RESEARCH FRONT

CSIRO PUBLISHING

Marine and Freshwater Research, 2020, **71**, 263–274
https://doi.org/10.1071/MF19267

Perspective

Ocean acidification as a multiple driver: how interactions between changing seawater carbonate parameters affect marine life

Catriona L. Hurd[A,B,M], *John Beardall*[C], *Steeve Comeau*[D], *Christopher E. Cornwall*[E], *Jonathan N. Havenhand*[F], *Philip L. Munday*[G], *Laura M. Parker*[H], *John A. Raven*[I,J,K] *and Christina M. McGraw*[L,M]

[A]Institute for Marine and Antarctic Studies, University of Tasmania, 20 Castray Esplanade, Battery Point, Hobart, Tas. 7004, Australia.
[B]Discipline of Biological Sciences, Private Bag 55, Hobart, Tas, 7001, Australia.
[C]School of Biological Sciences, Monash University, Clayton, Vic. 3800, Australia.
[D]Sorbonne Université, Centre National de la Recherche Scientifique – L'Institut national des sciences de l'Univers, Laboratoire d'Océanographie de Villefranche, 181 chemin du Lazaret, F-06230 Villefranche-sur-mer, France.
[E]School of Biological Sciences, Victoria University, Kelburn, Wellington, 6140, New Zealand.
[F]Department of Marine Sciences, University of Gothenburg, Tjärnö Marine Laboratory, SE-45296 Strömstad, Sweden.
[G]Australian Research Council Centre of Excellence for Coral Reef Studies, James Cook University, Townsville, Qld 4811, Australia.
[H]School of Biological, Earth and Environmental Sciences, University of New South Wales, Sydney, NSW 2052, Australia.
[I]Division of Plant Science, University of Dundee at the James Hutton Institute, Invergowrie, Dundee, DD2 5DA, UK.
[J]Climate Change Cluster, University of Technology Sydney, Ultimo, NSW 2007, Australia.
[K]School of Biological Science, University of Western Australia, 35 Stirling Highway, Crawley, WA 6008, Australia.
[L]Department of Chemistry, University of Otago, Dunedin, 9054, New Zealand.
[M]Corresponding authors. Email: catriona.hurd@utas.edu.au; christina.mcgraw@otago.ac.nz

Abstract. 'Multiple drivers' (also termed 'multiple stressors') is the term used to describe the cumulative effects of multiple environmental factors on organisms or ecosystems. Here, we consider ocean acidification as a multiple driver because many inorganic carbon parameters are changing simultaneously, including total dissolved inorganic carbon, CO_2, HCO_3^-, CO_3^{2-}, H^+ and $CaCO_3$ saturation state. With the rapid expansion of ocean acidification research has come a greater understanding of the complexity and intricacies of how these simultaneous changes to the seawater carbonate system are affecting marine life. We start by clarifying key terms used by chemists and biologists to describe the changing seawater inorganic carbon system. Then, using key groups of non-calcifying (fish, seaweeds, diatoms) and calcifying (coralline algae, coccolithophores, corals, molluscs) organisms, we consider how various physiological processes are affected by different components of the carbonate system.

Additional keywords: coccolithophores, coralline algae, corals, diatoms, fertilisation, fish, macroalgae, molluscs, seaweed.

Received 2 August 2019, accepted 23 September 2019, published online 29 November 2019

Introduction

A key factor in the development of ocean acidification (OA) as an independent research field over the past 15 years has been collaborations between chemists and biologists, particularly in experimental work to understand the effects of OA on marine life (Riebesell *et al.* 2010). This is exemplified by the OA research team at the University of Otago (New Zealand), led by Professor Keith Hunter: early collaborations focused on

biologists learning how to correctly manipulate the seawater carbonate system in a way that mimics 'natural future changes' (Hurd *et al.* 2009). This included the development of new culture systems to grow marine organisms that accurately measure and regulate the seawater carbonate system (McGraw *et al.* 2010; Cummings *et al.* 2011; Hoffmann *et al.* 2013) and fluctuate pH in a way that mimics coastal oceans (Cornwall *et al.* 2013; Reidenbach *et al.* 2017) while ensuring correct replication of experimental units (Cornwall and Hurd 2016).

With the rapid expansion of OA research has come a greater understanding of the complexity and intricacies of how the multiple changes to the seawater carbonate system are affecting marine life. At the same time, research is increasing into the many other environmental properties that are changing simultaneously. These include global drivers, such as temperature, underwater light regime, O_2 concentration and altered nutrient supply, as well as local drivers, such as eutrophication, urbanisation, pollution. These multiple drivers interact with each other, and with OA, in ways that can be unpredictable (Crain *et al.* 2008; Boyd *et al.* 2018).

OA itself can also be viewed as a multiple driver because there are many chemical parameters changing simultaneously, including total dissolved inorganic carbon (DIC), CO_2, HCO_3^-, CO_3^{2-}, H^+ and the carbonate saturation state (Ω). Each of these chemical parameters can have different physiological effects on an organism, which may affect growth and survival in unpredictable ways. For example, rates of gross calcification in mussels increase with OA because HCO_3^-, which is the source of carbonate for calcification, increases. However, at the same time, increased dissolution of the outer shell layer occurs as Ω decreases (Rodolfo-Metalpa *et al.* 2011). Therefore, the effect of OA on net calcification of bivalves and gastropods will be the balance of increasing DIC (positive) and decreasing Ω (negative), as well as the extent to which their outer shell is protected from decreasing Ω by the proteinaceous periostracum (Rodolfo-Metalpa *et al.* 2011).

Here, we begin by clarifying our understanding of the terms used by chemists and biologists to describe the changing seawater inorganic carbon system. Using key groups of calcifying and non-calcifying organisms known to be affected by OA, we then consider how the different components of the carbonate system affect them, both separately and interactively.

Seawater carbonate system

As pCO_2 in the atmosphere increases, more CO_2 is being driven into the surface seawater, increasing the concentration of aqueous CO_2. When CO_2 is absorbed by seawater, it will react with water to form carbonic acid (H_2CO_3):

$$CO_2(aq) + H_2O \rightleftharpoons H_2CO_3(aq) \quad (1)$$

H_2CO_3 will then dissociate to form bicarbonate (HCO_3^-) and a proton (H^+):

$$H_2CO_3(aq) \rightleftharpoons HCO_3^-(aq) + H^+(aq) \quad (2)$$

The H^+ can then react with carbonate (CO_3^{2-}), producing additional HCO_3^- at the expense of CO_3^{2-}:

$$CO_3^{2-}(aq) + H^+(aq) \rightleftharpoons HCO_3^-(aq) \quad (3)$$

These equilibrium reactions illustrate the consequence of increasing atmospheric CO_2 concentrations: CO_2, HCO_3^- and H^+ are increasing, while CO_3^{2-} is decreasing.

The term 'ocean acidification' comes from the increasing H^+ concentration (Eqn 2). However, because $[H^+]$ can vary by a factor of 10^{14} in water, acidity is usually expressed as pH, where an increasing acidity (i.e. increasing $[H^+]$), is expressed as a decreasing pH:

$$pH = -\log[H^+] \quad (4)$$

It is important to note that 'H^+' is chemical shorthand: in aqueous solutions, H^+ does not exist as written. Instead, the H^+ will be transferred from the proton donor (e.g. H_2CO_3) to water, forming a hydronium ion (H_3O^+). The H^+ will be transferred between water molecules, but will remain as H_3O^+ in seawater. However, in this paper, we will continue with the practice of expressing H_3O^+ as H^+ (or 'a proton').

In addition to the increasing acidity, the absorption of CO_2 leads to an increase in the total DIC:

$$DIC = [CO_2] + [H_2CO_3] + [HCO_3^-] + [CO_3^{2-}] \quad (5)$$

Because it is difficult to distinguish between $[CO_2]$ and $[H_2CO_3]$, these two terms are often combined into a single parameter, $[CO_2^*]$, which simplifies the expression for DIC:

$$DIC = [CO_2^*] + [HCO_3^-] + [CO_3^{2-}] \quad (6)$$

Although HCO_3^- is the dominant form of DIC in seawater, the relative amount of each species is dependent on seawater pH. Thus, it is important to recognise that a change in pH is an outcome of changes in the dissolved inorganic carbon system rather than a driver. As OA continues to decrease the pH of seawater, the position of the equilibrium shifts towards HCO_3^-, leading to substantial decreases in CO_3^{2-}. For example, as pCO_2 increases from 280 to 560 ppm, HCO_3^- is expected to increase by 12.3%, whereas CO_3^{2-} will decrease by 36.9% (Table 1). At the same time, the $CO_2 : HCO_3^-$ ratio will increase by 79.1%.

Decreasing CO_3^{2-} is also leading to decreasing Ω: Ω, which describes the thermodynamic stability of $CaCO_3$, decreases proportionally with decreasing $[CO_3^{2-}]$:

$$\Omega = \frac{[Ca^{2+}][CO_3^{2-}]}{K_{sp}} \quad (7)$$

where K_{sp} is the thermodynamic solubility product and will depend on the morphology of the $CaCO_3$ structure (e.g. calcite or aragonite) and the temperature, pressure, salinity and composition of the seawater. The appeal of Ω in describing the inorganic carbon system lies in its simplicity: when Ω is ≥ 1, the inorganic $CaCO_3$ structure is considered chemically stable; however, when Ω is <1, dissolution of the $CaCO_3$ structure is energetically favoured. It is important to remember that Ω refers only to the thermodynamic stability of the $CaCO_3$ structure, and not the ability of an organism to calcify.

