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LETTER

Particulate inorganic to organic carbon production as a predictor for coccolithophorid sensitivity to ongoing ocean acidification

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Scientific Significance Statement

Coccolithophores, calcifying phytoplankton, are a vital driver of the marine carbon cycle and mostly negatively influenced by ongoing ocean acidification. Yet, the reason for observed species and strain-specific sensitivities are elusive. Here, we identify the ratio of cellular calcification to photosynthesis as a predictor for individual sensitivity, allowing us to group species and strains accordingly.

Abstract

Ocean acidification (OA) can induce shifts in plankton community composition, with coccolithophores being mostly negatively impacted. This is likely to change particulate inorganic and organic carbon (PIC and POC, respectively) production, with impacts on the biological carbon pump. Hence, assessing and, most importantly, understanding species-specific sensitivities of coccolithophores is paramount. In a multispecies comparison, spanning more than two orders of magnitude in terms of POC and PIC production rates, among *Calcidiscus leptoporus*, *Coccolithus pelagicus subsp. braarudii*, *Emiliania huxleyi*, *Gephyrocapsa oceanica*, and *Scyphosphaera apsteinii*, we found that cellular PIC : POC was a good predictor for a species' OA sensitivity. This is likely related to the need for cellular pH homeostasis, which is challenged by the process of calcification producing protons internally, especially when seawater pH decreases in an OA scenario. With higher PIC : POC, species and strains being more sensitive to OA coccolithophores may shift toward less calcified varieties in the future.

Coccolithophores are an abundant component of marine phytoplankton assemblages, which can be found in almost all ocean ecosystems ranging from the subpolar regions to the equator (McIntyre and Bé 1967). Coccolithophores produce both particulate inorganic carbon (PIC) through calcification and particulate organic carbon (POC) through photosynthesis (Rost and Riebesell 2004). The amount of carbon produced by coccolithophores varies greatly between species, with, for instance, *Emiliania huxleyi* producing less than 6 pg C cell^{-1} and large species like *Scyphosphaera apsteinii* producing more than 200 times as much (Table 1). The ratio of inorganic to organic carbon production also varies greatly between and within species with ratios as low as 0.08 in *Chrysotila carterae* and as high as 3.29 in *Calcidiscus leptoporus* subsp. *quadriperforatus* (Table 1).

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Additional Supporting Information may be found in the online version of this article.

Table 1. Range and mean (bold) values of carbon production quotas consisting of total particulate carbon, PIC, and POC (pg C cell^{-1}), PIC : POC ratios, and cell diameter (μm), volume (μm^3), and surface area : volume ratio (SA : Vol) for *C. leptoporus* including subspecies *quadriperforatus*, *C. pelagicus* subsp. *braarudii*, *E. huxleyi*, *G. oceanica*, *S. apsteinii*, *H. carteri*, *C. carterae*, *S. pulchra*, and *U. sibogae*.

