxy for Miocene  $p\text{CO}_2$ .

By contrast, the broad, and apparently insensitive response by *C. pelagicus* to variable levels of  $p\text{CO}_2$  in culture also suggests adaptation to present-day  $p\text{CO}_2$  values, but with conservation of the ability to calcify normally under lower, as well as much higher  $\text{CO}_2$  concentrations, and therefore potentially represents an “evolutionary memory” of both the glacial low- $p\text{CO}_2$  and high- $p\text{CO}_2$  Palaeogene worlds. *Coccolithus pelagicus* tolerates conditions (S. Krug, personal communication) beyond the limit of modern surface waters (Orr et al., 2005), which indicates that this species has retained a signature of adaptation to a different past  $p\text{CO}_2$ . In comparison to other tested coccolithophores, *C. pelagicus* appears conservative in its evolutionary adaptation. This supposition is supported by our coccolith size data<sup>3</sup> for the *Coccolithus* morphospecies lineage. The morphological template and the mean coccolith size, linearly related to cell size (Henderiks and Pagani, in press), have remained largely constant for *Coccolithus* during the Cenozoic decline of  $p\text{CO}_2$  (Fig. 1b). In contrast, the Cenozoic ancestors of *Gephyrocapsa* and *Emiliana*, within the *Reticulofenestra* genus, have greatly varied in coccolith size, with largest morphospecies (14–20  $\mu\text{m}$ ) occurring during the late Eocene (Backman and Hermelin, 1986). Available morphometric data of *Calcidiscus* morphospecies show a secular decline in mean size since the late Miocene (in the equatorial and southern Indian Ocean; Knappertsbusch, 2000). Based on these data, we argue that such distinct evolutionary size patterns between the coccolithophore lineages imply that the different morphospecies may fall into two groups: morphospecies which are highly adaptive to changing ocean chemistry in terms of their calcification and size, and morphospecies which are highly conservative. Indeed, our fossil morphospecies likely group several cryptic species together (Saéz et al., 2003; deVargas et al., 2004). The coccolith size range of such cryptic species is in fact rather constant, whereas their biogeography and seasonality is highly variable (e.g. Renaud et al., 2002). In

this light, the total size range of fossil morphospecies could potentially be a measure for the degree of (cryptic) speciation within each coccolithophore lineage, and thus reflect evolutionary rates and/or genetic plasticity of coccolithogenesis.

Adaptation to a new ecological niche may result in a “cost” in terms of lost ancestral capabilities (e.g. Bell, 1997; Kassen, 2002) such that the adaptive *E. huxleyi* has lost the ability to succeed at high  $p\text{CO}_2$ . These capabilities may be lost either because they are no longer under selection (neutral) or because of a deleterious effect on fitness in a new niche. Evolution of the lightly calcified *Gephyrocapsa* and *E. huxleyi* with fast growing, small cells provides the advantage of investing energy in cell growth, and an ability to outcompete more heavily calcified coccolithophores in a low  $p\text{CO}_2$  world. Indeed the macroevolutionary decrease in coccolith (and thus cell) size during the Neogene (Henderiks et al., 2004) could indicate adaptation of the photosynthetic capacity of small reticulofenestrads and *Calcidiscus* morphospecies to lowered  $p\text{CO}_2$ . Cells optimise their photosynthesis by increasing surface area to volume, maximizing light acquisition, increasing Rubisco specificity (Tortell, 2000), and carbon concentrating mechanisms (Giordano et al., 2005). Today, *C. pelagicus* represents one of the largest coccolithophores alive, whereas *E. huxleyi* stands out as one of the smallest and appears best adapted to low  $p\text{CO}_2$  values, because this species shows no photoinhibition at high light levels or in continuous light (e.g. Nanninga and Tyrrell, 1996; Zondervan et al., 2002), blooms in the uppermost water column, has a carbon concentrating mechanism (Rost et al., 2002) and is likely an adaptive low- $p\text{CO}_2$  specialist. Additionally, *E. huxleyi* and *G. oceanica* cells are produced with an average PIC/POC ratio of 0.6–0.8 compared to 1.6–1.8 for both *C. leptoporus* and *C. pelagicus* (Zondervan et al., 2001, 2002; Langer et al., 2006) which could imply a different partitioning priority for energy between photosynthetic carbon fixation and calcification as a result of the  $p\text{CO}_2$  at their time of origin. This would imply that both *C. leptoporus* and *C. pelagicus* precipitate more  $\text{CaCO}_3$  per mole of C fixed and are efficient calcifiers. By contrast *E. huxleyi* prioritises photosynthetic carbon fixation under low  $p\text{CO}_2$  conditions and the cost of calcification to *E. huxleyi* and *G. oceanica* is proportionately larger and expendable as the ocean becomes more acidic.