Table 1. Dissolved inorganic carbon (DIC) parameters for current and future scenarios relative to the percentage change (Δ) since pre-Industrial values

Calculations were done with CO2sys (Lewis and Wallace 1998) using the carbonate equilibrium constants of Lueker *et al.* (2000), sulfate constants of Dickson (1990), hydrofluoric acid constants of Perez and Fraga (1987) and total boron of Lee *et al.* (2010). The calculations were based on the pCO_2 concentrations given in the table and alkalinity, temperature and salinity values of 2300 μmol kg^{-1}, 20°C and 35‰. [CO_2*], single parameter combining [CO_2] and [H_2CO_3]; Ω, saturation state

	Scenario				
	Pre-Industrial	Current	Δ (%)	2× Pre-industrial	Δ (%)
pCO_2 (μatm)	280	410	46.6	560	100
DIC (μmol kg^{-1})	1965.9	2044.4	4.0	2102.2	6.9
[CO_2*] (μmol kg^{-1})	9.0	13.2	46.7	18.1	101
[HCO_3^-] (μmol kg^{-1})	1726.3	1850.3	7.2	1938.7	12.3
[CO_3^{2-}] (μmol kg^{-1})	230.5	180.9	−21.5	145.4	−36.9
$CO_2 : HCO_3^-$	5.21×10^{-3}	7.13×10^{-3}	36.8	9.34×10^{-3}	79.1
[H^+] (mol kg^{-1})	6.7×10^{-9}	9.2×10^{-9}	36.6	1.2×10^{-8}	78.1
pH_T	8.17	8.04		7.92	
$\Omega_{calcite}$	5.51	4.33	−21.5	3.48	−36.9
$\Omega_{aragonite}$	3.58	2.81	−21.5	2.26	−36.9

Historical perspective on understanding the biological responses of marine organisms to OA

Early OA research focused on the effects on calcification, because calcifiers were considered particularly vulnerable to the declining CO_3^{2-} and Ω. From a biological perspective, the implication that seawater CO_3^{2-} is used directly in calcification was a distraction because most calcifying organisms acquire CO_3^{2-} for calcification by metabolic modification of CO_2 or HCO_3^-; indeed, the carbonate ion is rarely transported across cellular membranes (Hofmann and Todgham 2010). These processes can occur internally or externally, depending on the mechanisms of calcification (Roleda *et al.* 2012*a* and references therein), but are independent of seawater CO_3^{2-}. Gaining a more complete mechanistic understanding of how key groups, namely coccolithophores, corals, coralline algae and shellfish (i.e. gastropods and bivalves), calcify has been crucial in advancing our comprehension of how the various components of the seawater inorganic carbon system affect physiological and biochemical processes, and the relative susceptibility of species.

A theme that developed after the initial focus on calcifiers was how OA may affect non-calcifying marine life, including algae (i.e. phytoplankton such as diatoms and 'fleshy' seaweeds) and higher trophic levels, including fish (Hurd *et al.* 2018). A meta-analysis suggested that micro- and macro-algae will benefit in a future ocean because of a positive effect of increasing CO_2 on photosynthesis and growth (Kroeker *et al.* 2013). However, physiological studies on algae indicate a range of responses that can be positive, negative or neutral depending on the relative ability to use CO_2 and HCO_3^- and whether increasing H^+ affects cellular homeostasis (van der Loos *et al.* 2019). Similarly, changes in CO_2, HCO_3^- and H^+ can affect physiological processes in fish, with a range of downstream effects on individual performance (Heuer and Grosell 2014). Understanding the mechanisms by which the different components of the seawater carbonate system interact to affect organisms is key to understanding responses to OA.

Non-calcifiers

Fish

Environmental pCO_2 is the primary component of the changing inorganic carbon system that is of concern to fishes, but even here it can have diverse effects. An increase in environmental pCO_2 causes the pCO_2 in fish blood to rise. To counter the acidotic effect of higher extracellular pCO_2, fish excrete additional H^+ by the gills and buffer the plasma with extra HCO_3^-, which is mostly taken up from the water by the gills (Heuer and Grosell 2014). However, this change in the internal inorganic carbon chemistry to maintain a steady pH is not without consequences. For example, calcification processes are typically enhanced by increased pCO_2 in fishes. Several studies have found that the aragonitic otoliths of fishes are larger in larval fish reared under high CO_2 (Checkley *et al.* 2009). The likely explanation for this phenomenon is that the accumulation of HCO_3^- in the blood to buffer acid–base balance results in more HCO_3^- being transported into the fluid surrounding the otolith, providing more substrate for otolith formation (Heuer and Grosell 2014). Elevated internal CO_2 and HCO_3^- levels also lead to increased $CaCO_3$ precipitation in the fish gut (Heuer and Grosell 2016). Because fish contribute up to 15% of global production to the inorganic carbon cycle (Wilson *et al.* 2009), an increase in fish $CaCO_3$ excretion under OA conditions could be significant.

Less favourable effects of elevated CO_2 in fish are changes to sensory capacities and a wide range of altered behaviours (Nagelkerken and Munday 2016). The likely mechanism for the diverse behavioural changes that have been observed is the effect of elevated HCO_3^- levels on $GABA_A$ receptor function (Nilsson *et al.* 2012). GABA (γ-aminobutyric acid) is the primary inhibitory neurotransmitter in the vertebrate brain and the $GABA_A$ receptor is an ion channel with permeability for HCO_3^- and Cl^-. The increased concentration of extracellular HCO_3^- used to buffer blood pH can alter the function of the $GABA_A$ receptor, potentially altering its polarity, causing downstream effects on cognition and behaviour (Nilsson *et al.* 2012; Heuer *et al.* 2016; Schunter *et al.* 2016). New studies are showing that higher concentrations of H^+ in seawater associated with OA can alter the molecular structure of some odorants, having a direct effect on fish olfactory abilities (Roggatz *et al.* 2016; Velez *et al.* 2019).

Fleshy (non-calcifying) seaweeds

Seaweeds are (mostly) benthic coastal primary producers from three phyla (Rhodophyta, Ochrophyta and Chlorophyta) that dominate temperate waters where they provide 50% of primary production, recycle nutrients and form habitats for higher trophic levels (Hurd *et al.* 2014). Most seaweeds can take up

Table 2. Physiological processes affected by the changing dissolved inorganic carbon (DIC) parameters associated with ocean acidification
Ω, saturation state; CA, carbonic anhydrase; CCMs, CO_2-concentrating mechanisms

Organism	DIC parameter			
	H^+	CO_2	HCO_3^-	Ω
Fish	Indirect effect of altered odorant on fish olfactory system	Blood pH Otolith calcification Calcium carbonate precipitation in gut	Sensory capacity by GABA	
Fleshy seaweed	Cellular homeostasis	Downregulation of CCMs and external CA Bicarbonate transport Photosynthesis	Photosynthesis	
Diatoms	Silicification	Downregulation of CCMs and external CA Bicarbonate transport Photosynthesis	Photosynthesis	
Coccolithophores	Intracellular acid–base regulation	Photosynthesis	Photosynthesis Calcification	
Corals	Calcification, cellular homeostasis, photosynthesis		Calcification, photosynthesis, cellular homeostasis	Gross dissolution
Coralline algae	Calcification, cellular homeostasis		Calcification, cellular homeostasis	Gross dissolution
Molluscs	Gross calcification	Gross dissolution of the inner shell	Gross calcification	Gross calcification and gross dissolution
Fertilisation	Mitochondrial membrane potential Sperm motility	Sperm motility		

dissolved CO_2 by passive diffusion at a rate sufficient to saturate photosynthesis, an 'energetically cheap' acquisition strategy, provided that the CO_2 concentration is high enough (Raven *et al.* 2014). However, many seaweeds can also use HCO_3^-, the uptake of which has a higher energetic demand using direct active uptake or the extracellular conversion to CO_2 by the enzyme carbonic anhydrase (CA), with active transport occurring at the chloroplast membrane. Some tropical seaweeds appear to use only HCO_3^- (Diaz-Pulido *et al.* 2016). Algae that directly use HCO_3^- have a CO_2-concentrating mechanism (CCM species) that increases the supply of CO_2 to the active site of Rubisco (ribulose bisphosphate carboxylase–oxygenase), thereby enhancing its carboxylation efficiency. However, it is estimated that one-third of seaweed species take up CO_2 by diffusion only under present inorganic carbon supply conditions (mostly red seaweeds, Phylum Rhodophyta) and do not operate a CCM (termed 'non-CCM species'; Raven *et al.* 2014; Cornwall *et al.* 2015; Kübler and Dudgeon 2015). Hence, the energetics and mechanisms of DIC acquisition by seaweeds, and the relative availability of HCO_3^- and CO_2, and their changes in genetic adaptation will define their productivity in a future high-CO_2 ocean.

DIC 'uptake strategies' are being used to help predict the responses of seaweeds to OA. To date, six strategies have been identified based on field surveys (Hepburn *et al.* 2011; Cornwall *et al.* 2015, 2017*b*; Diaz-Pulido *et al.* 2016), laboratory studies (Cornwall *et al.* 2012; Britton *et al.* 2019; van der Loos *et al.* 2019) and a literature survey (van der Loos *et al.* 2019): (1) non-CCM with low affinity for DIC; (2) non-CCM with high affinity for DIC; (3) low-affinity CCM; (4) high-affinity CCM with no downregulation of the CCM; (5) high-affinity CCM with downregulation of the CCM; and (6) HCO_3^- only. Species with a low affinity for CO_2 or HCO_3^- are predicted to benefit in a future ocean because DIC will limit productivity under today's oceanic conditions, whereas no change is predicted for those with high-affinity uptake mechanisms because photosynthesis is saturated for DIC.

Testing the predicted responses of seaweeds requires a mechanistic understanding of DIC uptake processes (i.e. the six noted above), which are not well known for most seaweeds. For *Macrocystis pyrifera* and *Ulva rigida*, both of which have a high-affinity CCM, there was no effect of OA on rates of growth, photosynthesis or the activity of CA, supporting the predictions of Mechanism 4 above (Fernández *et al.* 2015; Rautenberger *et al.* 2015). A literature survey examining the growth responses of seaweeds to increased CO_2 revealed that for 55 CCM seaweeds, 21 had no response, whereas 15 had increased growth and 5 had decreased growth relative to ambient seawater, but the mechanisms of DIC uptake are unknown for most species (Table 2; van der Loos *et al.* 2019). The growth responses of only five non-CCM species have been tested, again with mixed results: the growth rates of three species were unaffected, whereas growth was enhanced in two species (Kübler *et al.* 1999; Kram *et al.* 2016; Britton *et al.* 2019; van der Loos *et al.* 2019).

In addition, some species may be sensitive to increasing H^+ concentrations, which could affect cellular homeostasis and may act antagonistically to any benefits of increasing DIC (van der Loos *et al.* 2019). However, for *M. pyrifera* gametophytes, the negative effects of H^+ on growth were ameliorated by the addition of CO_2, illustrating that CO_2 and H^+ have separate and interactive effects on metabolism (Roleda *et al.* 2012*b*).