Species	Total	Carbon quota inorganic	Organic	PIC : POC	Size (μm)	Volume (μm^3)	SA : Vol
Calculated sensitivity							
C. leptoporus^{H}	30.3–212 **(106)**	15.8–146 **(60.9)**	14.5–82 **(44.3)**	0.50–2.21 **(1.29)**	11.3–14.7 **(12.6)**	1043	0.48
C. pelagicus^{I}	355–1330 **(667)**	81–796 **(325)**	139–1071 **(347)**	0.11–2.57 **(1.15)**	10.5–19.9 **(16.0)**	2140	0.38
E. huxleyi^{L}	5.9–45.6 **(18.4)**	2.9–21.3 **(7.2)**	2.5–34.2 **(11.3)**	0.24–1.38 **(0.67)**	4.5–5.1 **(4.8)**	58.1	1.25
G. oceanica^{H}	15.5–151 **(45.9)**	7.5–90.8 **(24.5)**	7.3–73.5 **(21.5)**	0.52–2.44 **(1.25)**	6.1	119	0.98
S. apsteinii^{I}	506–1269 **(844)**	170–811 **(381)**	317–679 **(465)**	0.42–1.44 **(0.83)**	12.0–22.4 **(17.2)**	2678	0.35
Estimated sensitivity							
C. leptoporus 1168^{H}	377–525 **(468)**	251–382 **(308)**	126–224 **(160)**	1.37–3.13 **(2.08)**	19.7	4003	0.30
C. leptoporus 1141^{H}	122–233 **(183)**	93.4–153 **(124)**	28.7–81.3 **(58.5)**	0.72–3.29 **(2.01)**	13.3	1232	0.45
C. leptoporus 1130^{L}	81.6–119 **(98.5)**	29.1–58.0 **(40.9)**	52.5–61.0 **(57.6)**	0.51–0.75 **(0.61)**	12.1	928	0.50
H. carteri 1323^{H}	401–562 **(481)**	278–384 **(331)**	124–178 **(151)**	2.29–2.30 **(2.30)**	14.7	1646	0.41
H. carteri 1334^{H}	450–614 **(532)**	290–379 **(334)**	161–235 **(198)**	1.59–1.82 **(1.71)**	15.8	2080	0.38
C. carterae^{L}	75.7–522 **(219)**	7.9–59.7 **(21.5)**	60.9–463 **(197)**	0.08–0.27 **(0.17)**	3.8–11.7 **(8.2)**	284	0.74
S. pulchra^{L}	121–174 **(156)**	20.9–30.0 **(24.2)**	98.6–145 **(131)**	0.15–0.22 **(0.19)**	9.5–10.3 **(9.9)**	511	0.60
U. sibogae$^{L/I}$	43.9	16.7	27.2	0.62	N/A	N/A	N/A

Data compiled from Balch et al. (1992), Langer et al. (2006), Rahbari (2009), Fiorini et al. (2011*a*,*b*), Krug et al. (2011), Heinle (2013), Daniels et al. (2014), Diner et al. (2015), Sett et al. (2014), Bach et al. (2015), Šupraha et al. (2015), Zhang et al. (2015), Gafar and Schulz (2018), and N. A. Gafar et al. (2019). Note: volume and surface area to volume ratio are calculated using the mean cell diameter, and *C. leptoporus* strains from Diner et al. (2015) were estimated, whereas those from Langer et al. (2006) were calculated. $^{L/I/H}$denote low/intermediate/high OA sensitivity species. Bold values in brackets denote average values as currently stated in the table description.

Production of POC and PIC at the community level drives facets of the so-called biological carbon pump (Riebesell et al. 2009) and can be split into contributions to the organic carbon and carbonate counter pumps, respectively (Westbroek et al. 1993). Production of POC at the surface ocean and subsequent transport to depth increases the surface ocean uptake capacity for atmospheric CO_2, whereas the carbonate counter pump has the opposite effect (Rost and Riebesell 2004).

Carbon production and growth rates of coccolithophores are significantly affected by environmental conditions (i.e., temperature, light, and CO_2; Langer et al. 2006; Bach et al. 2011; Sett et al. 2014; Zhang et al. 2015; N. A. Gafar et al. 2019). Human-induced perturbations to the global carbon cycle, namely increasing concentrations of atmospheric CO_2 will drive changes in ocean surface temperatures, light availability, and carbonate chemistry speciation (Rost and Riebesell 2004). The latter process is termed ocean acidification and leads to reductions in oceanic pH and carbonate ion concentration, whereas CO_2 and bicarbonate concentrations increase (Doney et al. 2009). Within a physiologically broad CO_2/pH range, simulating OA, optimum curve responses for PIC and POC production, and growth rates have been observed in all coccolithophores so far (Langer et al. 2006; Bach et al. 2011; Sett et al. 2014; Zhang et al. 2015). Sensitivities of PIC, POC, and growth rates to OA vary as a result of differing substrate use (Kottmeier et al. 2016), with those for PIC production being the most sensitive (Bach et al. 2015; Gafar and Schulz 2018; N. A. Gafar et al. 2019). As a result, OA influences the relative production of PIC and POC in coccolithophores. Sensitivities to OA also vary between species with some such as *G. oceanica* being much more sensitive than others like *E. huxleyi* and *S. apsteinii* (Gafar et al. 2018*b*; Gafar and Schulz 2018; N. A. Gafar et al. 2019). It is not yet clear what drives these differences; however, they are likely to result in shifts in coccolithophore community composition under climate change conditions (Rost and Riebesell 2004; Gafar and Schulz 2018). Through effects on both community composition and physiological rates, OA has the potential to impact PIC and POC production on a global scale.