Conservation of an “evolutionary memory”, and phenotypic plasticity may be manifest as generalist or specialist behaviour of the different coccolithophore species with respect to  $p\text{CO}_2$  (Richmond et al., 2005). The preservation of *C. pelagicus*’ calcification ability may be enhanced because this species prefers a high  $p\text{CO}_2$  niche. *C. pelagicus* prevailed in Palaeocene equatorial regions (Haq and Lohmann, 1977), and was only restricted to high  $p\text{CO}_2$  subpolar and upwelling niches since the late Pliocene (Sato et al., 2004). By contrast *E. huxleyi* appears photosynthetically optimised for success in the modern ecological niche of low  $p\text{CO}_2$ . The distribution of the apparently cosmopolitan *E. huxleyi*, and to

<sup>3</sup>Coccolith morphometric data on 99 deep-sea sediment samples, from various (sub)tropical and temperate sites, ranging in age 2.2 ka–57.3 Ma. Mean coccolith size was determined from 100–200 individual coccolith measurements per sample collected from 4 replicate sprayed slides under polarised light microscopy (Henderiks and Törner, 2006). Further details will be presented and discussed elsewhere.

a lesser extent *C. leptoporus*, correlates well with the consistently low  $[\text{CO}_2(\text{aq})]$  across the tropical and temperate latitudes (Orr et al., 2005) such that the broad biogeographic distribution actually reflects a narrow ecological niche in terms of  $[\text{CO}_2(\text{aq})]$ .

The broad tolerance to  $[\text{CO}_2(\text{aq})]$  of *C. pelagicus* is likely a manifestation of the physiological plasticity, maintained at extra energetic cost, of this species and implies conservative evolution as reflected by the relative constancy of cell size and coccolith morphology throughout the last 63 million years (Fig. 1b). The optimisation of *E. huxleyi* to bloom and conquer the majority of the world's oceans has occurred at a cost incurring the loss of competitiveness at high  $\text{pCO}_2$ . A high degree of selectivity of the photosynthetic machinery for low  $\text{pCO}_2$  can be coupled with slow photosynthetic kinetics (Tcherkez et al., 2006) such that in high  $\text{pCO}_2$  conditions, the growth rate of *E. huxleyi* shows little response.

In terms of underlying genetic mechanisms, currently, little is known about genetic controls on calcification (e.g. Marsh, 2003; Nguyen et al., 2005), or the detailed photosynthetic mechanism of coccolithophores. Coupling of calcification with species-specific Rubisco specificity provides a tangible means to preserve the  $\text{CO}_2/\text{O}_2$  composition at the time of origin of photosynthetic phyla (Giordano et al., 2005; Tcherkez et al., 2006). The preservation of calcification ability at high  $\text{pCO}_2$  in *C. pelagicus* may occur through genetic redundancy (Wagner, 1999), or variance in genetic expression whilst the adaptation of *E. huxleyi* and *C. leptoporus* to the modern low  $\text{pCO}_2$  niche could be associated with gene inactivation of pathways associated with high  $\text{pCO}_2$  (Hittinger et al., 2004). The high proportion of duplicate genes within plant and algae genomes is indicative of a high rate of retention of duplicate genes (Lynch and Connery, 2000). Gene duplications contribute to the establishment of new gene functions, and may underlie the origin of evolutionary novelty. Duplicate genes can exist stably in a partially redundant state over a protracted evolutionary period (Moore and Purugganan, 2005). A half-life to silencing and loss of a plant gene duplicate is estimated at 23.4 million years such that remnant duplicate genes, which can be reactivated by environmental conditions to encode calcification within coccolithophores under "ancestral" conditions representative of ~60 Ma, appears reasonable.