Phytoplankton: diatoms

Diatoms (Phylum Ochrophyta) are an important group of marine phytoplankton, accounting for ~40% of oceanic primary productivity (Falkowski 1994; Field *et al.* 1998; Armbrust 2009). Any changes to the seawater carbonate system are likely to have profound effects on marine productivity and global carbon cycles.

Diatoms can take up both CO_2 and HCO_3^- from seawater, but the extent to which they show preferential use of one or other of these is highly species dependent. For example, *Nitzschia navis-varingica* predominantly uses CO_2, whereas *Stellarima stellaris* primarily uses HCO_3^- and *Pseudo-nitzschia multiseries* uses CO_2 or HCO_3^- approximately equally; only *N. navis-varingica* showed a significant decrease in the use of HCO_3^- at the lower pH associated with OA (Trimborn *et al.* 2008).

Diatom genomes encode several transporters implicated in HCO_3^- transport. In *Phaeodactylum tricornutum*, several plasmalemma-associated transporters are inducible under low CO_2 levels and are repressed at elevated CO_2, and orthologous transporter genes were reported in *Thalassiosira pseudonana* (Nakajima *et al.* 2013). There is little evidence of direct CO_2 active transport at the plasmalemma, but diatoms show external CA activity, responsible for accelerating the HCO_3^- to CO_2 conversion at the cell surface, a key component of some CCMs. This CCM is repressed when the uncatalysed CO_2 supply is greater than demand from photosynthesis, and is thus modulated by OA (see Smith-Harding *et al.* 2017 and references therein).

Similar to seaweeds, the response of diatom growth and photosynthesis to OA is also highly species dependent. Wu *et al.* (2014) and Sett *et al.* (2018) showed that elevated pCO_2 is likely to stimulate the growth rate of larger-celled diatoms more than in small-celled species because large cells are more prone to CO_2 depletion in their diffusive boundary layer. Gao and Campbell (2014) summarised the evidence in the literature for responses of diatom growth (and other physiological parameters) to OA and found that most studies reported either no effect or stimulation of growth under OA, although some did report (mostly low) inhibition of growth. Variability in response is likely due not only to species and strain differences, but also to interactions with other environmental conditions, such as light intensity (see Gao and Campbell 2014 and references therein).

The metabolic activities of algae potentially affect their internal acid–base balance. Thus, assimilation of HCO_3^- and NO_3^- will lead to generation of OH^- ions inside the cell; so, for maintenance of pH homeostasis, OH^- efflux or H^+ influx must occur. Similarly, if reduced N sources such as NH_4^+ are used, then H^+ efflux is required to maintain internal pH. Influx or efflux of OH^- or H^+ to achieve pH homeostasis requires energy, and so contributes to the maintenance energy requirements of the cell (Raven 2011).

The elevated H^+ concentration (i.e. lower pH) caused by increasing CO_2 is likely to alleviate the energy cost of acid–base regulation. As external H^+ increases, the H^+ free energy gradient between the seawater of an acidified ocean and cytosol, with pH values close to 7, will be decreased, although these energy savings are likely to be small (Raven 2011). Acidification of the cytosol associated with OA is likely to be small compared with external changes; Hervé *et al.* (2012) have shown that in *Thalassiosira weisflogii* a drop of 0.4 pH units in the medium (2.5-fold increase in H^+, set by adding acid rather than increased CO_2) is likely to result in an acidification of the cytosol of 0.17 units (1.5-fold increase in H^+). However, reduced pH may have larger implications for silicification. Although Hervé *et al.* (2012) showed only a small change in biogenic Si content per cell from pH 8.2 to pH 7.8, Milligan *et al.* (2004) have shown a fourfold in increase in Si dissolution of the frustules of *T. weisflogii* at a pCO_2 of 750 *v.* 100 ppm.

Calcifiers

Shelled molluscs

Shelled molluscs are among the most vulnerable organisms to OA, with consistently negative effects of OA measured across a wide range of physiological traits (Gazeau *et al.* 2013; Kroeker *et al.* 2013; Parker *et al.* 2013). Efforts to disentangle which components of the inorganic carbonate system affect shelled molluscs have focused primarily on net calcification and are the topic of considerable debate. Net calcification in these organisms is generally found to reduce under OA owing to a reduction in gross calcification (the amount of $CaCO_3$ that is precipitated into the shell) or an increase in gross dissolution (which can occur at the outer or inner surface of the shell). Correlations between $[CO_3^{2-}]$ or Ω and gross calcification have been made in several studies (Gazeau *et al.* 2011; Waldbusser *et al.* 2015*a*, 2015*b*; Ramesh *et al.* 2017; Li *et al.* 2018; Sanders *et al.* 2018; Thomsen *et al.* 2018), with decreasing Ω considered to make the kinetics of shell formation more energetically costly (kinetic–energetic constraint theory; Waldbusser *et al.* 2015*a*, 2015*b*, 2016). However, other studies suggest that gross calcification is controlled by the seawater $[HCO_3^-]/[H^+]$ ratio and not Ω (Cyronak *et al.* 2016*a*, 2016*b*; Thomsen *et al.* 2015, 2018; Ries *et al.* 2016; Lu *et al.* 2018). Here, $CaCO_3$ precipitation is thought to be balanced by the stimulating effect of HCO_3^- (the primary substrate for calcification in shelled molluscs) and the inhibitory effect of H^+ (which reduces the inside–outside concentration gradient of H^+, reducing the ability of shelled molluscs to remove H^+ from their calcifying fluid). During calcification, shelled molluscs transport HCO_3^- from the surrounding seawater to the extrapallial space (EPS), where inorganic $CaCO_3$ is precipitated by the reaction of Ca^{2+} and HCO_3^-. As a by-product of this reaction, H^+ ions are generated that must be removed from the EPS in order to avoid acidification of the extrapallial fluid (EPF) and allow further $CaCO_3$ precipitation to occur (Thomsen *et al.* 2015; Lu *et al.* 2018). Under OA, the seawater $[HCO_3^-]/[H^+]$ ratio becomes skewed towards $[H^+]$ (the inhibitor) as $[H^+]$ increases to a much greater extent than $[HCO_3^-]$. As a result, the inside–outside $[H^+]$ gradient shrinks, reducing the ability of shelled molluscs to remove H^+ ions from their EPS. This leads to acidosis of the EPF and inhibition of $CaCO_3$ precipitation (substrate-to-inhibitor ratio theory; Bach 2015; Thomsen *et al.* 2015, 2018; Ries *et al.* 2016; Lu *et al.* 2018).

However, gross calcification in shelled molluscs is not always negatively affected under OA. In fact, gross calcification under OA has been found to increase in some shelled mollusc species (Rodolfo-Metalpa *et al.* 2011). This phenomenon likely occurs because HCO_3^-, the primary source of carbonate used for calcification, is increased under OA. Instead, declines in net

calcification in these species are caused by an increase in gross dissolution of the outer shell surface at $\Omega < 1$ (Rodolfo-Metalpa *et al.* 2011). Such increases in gross dissolution of the outer shell surface of molluscs as Ω approaches or becomes <1 have been widely documented (Hall-Spencer *et al.* 2008; Marshall *et al.* 2008; Thomsen *et al.* 2010; Gazeau *et al.* 2013; Ries *et al.* 2016). The likelihood and degree of dissolution appears to be largely dependent on the presence and condition of an outer protective organic cover, known as the periostracum (Ries 2011; Rodolfo-Metalpa *et al.* 2011; Harvey *et al.* 2018), although the periostracum alone does not appear to prevent dissolution for all shelled mollusc species (Ries *et al.* 2016). An increase in gross dissolution under OA has also been shown to occur at the inner surface of mollusc shells (Melzner *et al.* 2011; Ramesh *et al.* 2017). Although efforts to disentangle which component of the inorganic carbonate system directly causes this effect are currently lacking, it has been suggested that increases in seawater pCO_2 cause an increase in pCO_2 in the EPF and haemolymph. As a result, Ω at the site of calcification is reduced and the inner surface of the shell is exposed to more corrosive conditions (Melzner *et al.* 2011; Ramesh *et al.* 2017).

Phytoplankton: coccolithophores

Coccolithophores are calcified photosynthetic marine members of the Phylum Haptophyta. Calcification is intracellular in coccolith-forming vesicles, followed by exocytosis of the coccoliths (Taylor *et al.* 2017). In present day air-equilibrium seawater, the dominant form of inorganic carbon entering the cells is HCO_3^- (Taylor *et al.* 2017; Liu *et al.* 2018). HCO_3^- is used for photosynthesis (generating OH^-) and calcification (consuming OH^-); because the particulate inorganic C (PIC) : particulate organic C (POC) ratio is rarely 1 : 1, acid–base regulation involves either H^+ influx or H^+ efflux (Raven and Crawfurd 2012; Taylor *et al.* 2017). Intracellular acid–base regulation is also affected by N assimilation, as discussed above (Raven 2011). In *Emiliania huxleyi*, increased CO_2 concentration eventually allows diffusive CO_2 to saturate the Form ID Rubisco, obviating the need for HCO_3^- influx (Boller *et al.* 2011; Bach *et al.* 2013; Taylor *et al.* 2017). Furthermore, with increasing concentrations of anthropogenic CO_2 in the ocean, there are variable (with experimental conditions and species) effects on growth rate and the PIC : POC ratio (Raven and Crawfurd 2012; Kottmeier *et al.* 2016). Thus, although the predicted decrease in calcification with external acidification does not always occur (Liu *et al.* 2018), the modelled effect on global calcification is an 11% decrease from the present rate by 2100 (Krumhardt *et al.* 2019). Although calcification alters the DIC system, causing OA, the decrease in coccolithophore calcification would have only a minor effect in decreasing OA. Decreased calcification decreases the ballasting that enhances the biological carbon pump (Raven 2017), although a lower PIC : POC ratio in the sinking particles means more atmospheric CO_2 burial per unit POC sinking than is the case without OA. Warming associated with OA may greatly reduce these effects (Milner *et al.* 2016).

Coralline algae and corals

Seawater carbonate chemistry affects coralline algae and corals by two main pathways: photosynthetic DIC uptake and net calcification. To date, the effects of separate components of seawater carbonate chemistry on photosynthetic rates of coralline algae are equivocal. The only two estimates of these effects found no significant responses (Cornwall *et al.* 2012; Comeau *et al.* 2018). By contrast, increasing seawater DIC can increase the photosynthetic rates of some coral species (Marubini *et al.* 2008; Comeau *et al.* 2017*b*, 2018). However, the positive effects of DIC addition on coral photosynthetic rates are only significant when DIC is elevated by several hundred micromoles per kilogram, which is much greater than the predicted increase in DIC associated with OA.