Given the potential effects of OA on coccolithophorid-driven biogeochemical element cycling and feedbacks to Earth's climate system outlined above, it becomes paramount to not only assess species-specific sensitivities but also to start investigating whether there are common underlying cellular characteristics. In the present study, we therefore examine how differences in cell size-related parameters (i.e., surface area and volume) and variation in the relative production of PIC and POC may influence species-specific sensitivities to high CO_2.

Methods

Species comparison

Previously published data on responses of calcification, photosynthesis, and growth to simulated ocean acidification (increasing CO_2 and decreasing pH) were collected for *E. huxleyi* (Sett et al. 2014; Gafar and Schulz 2018), *G. oceanica* (Sett et al. 2014; Zhang et al. 2015; Gafar et al. 2018*b*), *S. apsteinii* (N. A. Gafar et al. 2019), *C. leptoporus* (Langer et al. 2006), and *Coccolithus pelagicus* subsp. *braarudii* strains RCC1198 (Bach et al. 2015) and RCC1200 (Krug et al. 2011). All clones were isolated between 1992 and 2001 except *C. pelagicus* subsp. *braarudii* strain RCC1198, which was isolated in 1958. The fCO_2 level required to suppress rates to half of maximum rates ($K^1_{\frac{1}{2}fCO_2}$ inhib), or in other words, the sensitivity to OA was calculated from fits of the data to equations describing the modulating effects of temperature (T), light (I), and carbonate chemistry (substrate: S = CO_2 and HCO_3^- and H = $[H^+]$) on metabolic rates (MR) developed elsewhere (Gafar et al. 2018*b*), with Eq. 1 for species where carbonate chemistry conditions varied at constant light and temperature (*C. leptoporus* and *C. pelagicus*), Eq. 2 for species where carbonate chemistry and light intensities varied at a constant temperature (*S. apsteinii*), and Eq. 3 for species where carbonate chemistry, temperature, and light intensities varied (*G. oceanica* and *E. huxleyi*). $K^1_{\frac{1}{2}fCO_2}$ inhib values were calculated for each species and individual light and temperature condition. Using these half-inhibition fCO_2 levels is an effective way to normalize differences in absolute rates between species and between different temperature and light conditions.

$$\mathrm{MR}(\mathrm{S},\mathrm{H}) = \frac{k_1 \mathrm{S}}{k_2 \mathrm{H} + k_3 \mathrm{SH} + k_4 + k_5 \mathrm{S}} \quad (1)$$

$$\mathrm{MR}(I,S,H) = \frac{k_1 SI}{k_2 H + k_3 SH + k_4 I + k_5 SI + k_6 SHI^2} \quad (2)$$

$$\mathrm{MR}(T,I,S,H) = \frac{k_1 SIT}{k_2 HT + k_3 SHT + k_4 I + k_5 SI + SIT + k_6 SHI^2 T^2} \quad (3)$$

PIC : POC ratios were then calculated from modeled MR of calcification and photosynthesis at fCO_2 levels corresponding to $K^1_{\frac{1}{2}fCO_2}$ inhib for each rate for each species (*see* Gafar et al. [2018*a*] for calculated values). The influence of average cell size, cell volume, and surface area to volume ratio (calculated from Table 1) on OA sensitivity was also examined. Fit coefficients for *G. oceanica*, *E. huxleyi*, and *S. apsteinii* were taken from Gafar et al. (2018*b*), Gafar and Schulz (2018), and N. A. Gafar et al. (2019). Fit coefficients for *C. leptoporus* and *C. pelagicus* strains RCC1198 and RCC1200 are presented in Supporting Information Tables S1–S3, respectively.