#### 4 Concluding remarks

Our hypothesis for a novel proxy requires further foundation with experiments on an assortment of ancient morphospecies and strains, and investigation into calcification and photosynthesis under a range of ocean carbonate conditions, nutrient levels and temperatures. Nonetheless, our ideas have implications for the future ocean. With fossil fuel burning and a predicted decrease in pH of ~0.3 over the next 100 years (The Royal Society, 2005), the larger species will likely have

an advantage over the now prosperous *E. huxleyi*, as the carbonate system of the ocean reverses towards the acidity of the past. *C. pelagicus* has weathered large and abrupt changes in conditions in the geological past, e.g. the Palaeocene-Eocene thermal maximum (Gibbs et al., 2006), with no apparent impact on physiology, but the adaptive strategies of newcomer *E. huxleyi* may differ significantly, potentially leading to future non-calcifying descendants.

*Acknowledgements.* We are grateful to S. Krug for sharing his unpublished data with us and informing our hypothesis. We would like to thank M. Lachmann, P. Westbroek, F. Morel and P. Falkowski for their help with discussion and development of the ideas presented here. Interdisciplinary discussions with participants of the IGBP-SCOR workshop on ocean acidification (Lamont-Doherty Earth Observatory, September 2006) further strengthened the potential impacts of our hypothesis. We wish to thank three anonymous referees as well as U. Riebesell for their constructive and challenging comments.

Edited by: J. Bijma

#### Author contributions

The authors contributed equally to the preparation of this manuscript.

#### References

- Backman, J. and Hermelin, J. O. R.: Morphometry of the Eocene nannofossil *Reticulofenestra umbilicus* lineage and its biochronological consequences, *Palaeogeogr., Palaeoclim., Palaeoecol.*, 57, 103–116, 1986.
- Bell, G.: The basics of selection, Chapman and Hall, New York, 1997.
- Bijma, J., Spero, H. J., and Lea, D. W.: Reassessing foraminiferal stable isotope geochemistry: Impact of the oceanic carbonate system (experimental results), in: Use of Proxies in Paleoclimatology: Examples from the South Atlantic, edited by: Fischer, G. and Wefer, G., Springer-Verlag, Berlin Heidelberg, 489–512, 1999.
- Bown, P.: Calcareous nannoplankton evolution: a tale of two oceans, *Micropaleontology*, 51, 299–308, 2005.
- de Vargas, C. and Probert, I.: New keys to the Past: Current and future DNA studies in Coccolithophores, *Micropaleontology*, 50, 45–54, 2004.
- de Vargas, C., Sáez, A., Medlin, L., and Thierstein, H. R.: Super-Species in the calcareous plankton, in: Coccolithophores – From molecular processes to global impact, edited by: Thierstein, H. R. and Young, J., 271–298, 2004.
- Fujiwara, S., Sawada, M., and Someya, J.: Molecular phylogenetic analysis of *rbcL* in the Prymnesiophyta, *J. Phycology*, 30, 863–871, 1994.
- Gattuso, J.-P., Allemand, D., and Frankignoulle, M.: Photosynthesis and calcification at cellular, organismal and community levels in coral reefs: a review on interactions and control by carbonate chemistry, *Am. Zoologist*, 39, 160–183, 1999.