A reduction in net calcification rates with OA is generally observed with coralline algae and, to a lesser extent, corals (Noisette *et al.* 2013; Comeau *et al.* 2014; Johnson *et al.* 2014; Kornder *et al.* 2018). Attempts to disentangle the effects of individual components of the changing inorganic carbon system on coral calcification have collectively demonstrated that decreasing seawater DIC, pH and Ω (CO_3^{2-}) can reduce calcification (Marubini *et al.* 2008; Comeau *et al.* 2013, 2017*b*). However, results are inconsistent, and authors have concluded that $[CO_3^{2-}]$, $[HCO_3^-]$, Ω, $[DIC]/[H^+]$ and $[Ca^{2+}]$ may all affect calcification rates to varying extents (Gattuso *et al.* 1998; Marubini *et al.* 2003, 2008; Schneider and Erez 2006; Herfort *et al.* 2008; Jury *et al.* 2010; Comeau *et al.* 2013, 2017*b*). A stronger correlation between Ω and $[CO_3^{2-}]$ and coral calcification occurs in short-term experiments (minutes to hours), although little underlying physiology has been examined in the majority of short-term 'shock' experiments to elucidate the mechanisms responsible. Over longer time periods (months) that control for potential shock effects, seawater pH has been observed as the major controller of calcification rates, with increasing seawater DIC offsetting this somewhat for two species of coral and one crustose coralline alga (Comeau *et al.* 2018).

Jokiel (2011) proposed that reduced calcification under OA was by the 'proton flux' hypothesis, where increasing H^+ concentration gradients from internal to external reduces the capacity of organisms to export waste H^+ from the site of calcification, thereby causing reduced calcification rates. Under this hypothesis, as seawater pH declines, the gradient in H^+ between the site of calcification and seawater becomes steeper, thus reducing pH in the calcifying fluid (pH_{cf}; Ries 2011). By contrast, increasing DIC provides the inorganic carbon substrate needed for $CaCO_3$ precipitation at the internal site of calcification (Jokiel 2011). The linear correlation of the DIC : H^+ ratio with CO_3^{2-} concentrations could explain why Ω, CO_3^{2-} and HCO_3^- all appear to be correlated with faster calcification rates (Comeau *et al.* 2013; Jokiel 2013). Recent experimental evidence indicated that homeostasis of pH_{cf} was correlated with more robust responses to OA (McCulloch *et al.* 2012; Holcomb *et al.* 2015; Cornwall *et al.* 2017*a*), in part confirming this hypothesis. However, calcification rates and changes in pH_{cf} under OA are not always correlated (Comeau *et al.* 2017*a*; Venn *et al.* 2019). Studies on the mechanisms underlying the proton flux hypothesis demonstrate that seawater H^+ and DIC independently affect the chemistry within the calcifying fluid of corals, and most likely coralline algae, but that increasing seawater H^+ is the main driver of pH_{cf} and seawater DIC is the main driver of DIC in the calcifying fluid (DIC_{cf}; Comeau

et al. 2018). In addition, faster seawater velocity does not promote higher pH_{cf} for either corals or coralline algae (Comeau *et al.* 2019*a*), which was one of the major cornerstones of the proton flux hypothesis.

Although there is no biological mechanism in which Ω directly controls the internal precipitation of $CaCO_3$ in the semi-enclosed cell wall of coralline algae or the calcifying fluid of corals, dissolution of dead or exposed skeletal material still could be affected by Ω. Although proposed control of calcification by Ω is often invoked, particularly in earlier literature, there is little to no evidence of Ω affecting calcification in coralline algae and corals during long-term experiments. However, maintenance of constant Ω_{cf} is often correlated with constant calcification rates under OA. Different species of corals use specific physiological mechanisms to achieve this, either by the maintenance of constant pH_{cf}, slightly elevated DIC_{cf} or elevated $Ca^{2+}{}_{cf}$ (Schoepf *et al.* 2017; Comeau *et al.* 2018, 2019*b*; DeCarlo *et al.* 2018). The similar responses between both corals and coralline algae of vastly different evolutionary histories (Comeau *et al.* 2018, 2019*b*; Cornwall *et al.* 2018) is supportive of the general role that both DIC and H^+ play in affecting the calcification physiology of the majority of marine taxa under ocean acidification.

Cross-cutting processes: fertilisation success

The majority of marine macroorganisms reproduce by either broadcast spawning (releasing spermatozoa and eggs to the water column, where fertilisation takes place) or 'spermcasting' (in which spermatozoa are released to the water but eggs are retained for fertilisation; Bishop 1998). This process entails a chemical shift in the medium surrounding the gametes from the internal, intracellular conditions of the adult to those of the open water. Thus, seawater carbonate chemistry can have an overriding effect on invertebrate fertilisation. Available data, almost exclusively from marine invertebrates and fish, show that spermatozoa exhibit 'respiratory dilution', a process by which cellular respiration in dense sperm suspensions reduces internal cellular pH (pH_i) below 8.0 and thereby inhibits sperm metabolism and motility (Christen *et al.* 1983). Spermatozoa stored in reproductive organs of marine invertebrates typically have a pH_i of ≤7.5 and hence are quiescent, but, upon release to the surrounding seawater, begin to respire and swim at maximal rates. Classically, this process has been held to be mediated by H^+, although, recent work suggests that sperm motility may be more strongly inhibited by pH shifts induced by CO_2 rather than H^+ (Shi *et al.* 2017).

In many organisms OA will reduce the gradient between external and internal pH (e.g. Stumpp *et al.* 2012; Kelly and Hofmann 2013), thereby reducing the motility of newly released spermatozoa, and hence reducing fertilisation (e.g. Havenhand *et al.* 2008). However, fertilisation itself is a complex of several processes that includes not only sperm swimming (sperm speed, angular velocity and percentage motility), but also sperm chemoattraction to the egg (many eggs release sperm chemoattractants), sperm : egg binding, and fusion (which includes the potential for 'polyspermy'; Vogel *et al.* 1982). All these processes can be affected by OA.

The overwhelming majority of studies on the effects of OA on fertilisation success report negative, or no, response (see below), with very few positive effects reported (e.g. Caldwell *et al.* 2011). Note that these data are for marine invertebrates; to the best of our knowledge, there are no published data on how OA affects seaweed fertilisation processes, although a few studies have examined other aspects of reproduction (Olischläger *et al.* 2012; Graiff *et al.* 2017; Leal *et al.* 2017). The variability in strength of response is considerable, presumably reflecting the different habitats, environmental histories, physiologies and adaptations of the study organisms. However, choice of experimental design almost certainly has played a role. Although this is common to all experiments (e.g. Cornwall and Hurd 2016), a key issue for fertilisation experiments is that many studies have tested the effects of OA using gamete concentrations that yield >90% fertilisation in 'control' (typically present day) treatments. This greatly reduces the sensitivity of the assay (which is maximal at 50% fertilisation in controls; Marshall 2006) and precludes the detection of any positive effects of OA. Thus, we need to accept that our OA–fertilisation dataset is biased and we may have inadvertently tested only a subset of possible responses.

Notwithstanding these caveats, several interesting points emerge:

- Sperm swimming behaviour is typically negatively affected by OA (Havenhand *et al.* 2008; Shi *et al.* 2017; Falkenberg *et al.* 2019), although responses vary with exposure time (Eads *et al.* 2016), and sperm chemoattraction to eggs may be modified by OA (Foo *et al.* 2018), both of which will affect fertilisation success.
- Fertilisation success is often lower under OA (Sewell *et al.* 2014), although not always (Havenhand and Schlegel 2009), but the effects of OA are mediated by interactions with other drivers in both super- and subadditive ways (Campbell *et al.* 2014; Eads *et al.* 2016; Karelitz *et al.* 2017).
- Responses vary widely among populations and individuals. For example, independent studies on multiple populations of the Pacific oyster *Crassostrea gigas* from around the world have found negative (Parker *et al.* 2010; Barros *et al.* 2013) or no (Havenhand and Schlegel 2009; Riba *et al.* 2016) effects of OA on fertilisation. Within populations, OA can change the 'fitness-landscape' such that males that perform well under ambient conditions perform poorly under OA, and vice versa (Schlegel *et al.* 2012; Campbell *et al.* 2016; Falkenberg *et al.* 2019; Smith *et al.* 2019). This indicates strong potential for selection, although the heritability, and hence evolvability, of these traits remains to be demonstrated.

Mechanisms to explain these findings are beginning to be elucidated and indicate that effects may be occurring through changes in pH, as well as through other components of the carbonate system (Schlegel *et al.* 2015; Shi *et al.* 2017).

Conclusion

We have highlighted how OA acts as a multiple driver for marine life, with different components of the seawater carbonate system affecting different physiological processes within key organisms. Thus, the field of OA has moved from simplistic concepts (e.g. reduced pH or carbonate ion concentration will reduce calcification rates) to a mature understanding of how a changing carbonate

system affects physiological processes. Nevertheless, knowledge gaps remain for some key organisms: for coralline algae, there is high certainty that H^+ affects cellular homeostasis and calcification, but only moderate certainty on how HCO_3^- affects the same processes; for fleshy seaweeds, there is low certainty on responses to OA. Understanding the mechanistic processes by which OA affects organisms has led to much-improved predictions on how marine life will respond to OA; the challenge now is to understand how OA, itself a multiple driver, will interact with other ongoing local and global environmental changes.

Conflicts of interest

The authors declare that they have no conflicts of interest.

Declaration of funding

This work was not funded by a specific funding body. Christopher E. Cornwall was supported by a Rutherford Discovery Fellowship from The Royal Society of New Zealand Te Apārangi (RDF-VUW1701).

Acknowledgements

The authors dedicate this article to Prof. Keith Hunter, who established OA research in New Zealand and was a tremendous mentor and colleague to many of the authors. The University of Dundee is a registered Scottish charity (number 015096).