Results

There was no clear relationship between the five species mean cell diameter ($R^2 < 0.4135$, $F < 2.12$, $p > 0.24$), cell volume ($R^2 < 0.2755$, $F < 1.14$, $p > 0.36$), or surface area : volume

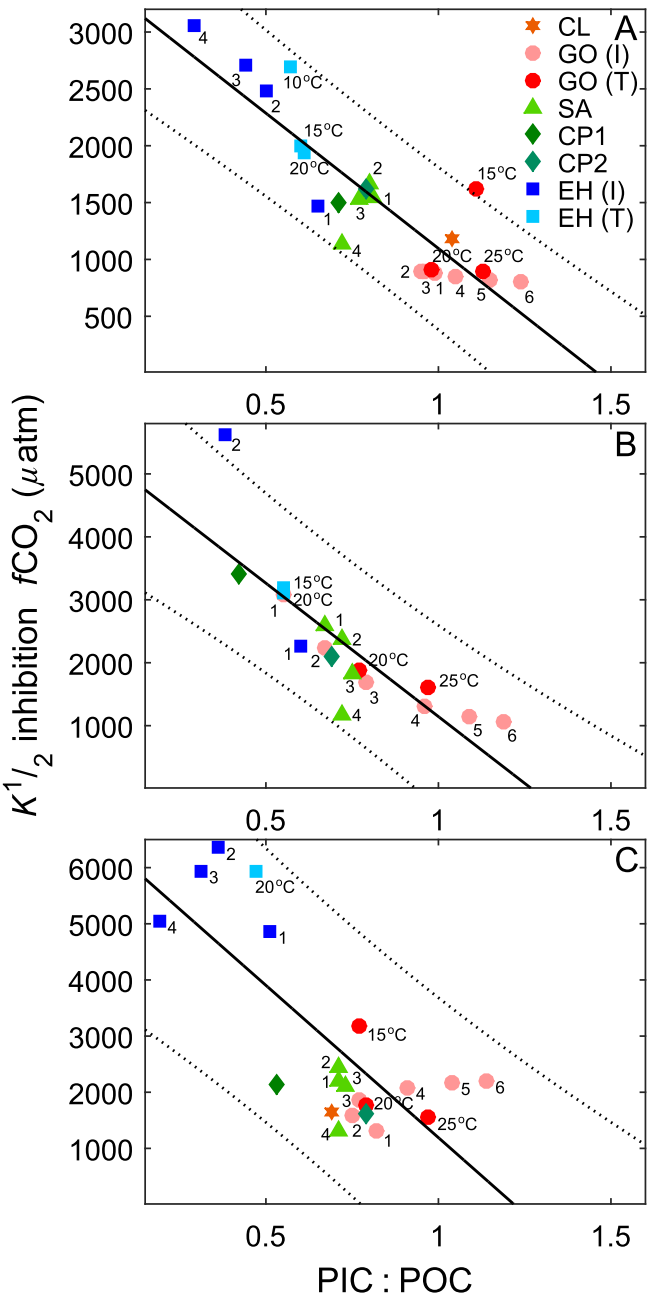

Fig. 1. PIC : POC ratios for *E. huxleyi* (EH), *G. oceanica* (GO), *S. apsteinii* (SA), *C. leptoporus* (CL), and *C. pelagicus* (CP) vs. the fCO_2 level at which (**A**) calcification, (**B**) photosynthetic carbon fixation, and (**C**) growth rates are inhibited to half of maximum $\left(K^1_{2CO_2}\text{ inhib}\right)$. (I) and (T) denote treatments under varying light and temperature conditions, respectively. Numbers 1–6 denote increasing light intensities for each species. The dotted line denotes the 95% prediction bounds for new observations. Note, under some conditions, rates were so insensitive to OA that suppression to half of maximum rates did not occur within the tested limits of our fit. In these cases, no values were calculated for PIC : POC.

ratio ($R^2 < 0.6107$, $F < 4.71$, $p > 0.12$; Table 1) and their mean OA sensitivity. However, there was an overall linear relationship of OA sensitivity and species-specific PIC : POC ratio at the fCO_2 of half-inhibition of calcification rates ($R^2 = 0.7727$, $F = 71.41$, $p < 0.05$, slope = −2380), photosynthetic rates ($R^2 = 0.6884$, $F = 35.35$, $p < 0.05$, slope = −4239), and growth rates ($R^2 = 0.5793$, $F = 26.16$, $p < 0.05$, slope = −5425), with higher ratios of PIC to POC resulting in inhibition already at lower fCO_2 (Fig. 1).