- Gibbs, S., Bralower, T. J., Bown, P. R., Zachos, J. C., and Bybell, L. M.: Shelf and open-ocean calcareous phytoplankton assemblages across the Paleocene-Eocene Thermal Maximum: Implications for global productivity gradients, *Geology*, 34, 233–236, 2006.
- Giordano, M., Beardall, J., and Raven, J. A.: CO<sub>2</sub> concentrating mechanisms in algae: Mechanisms, environmental modulation, and evolution, *Annu. Rev. Plant Biol.*, 56, 99–131, 2005.
- Haq, B. U. and Lohmann, G. P.: Early Cenozoic calcareous nannoplankton biogeography of the Atlantic Ocean, *Mar. Micropaleontol.*, 1, 119–194, 1976.
- Henderiks, J. and Pagani, M.: Refining ancient carbon dioxide estimates: Significance of coccolithophore cell size for alkenone-based pCO<sub>2</sub> records, *Paleoceanography*, doi:10.1029/2006PA001399, in press, 2007.
- Henderiks, J. and Törner, A.: Reproducibility of coccolith morphometry: Evaluation of spraying and smear slide preparation techniques, *Mar. Micropaleontol.*, 58, 207–218, 2006.
- Henderiks, J., Lindberg, L., and Törner, A.: Neogene patterns of coccolith size evolution and carbonate burial in the deep sea, *J. Nannoplankton Res.*, 26, 55–56, 2004.
- Hittinger, C. T., Rokas, A., Carroll S. B.: Parallel inactivation of multiple *GAL* pathway genes and ecological diversification in yeasts, *Proc. Nat. Acad. Sci.*, 101, 14 144–14 149, 2004.
- Kassen, R.: The experimental evolution of specialists, generalists, and the maintenance of diversity, *J. Evol. Biol.*, 15, 173–190, 2002.
- Kleypas, J. A., Buddemeier, R.W., Archer, D., Gattuso, J.-P., Langdon, C., and Opdyke, B. N.: Geochemical consequences of increased atmospheric carbon dioxide on coral reefs, *Science*, 284, 118–120, 1999.
- Kleypas, J. A., Feely, R. A., Fabry, V. J., Langdon, C., Sabine, C. L., and Robbins, L. L.: Impacts of Ocean Acidification on Coral Reefs and Other Marine Calcifiers: A Guide for Future Research, sponsored by NSF, NOAA, and the U.S. Geological Survey, St. Petersburg, USA, 88 pp., 2006.
- Knappertsbusch, M.: Morphologic evolution of the coccolithophorid *Calcidiscus leptoporus* from the Early Miocene to Recent, *J. Paleontol.*, 74, 712–730, 2000.
- Langer, G., Geisen, M., Baumann, K.-H., Kläs, J., Riebesell, U., Thoms, S., and Young, J. R.: Species specific responses of calcifying algae to changing seawater carbonate chemistry, *Geochem., Geophys., Geosyst.*, 7, Q09006, doi:10.1029/2005001227, 2006.
- Lowenstein, T. K. and Demicco, R. V.: Elevated Eocene atmospheric CO<sub>2</sub> and its subsequent decline, *Science*, 313, 1928, 2006.
- Lynch, M. and Conery, J. S.: The evolutionary fate and consequences of duplicate genes, *Science*, 290, 1151–1155, 2000.
- Maldonado, M., Carmona, M. C., Uriz, M. J., and Cruzado, A.: Decline in Mesozoic reef-building sponges explained by silicon limitation, *Nature*, 401, 785–788, 1999.
- Marsh, M. E.: Regulation of CaCO<sub>3</sub> formation in coccolithophores, *Comparative Biochemistry and Physiology Part B*, 136, 743–754, 2003.
- Moore, R. C. and Purugganan, M. D.: The evolutionary dynamics of plant duplicate genes, *Curr. Opin. Plant Biol.*, 8, 122–128, 2005.
- Nanninga, H. J. and Tyrrell, T.: The importance of light for the formation of algal blooms by *Emiliania huxleyi*, *Mar. Ecol. Progress Ser.*, 136, 195–203, 1996.
- Nguyen, B., Bowers, R. M., Wahlund, T. M., and Read, B. A.: Suppressive subtractive hybridization of and differences in gene expression content of calcifying and noncalcifying cultures of *Emiliania huxleyi* strain 1516, *Appl. Environ. Microbiol.*, 71, 2564–2575, 2005.
- Orr, J. C., Fabry, V. J., Aumont, O., Bopp, L., Doney, S. C., Feely, R. A., Gnanadesikan, A., Gruber, N., Ishida, A., Joos, F., Key, R. M., Lindsay, K., Maier-Reimer, E., Matear, R., Monfray, P., Mouchet, A., Najjar, R. G., Plattner, G.-K., Rodgers, K. B., Sabine, C. L., Sarmiento, J. L., Schlitzer, R., Slater, R. D., Totterdell, I. J., Weirig, M.-F., Yamanaka, Y., and Yool, A.: Anthropogenic ocean acidification over the twenty-first century and its impact on calcifying organisms, *Nature*, 437, 681–686, 2005.