References

Armbrust, E. V. (2009). The life of diatoms in the world's oceans. *Nature* **459**, 185–192. doi:10.1038/NATURE08057

Bach, L. T. (2015). Reconsidering the role of carbonate ion concentration in calcification by marine organisms. *Biogeosciences* **12**, 4939–4951. doi:10.5194/BG-12-4939-2015

Bach, L. T., Mackinder, L., Schulz, K. G., Wheeler, G., Schroeder, D. C., Brownlee, C., and Riebesell, U. (2013). Dissecting the impact of CO_2 and pH on the mechanisms of photosynthesis and calcification in the coccolithophore *Emiliania huxleyi*. *New Phytologist* **199**(1), 121–134. doi:10.1111/NPH.12225

Barros, P., Sobral, P., Range, P., Chícharo, L., and Matias, D. (2013). Effects of sea-water acidification on fertilization and larval development of the oyster *Crassostrea gigas*. *Journal of Experimental Marine Biology and Ecology* **440**, 200–206. doi:10.1016/J.JEMBE.2012.12.014

Bishop, J. (1998). Fertilization in the sea: are the hazards of broadcast spawning avoided when free-spawned sperm fertilize retained eggs? *Proceedings of the Royal Society of London – B. Biological Sciences* **265**(1397), 725–731. doi:10.1098/RSPB.1998.0353

Boller, A. J., Thomas, P. J., Cavanaugh, C. M., and Scott, K. M. (2011). Low stable carbon isotope fractionation by coccolithophore RubisCO. *Geochimica et Cosmochimica Acta* **75**(22), 7200–7207. doi:10.1016/J.GCA.2011.08.031

Boyd, P. W., Collins, S., Dupont, S., Fabricius, K., Gattuso, J.-P., Havenhand, J., Hutchins, D. A., Riebesell, U., Rintoul, M. S., Vichi, M., Biswas, H., Ciotti, A., Gao, K., Gehlen, M., Hurd, C. L., Kurihara, H., McGraw, C. M., Navarro, J. M., Nilsson, G. E., Passow, U., and Pörtner, H.-O. (2018). Experimental strategies to assess the biological ramifications of multiple drivers of global ocean change – a review. *Global Change Biology* **24**(6), 2239–2261. doi:10.1111/GCB.14102

Britton, D., Mundy, C. N., McGraw, C. M., Revill, A. T., and Hurd, C. L. (2019). Responses of seaweeds that use CO_2 as their sole inorganic carbon source to ocean acidification: differential effects of fluctuating pH but little benefit of CO_2 enrichment. *ICES Journal of Marine Science* **12**, 12–34. doi:10.1093/ICESJMS/FSZ070

Caldwell, G. S., Fitzer, S., Gillespie, C. S., Pickavance, G., Turnbull, E., and Bentley, M. G. (2011). Ocean acidification takes sperm back in time. *Invertebrate Reproduction & Development* **55**(4), 217–221. doi:10.1080/07924259.2011.574842

Campbell, A. L., Mangan, S., Ellis, R. P., and Lewis, C. (2014). Ocean acidification increases copper toxicity to the early life history stages of the polychaete *Arenicola marina* in artificial seawater. *Environmental Science & Technology* **48**(16), 9745–9753. doi:10.1021/ES502739M

Campbell, A. L., Levitan, D. R., Hosken, D. J., and Lewis, C. (2016). Ocean acidification changes the male fitness landscape. *Scientific Reports* **6**, 31250. doi:10.1038/SREP31250

Checkley, D. M. J., Dickson, A. G., Takahashi, M., Radich, A., Eisenkolb, N., and Asch, R. (2009). Elevated CO_2 enhances otolith growth in young fish. *Science* **324**(5935), 1683–1683. doi:10.1126/SCIENCE.1169806

Christen, R., Schackmann, R., and Shapiro, B. (1983). Metabolism of sea urchin sperm. Interrelationships between intracellular pH, ATPase activity, and mitochondrial respiration. *The Journal of Biological Chemistry* **258**(9), 5392–5399.

Comeau, S., Carpenter, R. C., and Edmunds, P. J. (2013). Coral reef calcifiers buffer their responses to ocean acidification using both bicarbonate and carbonate. *Proceedings of the Royal Society of London – B. Biological Sciences* **280**(1753), 20122374. doi:10.1098/RSPB.2012.2374

Comeau, S., Edmunds, P. J., Spindel, N. B., and Carpenter, R. C. (2014). Fast coral reef calcifiers are more sensitive to ocean acidification in short-term laboratory incubations. *Limnology and Oceanography* **59**(3), 1081–1091. [Published erratum appears in *Limnology and Oceanography* **2018**, 64(3), 1422–1422]. doi:10.4319/LO.2014.59.3.1081

Comeau, S., Cornwall, C. E., and McCulloch, M. T. (2017*a*). Decoupling between the response of coral calcifying fluid pH and calcification to ocean acidification. *Scientific Reports* **7**(1), 7573. doi:10.1038/S41598-017-08003-Z

Comeau, S., Tambutté, E., Carpenter, R. C., Edmunds, P. J., Evensen, N. R., Allemand, D., Ferrier-Pagès, C., Tambutté, S., and Venn, A. A. (2017*b*). Coral calcifying fluid pH is modulated by seawater carbonate chemistry not solely seawater pH. *Proceedings of the Royal Society of London – B. Biological Sciences* **284**, 20161669. doi:10.1098/RSPB.2016.1669

Comeau, S., Cornwall, C. E., DeCarlo, T. M., Krieger, E., and McCulloch, M. T. (2018). Similar controls on calcification under ocean acidification across unrelated coral reef taxa. *Global Change Biology* **24**(10), 4857–4868. doi:10.1111/GCB.14379

Comeau, S., Cornwall, C., Pupier, C., DeCarlo, T., Alessi, C., Trehern, R., and McCulloch, M. (2019*a*). Flow-driven micro-scale pH variability affects the physiology of corals and coralline algae under ocean acidification. *Scientific Reports* **9**(1), 12829. doi:10.1038/S41598-019-49044-W

Comeau, S., Cornwall, C. E., DeCarlo, T. M., Doo, S. S., Carpenter, R. C., and McCulloch, M. T. (2019*b*). Resistance to ocean acidification in coral reef taxa is not gained by acclimatization. *Nature Climate Change* **9**(6), 477–483. doi:10.1038/S41558-019-0486-9

Cornwall, C. E., and Hurd, C. L. (2016). Experimental design in ocean acidification research: problems and solutions. *ICES Journal of Marine Science* **73**(3), 572–581. doi:10.1093/ICESJMS/FSV118

Cornwall, C. E., Hepburn, C. D., Pritchard, D. W., McGraw, C. M., Currie, K. I., Hunter, K. A., and Hurd, C. L. (2012). Carbon-use strategies in macroalgae: differential responses to lowered pH and implications for ocean acidification. *Journal of Phycology* **48**, 137–144. doi:10.1111/J.1529-8817.2011.01085.X

Cornwall, C. E., Hepburn, C. D., McGraw, C. M., Currie, K. I., Pilditch, C. A., Hunter, K. A., Boyd, P. W., and Hurd, C. L. (2013). Diurnal fluctuations in seawater pH influence the response of a calcifying macroalga to ocean acidification. *Proceedings of the Royal Society of London – B. Biological Sciences* **280**, 20132201. doi:10.1098/RSPB.2013.2201

Cornwall, C. E., Revill, A. T., and Hurd, C. L. (2015). High prevalence of diffusive uptake of CO_2 by macroalgae in a temperate subtidal

ecosystem. *Photosynthesis Research* **124**(2), 181–190. doi:10.1007/S11120-015-0114-0

Cornwall, C. E., Comeau, S., and McCulloch, M. T. (2017*a*). Coralline algae elevate pH at the site of calcification under ocean acidification. *Global Change Biology* **23**(10), 4245–4256. doi:10.1111/GCB.13673

Cornwall, C. E., Revill, A. T., Hall-Spencer, J. M., Milazzo, M., Raven, J. A., and Hurd, C. L. (2017*b*). Inorganic carbon physiology underpins macroalgal responses to elevated CO_2. *Scientific Reports* **7**, 46297. doi:10.1038/SREP46297

Cornwall, C. E., Comeau, S., DeCarlo, T. M., Moore, B., D'Alexis, Q., and McCulloch, M. T. (2018). Resistance of corals and coralline algae to ocean acidification: physiological control of calcification under natural pH variability. *Proceedings of the Royal Society of London – B. Biological Sciences* **285**(1884), 20181168. doi:10.1098/RSPB.2018.1168

Crain, C. M., Kroeker, K. J., and Halpern, S. (2008). Interactive and cumulative effects of multiple human stressors in marine systems. *Ecology Letters* **11**, 1304–1315. doi:10.1111/J.1461-0248.2008.01253.X

Cummings, V., Hewitt, J. E., Van Rooyen, A., Currie, K. I., Beard, S., Thrush, S. F., Norkko, J., Barr, N. G., Heath, P., Halliday, N. J., Sedcole, R., Gomez, A., McGraw, C. M., and Metcalf, V. (2011). Ocean acidification at high latitudes: potential effects on functioning of the Antarctic bivalve *Laternula elliptica*. *PLoS One* **6**(1), e16069. doi:10.1371/JOURNAL.PONE.0016069

Cyronak, T., Schulz, K. G., and Jokiel, P. L. (2016*a*). The Omega myth: what really drives lower calcification rates in an acidifying ocean. *ICES Journal of Marine Science* **73**(3), 558–562. doi:10.1093/ICESJMS/FSV075

Cyronak, T., Schulz, K. G., and Jokiel, P. L. (2016*b*). Response to Waldbusser *et al.* (2016) 'Calcium carbonate saturation state: on myths and this or that stories'. *ICES Journal of Marine Science* **73**(3), 569–571. doi:10.1093/ICESJMS/FSV224

DeCarlo, T. M., Comeau, S., Cornwall, C. E., and McCulloch, M. T. (2018). Coral resistance to ocean acidification linked to increased calcium at the site of calcification. *Proceedings of the Royal Society of London – B. Biological Sciences* **285**(1878), 20180564. doi:10.1098/RSPB.2018.0564

Diaz-Pulido, G., Cornwall, C. E., Gartrell, P., Hurd, C. L., and Tran, D. V. (2016). Strategies of dissolved inorganic carbon use in macroalgae across a gradient of terrestrial influence: implications for the Great Barrier Reef in the context of ocean acidification. *Coral Reefs* **35**(4), 1327–1341. doi:10.1007/S00338-016-1481-5