Modulation of PIC : POC ratios by light and temperature generally influenced $K^1_{2fCO_2}$ inhib as follows. In *E. huxleyi*, an increase in light availability decreased PIC : POC ratios and hence the sensitivity to OA (higher $K^1_{2fCO_2}$ inhib), whereas an increase in temperature had the opposite effect (Fig. 1). In *G. oceanica*, an increase in both light intensities and temperature increased both PIC : POC ratios and sensitivity to OA (lower $K^1_{2fCO_2}$ inhib; Fig. 1). Finally, in *S. apsteinii*, there was no clear trend of light availability on PIC : POC ratios and hence sensitivity to OA (Fig. 1).

Discussion

The influence of cellular metrics on OA sensitivity

Changes in cell size-related geometry and variation in the relative production of PIC and POC both could influence a species sensitivity to OA. $CaCO_3$ formation, via calcification, is thought to result in generation of H^+ in the coccolith producing vesicle (Paasche 2001; Brownlee and Taylor 2004) and has been observed to result in a decrease in intracellular pH in *E. huxleyi* (Suffrian et al. 2011; Fig. 2). As such, more heavily calcifying species produce more H^+. As coccolithophores need to maintain intracellular pH homeostasis, this excess H^+ must be dealt with. One option is through removal across the plasma membrane (Suffrian et al. 2011; Taylor et al. 2011). Another option is neutralization of excess H^+, for example, by reacting with HCO_3^- in the cytosol to form CO_2 for photosynthesis (Fig. 2) or by reacting with hydroxyl ions (OH^-) exported from the chloroplasts after internal conversion of HCO_3^- into CO_2 (Anning et al. 1996; Buitenhuis et al. 1999; Paasche 2001). Removing/neutralizing excess H^+ is likely to be more difficult for more heavily calcified species, as they must remove larger amounts of H^+ relative to less calcified species (Bach et al. 2015). Removing/neutralizing excess H^+ is also likely to be more difficult for larger species as they have a much lower surface area, relative to volume, to transport H^+ across. In addition to this, the removal of H^+ will become more difficult under high CO_2/low pH conditions because of a reduced electrochemical gradient across the plasma membrane, so it would be expected that species with high PIC : POC ratios may be more quickly unable to maintain optimal intracellular pH as seawater pH levels decrease. As a result, large, high PIC : POC species would be expected to be more strongly inhibited by H^+ at lower CO_2 levels than lighter and smaller PIC : POC species.

It should be noted that the influence of PIC : POC on the OA sensitivities of growth rates is not as clear as those for calcification and photosynthetic carbon fixation rates. Interestingly,