- Pagani, M., Zachos, J. C., Freeman, K. H., Tripple, B., and Bohaty, S.: Marked decline in atmospheric carbon dioxide concentrations during the Paleogene, *Science*, 309, 600–603, 2005.
- Pearson, P. N. and Palmer, M. R.: Atmospheric carbon dioxide concentrations over the past 60 million years, *Nature*, 406, 695–699, 2000.
- Raffi, I., Backman, J., Fornaciari, E., Pälke, H., Rio, D., Lourens, L., and Hilgen, F.: A review of calcareous nannofossil astrochronology encompassing the past 25 million years, *Quaternary Sci. Rev.*, 25, 3113–3137, 2006.
- Renaud, S., Ziveri, P., and Broerse, A. T. C.: Geographical and seasonal differences in morphology and dynamics of the coccolithophore *Calcidiscus leptoporus*, *Mar. Micropaleontol.*, 46, 363–385, 2002.
- Richmond, C. E., Breitburg, D. L., and Rose, K. A.: The role of environmental generalist species in ecosystem function, *Ecol. Model.*, 188, 279–295, 2005.
- Riebesell, U., Zondervan, I., Rost, B., Tortell, P. D., Zeebe, R. E., and Morel, F. M. M.: Reduced calcification of marine plankton in response to increased atmospheric CO<sub>2</sub>, *Nature*, 407, 364–367, 2000.
- Rost, B., Zondervan, I., and Riebesell, U.: Light-dependent carbon isotope fractionation in the coccolithophorid *Emiliania huxleyi*, *Limn. Oceanogr.*, 47, 120–128, 2002.
- Sáez, A. G., Probert, I., Geisen, M., Quinn, P., Young, J., and Medlin, L. K.: Pseudo-cryptic speciation in coccolithophores, *Proc. Nat. Acad. Sci.*, 100, 7163–7168, 2003.
- Sáez, A. G., Probert, I., Young, J., Edvardsen, B., Eikrem, W., and Medlin, L. K.: A review of the phylogeny of the Haptophyta, in: *Coccolithophores – From molecular processes to global impact*, edited by: Thierstein, H. R. and Young, J., 251–269, 2004.
- Sato, T., Yuguchi, S., Takayama, T., and Kameo, K.: Drastic change in the geographical distribution of the cold-water nannofossil *Coccolithus pelagicus* (Wallich) Schiller at 2.74 Ma in the late Pliocene, with special reference to glaciation in the Arctic Ocean, *Mar. Micropaleontol.*, 52, 181–193, 2004.
- Tcherkez, G. G. B., Farquhar, G. D., and Andrews, T. J.: Despite slow catalysis and confused substrate specificity, all ribulose biphosphate carboxylases may be nearly perfectly optimized, *Proc. Nat. Acad. Sci.*, 103, 7246–7251, 2006.
- The Royal Society: Ocean acidification due to increasing atmospheric carbon dioxide, Policy Document 12/05, 60 pp., 2005.
- Thierstein, H. R., Geitzenauer, K. R., Molfino, B., and Shackleton, N. J.: Global synchronicity of late Quaternary coccolith datum levels: Validation by oxygen isotopes, *Geology*, 5, 400–404,

- 1977.
- Tortell, P. D.: Evolutionary and ecological perspectives on carbon acquisition in phytoplankton, *Limnol. Oceanogr.*, 45, 744–750, 2000.
- Tyrrell, T. and Zeebe, R. E.: History of carbonate ion concentration over the last 100 million years, *Geochim. Cosmochim. Acta*, 68, 3521–3530, 2004.
- Wagner, A.: Redundant gene functions and natural selection, *J. Evolution. Biol.*, 12, 1–16, 1999.
- Young, J. R. and Bown, P. R.: Higher classification of calcareous nannofossils, *J. Nannoplankton Res.*, 19, 15–20, 1997.
- Zondervan, I., Rost, B., and Riebesell, U.: Effect of CO<sub>2</sub> concentration on the PIC/POC ratio in the coccolithophore *Emiliana huxleyi* grown under light-limiting conditions and different daylengths, *J. Exp. Mar. Biol. Ecol.*, 272, 55–70, 2002.
- Zondervan, I., Zeebe, R. E., Rost, B., and Riebesell, U.: Decreasing marine biogenic calcification: A negative feedback on rising atmospheric pCO<sub>2</sub>, *Global Biogeochem. Cy.*, 15, doi:10.1029/2000GB001321, 507–516, 2001.