Dickson, A. G. (1990). Standard potential of the reaction: $AgCl(s) + 12H_2(g) = Ag(s) + HCl(aq)$, and the standard acidity constant of the ion HSO_4^- in synthetic sea water from 273.15 to 318.15 K. *The Journal of Chemical Thermodynamics* **22**(2), 113–127. doi:10.1016/0021-9614(90)90074-Z

Eads, A. R., Kennington, W. J., and Evans, J. P. (2016). Interactive effects of ocean warming and acidification on sperm motility and fertilization in the mussel *Mytilus galloprovincialis*. *Marine Ecology Progress Series* **562**, 101–111. doi:10.3354/MEPS11944

Falkenberg, L. J., Styan, C. A., and Havenhand, J. N. (2019). Sperm motility of oysters from distinct populations differs in response to ocean acidification and freshening. *Scientific Reports* **9**(1), 7970. doi:10.1038/S41598-019-44321-0

Falkowski, P. G. (1994). The role of phytoplankton photosynthesis in global biogeochemical cycles. *Photosynthesis Research* **39**(3), 235–258. doi:10.1007/BF00014586

Fernández, P. A., Roleda, M. Y., and Hurd, C. L. (2015). Effects of ocean acidification on the photosynthetic performance, carbonic anhydrase activity and growth of the giant kelp. *Macrocystis pyrifera*. *Photosynthesis Research* **124**(3), 293–304. doi:10.1007/S11120-015-0138-5

Field, C. B., Behrenfeld, M. J., Randerson, J. T., and Falkowski, P. (1998). Primary production of the biosphere: integrating terrestrial and oceanic components. *Science* **281**(5374), 237–240. doi:10.1126/SCIENCE.281.5374.237

Foo, S. A., Deaker, D., and Byrne, M. (2018). *Cherchez la femme* – impact of ocean acidification on the egg jelly coat and attractants for sperm. *The Journal of Experimental Biology* **221**(13), jeb177188. doi:10.1242/JEB.177188

Gao, K., and Campbell, D. A. (2014). Photophysiological responses of marine diatoms to elevated CO_2 and decreased pH: a review. *Functional Plant Biology* **41**(5), 449–459. doi:10.1071/FP13247

Gattuso, J.-P., Frankignoulle, M., Bourge, I., Romaine, S., and Buddemeier, R. (1998). Effect of calcium carbonate saturation of seawater on coral calcification. *Global and Planetary Change* **18**(1–2), 37–46. doi:10.1016/S0921-8181(98)00035-6

Gazeau, F., Gattuso, J.-P., Greaves, M., Elderfield, H., Peene, J., Heip, C. H., and Middelburg, J. J. (2011). Effect of carbonate chemistry alteration on the early embryonic development of the Pacific oyster (*Crassostrea gigas*). *PLoS One* **6**(8), e23010. doi:10.1371/JOURNAL.PONE.0023010

Gazeau, F., Parker, L. M., Comeau, S., Gattuso, J.-P., O'Connor, W. A., Martin, S., Pörtner, H.-O., and Ross, P. M. (2013). Impacts of ocean acidification on marine shelled molluscs. *Marine Biology* **160**(8), 2207–2245. doi:10.1007/S00227-013-2219-3

Graiff, A., Dankworth, M., Wahl, M., Karsten, U., and Bartsch, I. (2017). Seasonal variations of *Fucus vesiculosus* fertility under ocean acidification and warming in the western Baltic Sea. *Botanica Marina* **60**(3), 239–255. doi:10.1515/BOT-2016-0081

Hall-Spencer, J. M., Rodolfo-Metalpa, R., Martin, S., Ransome, E., Fine, M., Turner, S. M., Rowley, S. J., Tedesco, D., and Buia, M. C. (2008). Volcanic carbon dioxide vents show ecosystem effects of ocean acidification. *Nature* **454**(7200), 96–99. doi:10.1038/NATURE07051

Harvey, B. P., Agostini, S., Wada, S., Inaba, K., and Hall-Spencer, J. M. (2018). Dissolution: the Achilles' heel of the triton shell in an acidifying ocean. *Frontiers in Marine Science* **5**, 371. doi:10.3389/FMARS.2018.00371

Havenhand, J., and Schlegel, P. (2009). Near-future levels of ocean acidification do not affect sperm motility and fertilization kinetics in the oyster *Crassostrea gigas*. *Biogeosciences* **6**(12), 3009–3015. doi:10.5194/BG-6-3009-2009

Havenhand, J. N., Buttler, F.-R., Thorndyke, M. C., and Williamson, J. E. (2008). Near-future levels of ocean acidification reduce fertilization success in a sea urchin. *Current Biology* **18**(15), R651–R652. doi:10.1016/J.CUB.2008.06.015

Hepburn, C. D., Pritchard, D. W., Cornwall, C. E., McLeod, R. J., Beardall, J., Raven, J. A., and Hurd, C. L. (2011). Diversity of carbon use strategies in a kelp forest community: implications for a high CO_2 ocean. *Global Change Biology* **17**, 2488–2497. doi:10.1111/J.1365-2486.2011.02411.X

Herfort, L., Thake, B., and Taubner, I. (2008). Bicarbonate stimulation of calcification and photosynthesis in two hermatypic corals. *Journal of Phycology* **44**, 91–98. doi:10.1111/J.1529-8817.2007.00445.X

Hervé, V., Derr, J., Douady, S., Quinet, M., Moisan, L., and Lopez, P. J. (2012). Multiparametric analyses reveal the pH-dependence of silicon biomineralization in diatoms. *PLoS One* **7**(10), e46722. doi:10.1371/JOURNAL.PONE.0046722

Heuer, R. M., and Grosell, M. (2014). Physiological impacts of elevated carbon dioxide and ocean acidification on fish. *American Journal of Physiology – Regulatory, Integrative and Comparative Physiology* **307**(9), R1061–R1084. doi:10.1152/AJPREGU.00064.2014

Heuer, R. M., and Grosell, M. (2016). Elevated CO_2 increases energetic cost and ion movement in the marine fish intestine. *Scientific Reports* **6**, 34480. doi:10.1038/SREP34480

Heuer, R. M., Welch, M. J., Rummer, J. L., Munday, P. L., and Grosell, M. (2016). Altered brain ion gradients following compensation for elevated CO_2 are linked to behavioural alterations in a coral reef fish. *Scientific Reports* **6**, 33216. doi:10.1038/SREP33216

Hoffmann, L. J., Breitbarth, E., McGraw, C. M., Law, C. S., Currie, K. I., and Hunter, K. A. (2013). A trace-metal clean, pH-controlled incubator system for ocean acidification incubation studies. *Limnology and Oceanography, Methods* **11**, 53–61. doi:10.4319/LOM.2013.11.53

Hofmann, G. E., and Todgham, A. E. (2010). Living in the now: physiological mechanisms to tolerate a rapidly changing environment. *Annual Review of Physiology* **72**(1), 127–145. doi:10.1146/ANNUREV-PHYSIOL-021909-135900

Holcomb, M., Venn, A. A., Tambutté, S., Allemand, D., Trotter, J., and McCulloch, M. (2015). Coral calcifying fluid pH dictates response to ocean acidification. *Scientific Reports* **4**, 5207. doi:10.1038/SREP05207

Hurd, C. L., Hepburn, C. D., Currie, K. I., Raven, J. A., and Hunter, K. A. (2009). Testing methods of ocean acidification on algal metabolism: consideration for experimental designs. *Journal of Phycology* **45**, 1236–1251. doi:10.1111/J.1529-8817.2009.00768.X

Hurd, C. L., Harrison, P. J., Bischof, K., and Lobban, C. S. (2014). 'Seaweed Ecology and Physiology', 2nd edn. (Cambridge University Press: Cambridge, UK.)

Hurd, C. L., Lenton, A., Tilbrook, B., and Boyd, P. W. (2018). Current understanding and challenges for oceans in a higher-CO_2 world. *Nature Climate Change* **8**(8), 686–694. doi:10.1038/S41558-018-0211-0

Johnson, M. D., Price, N. N., and Smith, J. E. (2014). Contrasting effects of ocean acidification on tropical fleshy and calcareous algae. *PeerJ* **2**, e411. doi:10.7717/PEERJ.411

Jokiel, P. L. (2011). Ocean acidification and control of reef coral calcification by boundary layer limitation of proton flux. *Bulletin of Marine Science* **87**, 639–657. doi:10.5343/BMS.2010.1107

Jokiel, P. L. (2013). Coral reef calcification: carbonate, bicarbonate and proton flux under conditions of increasing ocean acidification. *Proceedings of the Royal Society of London – B. Biological Sciences* **280**(1764), 20130031. doi:10.1098/RSPB.2013.0031

Jury, C. P., Whitehead, R. F., and Szmant, A. M. (2010). Effects of variations in carbonate chemistry on the calcification rates of *Madracis auretenra* (= *Madracis mirabilis sensu* Wells, 1973): bicarbonate concentrations best predict calcification rates. *Global Change Biology* **16**(5), 1632–1644. doi:10.1111/J.1365-2486.2009.02057.X

Karelitz, S. E., Uthicke, S., Foo, S. A., Barker, M. F., Byrne, M., Pecorino, D., and Lamare, M. D. (2017). Ocean acidification has little effect on developmental thermal windows of echinoderms from Antarctica to the tropics. *Global Change Biology* **23**(2), 657–672. doi:10.1111/GCB.13452

Kelly, M. W., and Hofmann, G. E. (2013). Adaptation and the physiology of ocean acidification. *Functional Ecology* **27**(4), 980–990. doi:10.1111/J.1365-2435.2012.02061.X

Kornder, N. A., Riegl, B. M., and Figueiredo, J. (2018). Thresholds and drivers of coral calcification responses to climate change. *Global Change Biology* **24**(11), 5084–5095. doi:10.1111/GCB.14431

Kottmeier, D. M., Rokitta, S. D., and Rost, B. (2016). Acidification, not carbonation, is the major regulator of carbon fluxes in the coccolithophore *Emiliania huxleyi*. *New Phytologist* **211**(1), 126–137. doi:10.1111/NPH.13885

Kram, S. L., Price, N. N., Donham, E. M., Johnson, M. D., Kelly, E. L. A., Hamilton, S. L., and Smith, J. E. (2016). Variable responses of temperate calcified and fleshy macroalgae to elevated pCO_2 and warming. *ICES Journal of Marine Science* **73**(3), 693–703. doi:10.1093/ICESJMS/FSV168