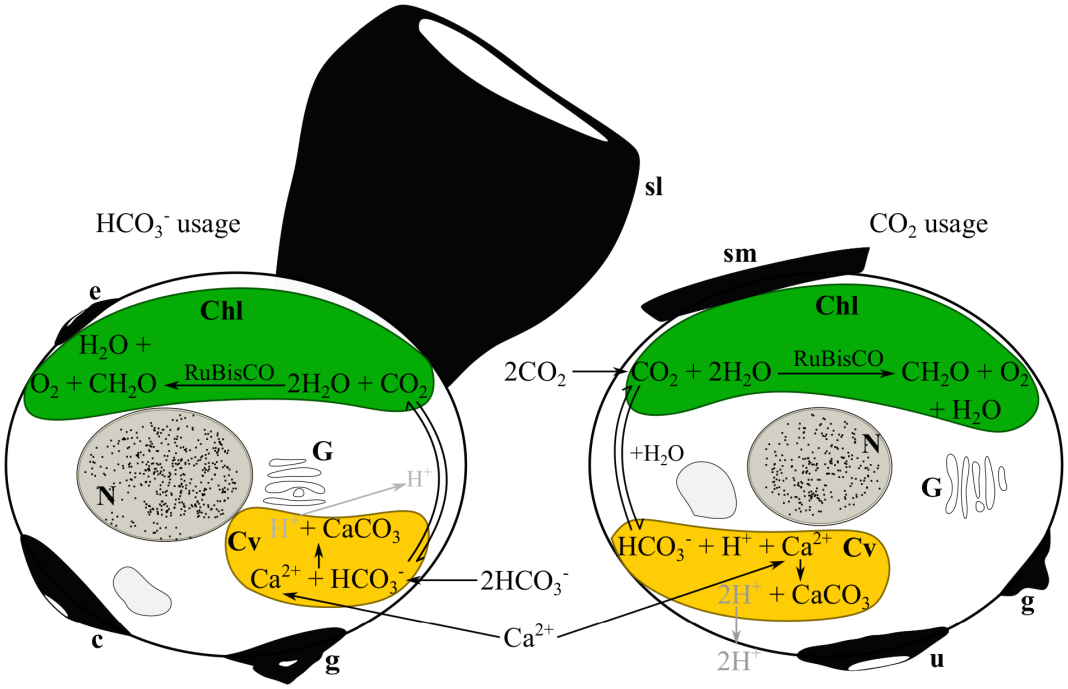

Fig. 2. Schematic drawing depicting various modes of inorganic carbon uptake (CO_2 and HCO_3^-) via the plasma membrane, their mechanisms of use by photosynthetic carbon fixation in the chloroplast (Chl) and by calcification in the coccolith production vesicle (Cv) and the related H^+ budget. Note neutralization of excess H^+, produced via calcification, can occur when photosynthesis is fuelled by HCO_3^-. While N denotes the nucleus, G depicts the Golgi vesicle. The variety of coccolith morphology in various species (e, *E. huxleyi*; u, *Umbillicosphaera* sp.; g. *Gephyrocapsa* sp.; c, *Calcidiscus* sp.; sm, *S. apsteinii* murolith; sl, *S. apsteinii* lopadolith) is illustrated by silhouettes.

it is only the larger species, of intermediate sensitivity, which do not follow the PIC : POC pattern observed for the other rates. It is possible that as cell sizes get larger that cell size begins to influence sensitivities to OA more than differences in PIC : POC (which influences small species more strongly). The reason that a similar pattern is not observed for all rates is likely because of the higher sensitivity of photosynthetic carbon fixation and calcification to OA. The higher OA sensitivity likely strengthens the influence of PIC : POC on OA sensitivity to the point that size-related effects are obscured. This would also explain why the relationship of PIC : POC and OA sensitivity becomes clearer/stronger as we move from growth (least sensitive) to photosynthetic carbon fixation (intermediate sensitivity) and finally to calcification (most sensitive). It is important to keep in mind, however, that already relatively small decreases in growth rate of much less than a 50% reduction as shown in Fig. 1C have been found and calculated to highly impact bloom formation potential and hence community production, with a ~ 50% decline in cellular standing stocks and a 1.8 times decrease in community production of $CaCO_3$ with a growth rate decrease of 10% (*see* Gafar et al. [2018*b*] and references therein).

When all five coccolithophore species are combined, a significant relationship is found between PIC : POC ratio and CO_2 sensitivity, with higher PIC : POC ratios resulting in a greater sensitivity to OA in all rates (Fig. 1). This indicates that PIC : POC ratios may be a driving factor behind species-specific sensitivities to OA, especially in terms of PIC and POC production. As $K^1_{\frac{1}{2}fCO_2}$ inhib for all rates are generally lower (indicating higher sensitivity) in higher PIC : POC species, it would be expected that all rates will be more heavily suppressed vs. lower PIC : POC species at the same CO_2 level. Relative growth rates, and the photosynthetic rates which support them, are an important measure of competitive ability between species (Gafar and Schulz 2018). It has been estimated that the relatively higher decrease in growth rates of species which are more sensitive to high CO_2 will result in a drop in relative abundance/dominance under rising CO_2 levels (i.e., *G. oceanica* vs. *E. huxleyi* in Gafar and Schulz [2018]). It is this reduction in relative competitive ability (driven by differences in PIC : POC ratios) that may shift the balance toward greater relative abundances of lower PIC : POC species under future ocean conditions.

$CaCO_3$ production in the surface ocean shifts carbonate chemistry speciation toward CO_2; hence, a reduction in overall PIC production increases the storage capacity for atmospheric CO_2 (Zeebe and Wolf-Gladrow 2001). At the same time, it appears that PIC, through ballasting of organic carbon aggregates, is responsible for an effective mode of POC export to depth (Klaas and Archer 2002), although there are most likely other processes at work and their individual contribution, especially at the local scale are difficult to reconcile

(*see* Bach et al. [2016] and references therein). A reduction in PIC-driven POC export would then weaken the biological pump and decrease the ocean's storage capacity for atmospheric CO_2 (Barker et al. 2003). When both effects are considered together, model results suggest that the effect of reduced PIC production and reduced ballasting can nullify each other (Barker et al. 2003; Ridgwell 2003; Heinze 2004; Hofmann and Schellnhuber 2009), but the actual net effect depends on a number of assumptions, i.e., remineralization length scales, PIC : POC ballasting effect, influence of OA on PIC production (based on *E. huxleyi* only in most models), and sediment feedbacks ($CaCO_3$ compensation when riverine input is mismatched by deep-sea burial). Hence, what the overall effect of a shift to lower PIC : POC species, due to OA, for atmospheric CO_2 would be is difficult to judge at the moment.

Unlike PIC : POC ratios, cell size-related geometry does not appear to affect sensitivities and responses to changing carbonate chemistry, at least for calcification and photosynthesis. For instance, *G. oceanica* has similar sensitivities to *C. leptoporus* (798–1626 vs. 1183 μatm) even though *C. leptoporus* has 10 times the volume (Table 1; Fig. 1A). At the same time, *E. huxleyi*, which is a similar size to *G. oceanica*, is half as sensitive to OA (1462–3058 μatm). It may be that the differences in size between the five species are simply not large enough in comparison to the differences in PIC : POC ratio to clearly observe a size-related effect on OA sensitivity. In addition, it may be that any effect of size which is present is being masked by the differences in PIC : POC ratio between the species (as proposed for growth rates above). Comparison of species of different sizes and similar PIC : POC ratio may be required for the presence/absence of a cell size influence to be determined.

Interestingly, the finding that calcification rates are typically more sensitive to OA than photosynthetic carbon fixation and growth rates in a single species (i.e., Sett et al. 2014; Bach et al. 2015; Zhang et al. 2015; Gafar and Schulz 2018) does also apply to our multispecies analysis. This is reflected in process-specific slopes of sensitivity to PIC : POC ratios (e.g., Fig. 1), which are the least steep (most sensitive) for calcification, followed by photosynthetic carbon fixation and growth rates (−2380, −4239, and −5425, respectively).

The influence of temperature and light on PIC : POC ratios and high CO_2/H^+ sensitivity

Light and temperature are known to individually modulate PIC and POC production of coccolithophores (Sett et al. 2014; Zhang et al. 2015; Gafar and Schulz 2018; N. A. Gafar et al. 2019), and in most cases, the resulting changes in PIC : POC ratio were directly reflected in corresponding shifts in $K^1_{\frac{1}{2}fCO_2}$ inhib (i.e., following the fit line in Fig. 1A,B). This was generally the case for *E. huxleyi*, where increasing light intensities lowered PIC : POC ratios and OA sensitivity, whereas for *G. oceanica* both increased. In contrast, there was no clear trend of light on PIC : POC ratios and associated OA sensitivity in *S. apsteinii*, and it appeared that in some instances (e.g., *G. oceanica* at 15°C and *S. apsteinii* at high light), there might be effects of light and temperature on other cellular functions, resulting in an unrelated PIC : POC and sensitivity response. It is clear that, at least, the effect of changing light intensity on PIC : POC, and therefore, sensitivities to OA, may be species specific. As a result, different species may become more or less sensitive to OA under the changes in light availability and temperature predicted over the following decades (Rost and Riebesell 2004; Stocker et al. 2013). As such, differences in temperature and light conditions should be kept in mind when comparing coccolithophore responses to OA.

Estimating OA sensitivity using current day PIC : POC ratios

Coccolithophores can be broadly split into three groups with low (*E. huxleyi*), intermediate (*S. apsteinii* and *C. pelagicus*), and high (*G. oceanica* and *C. leptoporus*) sensitivity to OA (Fig. 1). All calcifying coccolithophores studied to date show either no change or a general decrease in PIC : POC ratio with increasing pCO_2 (e.g., Krug et al. 2011; Fiorini et al. 2011*b*; Sett et al. 2014; Diner et al. 2015). As such, it should be possible to tentatively place species for which little or no CO_2 response data exist, into these three general sensitivity groups using what PIC : POC ratios are available.