Kroeker, K. J., Kordas, R. L., Crim, R., Hendriks, I. E., Ramajo, L., Singh, G. S., Duarte, C. M., and Gattuso, J. P. (2013). Impacts of ocean acidification on marine organisms: quantifying sensitivities and interaction with warming. *Global Change Biology* **19**, 1884–1896. doi:10.1111/GCB.12179

Krumhardt, K. M., Lovenduski, N. S., Long, M. C., Levy, M., Lindsay, K., Moore, J. K., and Nissen, C. (2019). Coccolithophore growth and calcification in an acidified ocean: insights from community earth system model simulations. *Journal of Advances in Modeling Earth Systems* **11**, 1418–1437. doi:10.1029/2018MS001483

Kübler, J. E., and Dudgeon, S. R. (2015). Predicting effects of ocean acidification and warming on algae lacking carbon concentrating mechanisms. *PLoS One* **10**(7), e0132806. doi:10.1371/JOURNAL.PONE.0132806

Kübler, J. E., Johnston, A. M., and Raven, J. A. (1999). The effects of reduced and elevated CO_2 and O_2 on the seaweed *Lomentaria articulata*. *Plant, Cell & Environment* **22**, 1303–1310. doi:10.1046/J.1365-3040.1999.00492.X

Leal, P. P., Hurd, C. L., Fernández, P. A., and Roleda, M. Y. (2017). Meiospore development of the kelps *Macrocystis pyrifera* and *Undaria pinnatifida* under ocean acidification and ocean warming: independent effects are more important than their interaction. *Marine Biology* **164**(1), 7. doi:10.1007/S00227-016-3039-Z

Lee, K., Kim, T.-W., Byrne, R. H., Millero, F. J., Feely, R. A., and Liu, Y.-M. (2010). The universal ratio of boron to chlorinity for the North Pacific and North Atlantic oceans. *Geochimica et Cosmochimica Acta* **74**(6), 1801–1811. doi:10.1016/J.GCA.2009.12.027

Lewis, E. and Wallace, D. (1998). Program developed for CO_2 system calculations. ORNL/CDIAC-105. Carbon Dioxide Information Analysis Center, Oak Ridge National Laboratory, US Department of Energy, Oak Ridge, TN, USA.

Li, J., Mao, Y., Jiang, Z., Zhang, J., Bian, D., and Fang, J. (2018). Impact of seawater carbonate variables on post-larval bivalve calcification. *Journal of Oceanology and Limnology* **36**(2), 405–413. doi:10.1007/S00343-017-6277-0

Liu, Y.-W., Eagle, R. A., Aciego, S. M., Gilmore, R. E., and Ries, J. B. (2018). A coastal coccolithophore maintains pH homeostasis and switches carbon sources in response to ocean acidification. *Nature Communications* **9**(1), 2857. doi:10.1038/S41467-018-04463-7

Lu, Y., Wang, L., Wang, L., Cong, Y., Yang, G., and Zhao, L. (2018). Deciphering carbon sources of mussel shell carbonate under experimental ocean acidification and warming. *Marine Environmental Research* **142**, 141–146. doi:10.1016/J.MARENVRES.2018.10.007

Lueker, T. J., Dickson, A. G., and Keeling, C. D. (2000). Ocean pCO_2 calculated from dissolved inorganic carbon, alkalinity, and equations for K_1 and K_2: validation based on laboratory measurements of CO_2 in gas and seawater at equilibrium. *Marine Chemistry* **70**(1–3), 105–119. doi:10.1016/S0304-4203(00)00022-0

Marshall, D. J. (2006). Reliably estimating the effect of toxicants on fertilization success in marine broadcast spawners. *Marine Pollution Bulletin* **52**(7), 734–738. doi:10.1016/J.MARPOLBUL.2006.05.005

Marshall, D. J., Santos, J. H., Leung, K. M., and Chak, W. H. (2008). Correlations between gastropod shell dissolution and water chemical properties in a tropical estuary. *Marine Environmental Research* **66**(4), 422–429. doi:10.1016/J.MARENVRES.2008.07.003

Marubini, F., Ferrier–Pages, C., and Cuif, J. P. (2003). Suppression of skeletal growth in scleractinian corals by decreasing ambient carbonate-ion concentration: a cross-family comparison. *Proceedings of the Royal Society of London – B. Biological Sciences* **270**(1511), 179–184. doi:10.1098/RSPB.2002.2212

Marubini, F., Ferrier-Pages, C., Furla, P., and Allemand, D. (2008). Coral calcification responds to seawater acidification: a working hypothesis towards a physiological mechanism. *Coral Reefs* **27**(3), 491–499. doi:10.1007/S00338-008-0375-6

McCulloch, M. T., Falter, J., Trotter, J., and Montagna, P. (2012). Coral resilience to ocean acidification and global warming through pH up-regulation. *Nature Climate Change* **2**, 623–627. doi:10.1038/NCLIMATE1473

McGraw, C. M., Cornwall, C. E., Reid, M. R., Currie, K. I., Hepburn, C. D., Boyd, P., Hurd, C. L., and Hunter, K. A. (2010). An automated pH-controlled culture system for laboratory-based ocean acidification experiments. *Limnology and Oceanography, Methods* **8**, 686–694.

Melzner, F., Stange, P., Trübenbach, K., Thomsen, J., Casties, I., Panknin, U., Gorb, S. N., and Gutowska, M. A. (2011). Food supply and seawater pCO_2 impact calcification and internal shell dissolution in the blue mussel *Mytilus edulis*. *PLoS One* **6**(9), e24223. doi:10.1371/JOURNAL.PONE.0024223

Milligan, A. J., Varela, D. E., Brzezinski, M. A., and Morel, F. M. M. (2004). Dynamics of silicon metabolism and silicon isotopic discrimination in a marine diatoms: a function of pCO_2. *Limnology and Oceanography* **49**(2), 322–329. doi:10.4319/LO.2004.49.2.0322

Milner, S., Langer, G., Grelaud, M., and Ziveri, P. (2016). Ocean warming modulates the effects of acidification on *Emiliania huxleyi* calcification and sinking. *Limnology and Oceanography* **61**(4), 1322–1336. doi:10.1002/LNO.10292

Nagelkerken, I., and Munday, P. L. (2016). Animal behaviour shapes the ecological effects of ocean acidification and warming: moving from individual to community-level responses. *Global Change Biology* **22**(3), 974–989. doi:10.1111/GCB.13167

Nakajima, K., Tanaka, A., and Matsuda, Y. (2013). SLC4 family transporters in a marine diatom directly pump bicarbonate from seawater. *Proceedings of the National Academy of Sciences of the United States of America* **110**(5), 1767–1772. doi:10.1073/PNAS.1216234110

Nilsson, G. E., Dixson, D. L., Domenici, P., McCormick, M. I., Sørensen, C., Watson, S. A., and Munday, P. L. (2012). Near-future carbon dioxide levels alter fish behaviour by interfering with neurotransmitter function. *Nature Climate Change* **2**, 201–204. doi:10.1038/NCLIMATE1352

Noisette, F., Egilsdottir, H., Davoult, D., and Martin, M. (2013). Physiological responses of three temperate coralline algae from contrasting habitats to near-future ocean acidification. *Journal of Experimental Marine Biology and Ecology* **448**, 179–187. doi:10.1016/J.JEMBE.2013.07.006

Olischläger, M., Bartsch, I., Gutow, L., and Wiencke, C. (2012). Effects of ocean acidification on different life-cycle stages of the kelp *Laminaria hyperborea* (Phaeophyceae). *Botanica Marina* **55**(5), 511–525. doi:10.1515/BOT-2012-0163

Parker, L. M., Ross, P. M., and O'Connor, W. A. (2010). Comparing the effect of elevated pCO_2 and temperature on the fertilization and early development of two species of oysters. *Marine Biology* **157**(11), 2435–2452. doi:10.1007/S00227-010-1508-3

Parker, L., Ross, P., O'Connor, W., Pörtner, H., Scanes, E., and Wright, J. (2013). Predicting the response of molluscs to the impact of ocean acidification. *Biology (Basel)* **2**(2), 651–692. doi:10.3390/BIOLOGY2020651

Perez, F. F., and Fraga, F. (1987). Association constant of fluoride and hydrogen ions in seawater. *Marine Chemistry* **21**(2), 161–168. doi:10.1016/0304-4203(87)90036-3

Ramesh, K., Hu, M. Y., Thomsen, J., Bleich, M., and Melzner, F. (2017). Mussel larvae modify calcifying fluid carbonate chemistry to promote calcification. *Nature Communications* **8**(1), 1709. doi:10.1038/S41467-017-01806-8

Rautenberger, R., Fernandez, P. A., Strittmatter, M., Heesch, S., Cornwall, C. E., Hurd, C. L., and Roleda, M. Y. (2015). Saturating light and not increased carbon dioxide under ocean acidification drives photosynthesis and growth in *Ulva rigida* (Chlorophyta). *Ecology and Evolution* **5**(4), 874–888. doi:10.1002/ECE3.1382

Raven, J. A. (2011). Praeger Review: effects on marine algae of changed seawater chemistry with increasing atmospheric CO_2. *Biology and Environment* **111**(1), 1–17. doi:10.3318/BIOE.2011.01

Raven, J. A. (2017). The possible roles of algae in restricting the increase in atmospheric CO_2 and global temperature. *European Journal of Phycology* **52**(4), 506–522. doi:10.1080/09670262.2017.1362593

Raven, J. A., and Crawfurd, K. (2012). Environmental controls on coccolithophore calcification. *Marine Ecology Progress Series* **470**, 137–166. doi:10.3354/MEPS09993

Raven, J. A., Beardall, J., and Giordano, M. (2014). Energy costs of carbon dioxide concentrating mechanisms in aquatic organisms. *Photosynthesis Research* **121**, 111–124. doi:10.1007/S11120-013-9962-7

Reidenbach, L. B., Fernandez, P. A., Leal, P. P., Noisette, F., McGraw, C. M., Revill, A. T., Hurd, C. L., and Kübler, J. E. (2017). Growth, ammonium metabolism, and photosynthetic properties of *Ulva australis* (Chlorophyta) under decreasing pH and ammonium enrichment. *PLoS One* **12**(11), e0188389. doi:10.1371/JOURNAL.PONE.0188389

Riba, I., Gabrielyan, B., Khosrovyan, A., Luque, A., and Del Valls, T. A. (2016). The influence of pH and waterborne metals on egg fertilization of the blue mussel (*Mytilus edulis*), the oyster (*Crassostrea gigas*) and the sea urchin (*Paracentrotus lividus*). *Environmental Science and Pollution Research International* **23**(14), 14580–14588. doi:10.1007/S11356-016-6611-7

Riebesell, U., Fabry, V. J., Hansson, L., and Gattuso, J. P. (2010). 'Guide to Best Practices for Ocean Acidification Research and Data Reporting.' (Publications Office of the European Union: Luxembourg, Luxembourg.)