PIC : POC ratios of below 0.3 for *Syracosphaera pulchra* (Fiorini et al. 2011*a*,*b*) and *C. carterae* (Heinle 2013) suggest that these two species belong to the low-sensitivity group with *E. huxleyi*. Atlantic (RCC1323) and Mediterranean (RCC1334) strains of *Helicosphaera carteri* with average PIC : POC ratios of 2.30 and 1.71, respectively (Šupraha et al. 2015), likely belong in the high-sensitivity group with *G. oceanica*. Finally, *Umbilicosphaera sibogae*, with a PIC : POC ratio of 0.62 (Balch et al. 1992), is in between the low- and intermediate-sensitivity groups.

Concerning strain-specific sensitivities to OA, it appears that the PIC : POC ratio may also be the driving factor. For instance, calcification rates in *C. leptoporus* decreased markedly with a tripling of pCO_2 in two strains with relatively high average PIC : POC ratios of 2.01 and 2.08 (RCC1141 and RCC1168; Diner et al. 2015), suggesting placement in the high-sensitivity group. In contrast, calcification and photosynthetic rates in strain RCC1130, with a comparatively low average PIC : POC ratio of 0.61, were hardly affected, placing it in the low-sensitivity group with *E. huxleyi*.

Differences in OA response between different strains have also been observed for *E. huxleyi*, with differences in response also observed between morphotypes (Langer et al. 2009; Müller et al. 2015). Morphotypes of *E. huxleyi* are physiological variants, which differ in coccolith morphology and degree of calcification of individual liths (Hagino et al. 2011). As such, it might be expected that different morphotypes may have a different degree of cellular calcification (or PIC : POC ratio) and thus sensitivity to OA. Some morphotypes with heavily calcified liths (A overcalcified) do indeed have higher cellular PIC

quotas and PIC : POC ratios than strains of morphotypes with less calcified liths (B/C) (Beaufort et al. 2011; Müller et al. 2015). At the same time, there have also been observations of no real difference in cellular PIC quota and PIC : POC ratios between different morphotypes like the R overcalcified type RCC1216, delicate B type RCC1212, and two intermediate A types RCC1238 and RCC1256 (Langer et al. 2009) and the R overcalcified types CH352 and CH360 and the moderately calcified CH428 (von Dassow et al. 2018). Interestingly, in the latter study, the similarity in cellular PIC : POC between strains/morphotypes was associated with a similar OA sensitivity. This disconnect between visual and measured degree of calcification of a strain could provide an explanation of reports of "heavily calcified" morphotypes of *E. huxleyi* dominating in relatively low pH waters in winter in the Bay of Biscay (Smith et al. 2012), the Benguela coastal upwelling (Henderiks et al. 2012), and the coastal zone of central Chile (Beaufort et al. 2011; von Dassow et al. 2018).

Finally, it is interesting to note that the two bloom-forming species *E. huxleyi* and *G. oceanica* belong to opposite ends of the CO_2 sensitivity scale. Despite this, both species are bloom formers and the most commonly observed species in modern coccolithophore communities (McIntyre and Bé 1967; Roth and Coulbourn 1982). It is likely that current day CO_2 levels are not yet high enough to impact the blooming/success of either species. As a result, OA sensitivity does not currently dictate their relative success when compared to other environmental factors like temperature (Gafar and Schulz 2018).

Conclusion

Combined data from *S. apsteinii*, *E. huxleyi*, *G. oceanica*, *C. pelagicus*, and *C. leptoporus* indicate that the PIC : POC ratio is a major driver behind species and potentially also strain-specific sensitivities of calcification, photosynthesis, and growth to OA (high CO_2/H^+), with higher PIC : POC ratios resulting in greater sensitivity. Meanwhile, cell size, volume, and surface area to volume ratio have no clear influence on OA sensitivities. Hence, it may be expected that, under future ocean conditions, coccolithophore communities may shift toward greater abundances of lower sensitivity, low PIC : POC species and strains. This has the potential to impact the marine carbon cycle on the regional scale.

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