Ries, J. B. (2011). Acid ocean cover up. *Nature Climate Change* **1**, 294–295. doi:10.1038/NCLIMATE1204

Ries, J. B., Ghazaleh, M. N., Connolly, B., Westfield, I., and Castillo, K. D. (2016). Impacts of seawater saturation state ($\Omega A = 0.4–4.6$) and temperature (10, 25°C) on the dissolution kinetics of whole-shell biogenic carbonates. *Geochimica et Cosmochimica Acta* **192**, 318–337. doi:10.1016/J.GCA.2016.07.001

Rodolfo-Metalpa, R., Houlbrèque, F., Tambutté, É., Boisson, F., Baggini, C., Patti, F. P., Jeffree, R., Fine, M., Foggo, A., Gattuso, J. P., and Hall-Spencer, J. M. (2011). Coral and mollusc resistance to ocean acidification adversely affected by warming. *Nature Climate Change* **1**, 308–312. doi:10.1038/NCLIMATE1200

Roggatz, C. C., Lorch, M., Hardege, J. D., and Benoit, D. M. (2016). Ocean acidification affects marine chemical communication by changing structure and function of peptide signalling molecules. *Global Change Biology* **22**(12), 3914–3926. doi:10.1111/GCB.13354

Roleda, M. Y., Boyd, P. W., and Hurd, C. L. (2012*a*). Before ocean acidification: calcifier chemistry lessons. *Journal of Phycology* **48**(4), 840–843. doi:10.1111/J.1529-8817.2012.01195.X

Roleda, M. Y., Morris, J. N., McGraw, C. M., and Hurd, C. L. (2012*b*). Ocean acidification and seaweed reproduction: increased CO_2 ameliorates the negative effect of lowered pH on meiospore germination in the giant kelp *Macrocystis pyrifera* (Laminariales, Phaeophyceae). *Global Change Biology* **18**, 854–864. doi:10.1111/J.1365-2486.2011.02594.X

Sanders, T., Schmittmann, L., Nascimento-Schulze, J. C., and Melzner, F. (2018). High calcification costs limit mussel growth at low salinity. *Frontiers in Marine Science* **5**, 352. doi:10.3389/FMARS.2018.00352

Schlegel, P., Havenhand, J. N., Gillings, M. R., and Williamson, J. E. (2012). Individual variability in reproductive success determines winners and losers under ocean acidification: a case study with sea urchins. *PLoS One* **7**(12), e53118. doi:10.1371/JOURNAL.PONE.0053118

Schlegel, P., Binet, M. T., Havenhand, J. N., Doyle, C. J., and Williamson, J. E. (2015). Ocean acidification impacts on sperm mitochondrial membrane potential bring sperm swimming behaviour near its tipping point. *The Journal of Experimental Biology* **218**(7), 1084–1090. doi:10.1242/JEB.114900

Schneider, K., and Erez, J. (2006). The effect of carbonate chemistry on calcification and photosynthesis in the hermatypic coral. *Acropora eurystoma. Limnology and Oceanography* **51**(3), 1284–1293. doi:10.4319/LO.2006.51.3.1284

Schoepf, V., Jury, C. P., Toonen, R. J., and McCulloch, M. T. (2017). Coral calcification mechanisms facilitate adaptive responses to ocean acidification. *Proceedings of the Royal Society of London – B. Biological Sciences* **284**(1868), 20172117. doi:10.1098/RSPB.2017.2117

Schunter, C., Welch, M. J., Ryu, T., Zhang, H., Berumen, M. L., Nilsson, G. E., Munday, P. L., and Ravasi, T. (2016). Molecular signatures of transgenerational response to ocean acidification in a species of reef fish. *Nature Climate Change* **6**, 1014–1018. doi:10.1038/NCLIMATE3087

Sett, S., Schulz, K. G., Bach, L. T., and Riebesell, U. (2018). Shift towards larger diatoms in a natural phytoplankton assemblage under combined high-CO_2 and warming conditions. *Journal of Plankton Research* **40**(4), 391–406. doi:10.1093/PLANKT/FBY018

Sewell, M. A., Millar, R. B., Yu, P. C., Kapsenberg, L., and Hofmann, G. E. (2014). Ocean acidification and fertilization in the Antarctic sea urchin *Sterechinus neumayeri*: the importance of polyspermy. *Environmental Science & Technology* **48**(1), 713–722. doi:10.1021/ES402815S

Shi, W., Zhao, X., Han, Y., Guo, C., Liu, S., Su, W., Wang, Y., Zha, S., Chai, X., and Fu, W. (2017). Effects of reduced pH and elevated pCO_2 on sperm motility and fertilisation success in blood clam, *Tegillarca granosa*. *New Zealand Journal of Marine and Freshwater Research* **51**(4), 543–554. doi:10.1080/00288330.2017.1296006

Smith, K. E., Byrne, M., Deaker, D., Hird, C. M., Nielson, C., Wilson-McNeal, A., and Lewis, C. (2019). Sea urchin reproductive performance in a changing ocean: poor males improve while good males worsen in response to ocean acidification. *Proceedings of the Royal Society of London – B. Biological Sciences* **286**(1907), 20190785. doi:10.1098/RSPB.2019.0785

Smith-Harding, T. J., Beardall, J., and Mitchell, J. G. (2017). The role of external carbonic anhydrase in photosynthesis during growth of the marine diatom *Chaetoceros muelleri*. *Journal of Phycology* **53**(6), 1159–1170. doi:10.1111/JPY.12572

Stumpp, M., Hu, M. Y., Melzner, F., Gutowska, M. A., Dorey, N., Himmerkus, N., Holtmann, W. C., Dupont, S. T., Thorndyke, M. C., and Bleich, M. (2012). Acidified seawater impacts sea urchin larvae pH regulatory systems relevant for calcification. *Proceedings of the National Academy of Sciences of the United States of America* **109**(44), 18192–18197. doi:10.1073/PNAS.1209174109

Taylor, A. R., Brownlee, C., and Wheeler, G. (2017). Coccolithophore cell biology: chalking up progress. *Annual Review of Marine Science* **9**(1), 283–310. doi:10.1146/ANNUREV-MARINE-122414-034032

Thomsen, J., Gutowska, M., Saphörster, J., Heinemann, A., Trübenbach, K., Fietzke, J., Hiebenthal, C., Eisenhauer, A., Körtzinger, A., and Wahl, M. (2010). Calcifying invertebrates succeed in a naturally CO_2 enriched coastal habitat but are threatened by high levels of future acidification. *Biogeosciences* **7**(11), 3879–3891. doi:10.5194/BG-7-3879-2010

Thomsen, J., Haynert, K., Wegner, K. M., and Melzner, F. (2015). Impact of seawater carbonate chemistry on the calcification of marine bivalves. *Biogeosciences* **12**(2), 1543–1571. doi:10.5194/BGD-12-1543-2015

Thomsen, J., Ramesh, K., Sanders, T., Bleich, M., and Melzner, F. (2018). Calcification in a marginal sea – influence of seawater [Ca^{2+}] and carbonate chemistry on bivalve shell formation. *Biogeosciences* **15**, 1469–1482. doi:10.5194/BG-15-1469-2018

Trimborn, S., Lundholm, N., Thoms, S., Richter, K. U., Krock, B., Hansen, P. J., and Rost, B. (2008). Inorganic carbon acquisition in potentially toxic and non-toxic diatoms: the effect of pH-induced changes in seawater carbonate chemistry. *Physiologia Plantarum* **133**, 92–105. doi:10.1111/J.1399-3054.2007.01038.X

van der Loos, L. M., Schmid, M., Leal, P. P., McGraw, C. M., Britton, D., Revill, A. T., Virtue, P., Nichols, P. D., and Hurd, C. L. (2019). Responses of macroalgae to CO_2 enrichment cannot be inferred solely from their inorganic carbon uptake strategy. *Ecology and Evolution* **9**(1), 125–140.

Velez, Z., Roggatz, C. C., Benoit, D. M., Hardege, J. D., and Hubbard, P. C. (2019). Short- and medium-term exposure to ocean acidification reduces olfactory sensitivity in gilthead seabream. *Frontiers in Physiology* **10**(731), 731. doi:10.3389/FPHYS.2019.00731

Venn, A., Tambutté, E., Caminiti-Segonds, N., Techer, N., Allemand, D., and Tambutté, S. (2019). Effects of light and darkness on pH regulation in three coral species exposed to seawater acidification. *Scientific Reports* **9**(1), 2201. doi:10.1038/S41598-018-38168-0

Vogel, H., Czihak, G., Chang, P., and Wolf, W. (1982). Fertilization kinetics of sea urchin eggs. *Mathematical Biosciences* **58**(2), 189–216. doi:10.1016/0025-5564(82)90073-6

Waldbusser, G. G., Hales, B., Langdon, C. J., Haley, B. A., Schrader, P., Brunner, E. L., Gray, M. W., Miller, C. A., and Gimenez, I. (2015*a*). Saturation-state sensitivity of marine bivalve larvae to ocean acidification. *Nature Climate Change* **5**(3), 273–280. doi:10.1038/NCLIMATE2479

Waldbusser, G. G., Hales, B., Langdon, C. J., Haley, B. A., Schrader, P., Brunner, E. L., Gray, M. W., Miller, C. A., Gimenez, I., and Hutchinson, G. (2015*b*). Ocean acidification has multiple modes of action on bivalve larvae. *PLoS One* **10**(6), e0128376. doi:10.1371/JOURNAL.PONE.0128376

Waldbusser, G. G., Hales, B., and Haley, B. A. (2016). Calcium carbonate saturation state: on myths and this or that stories. *ICES Journal of Marine Science* **73**(3), 563–568. doi:10.1093/ICESJMS/FSV174

Wilson, R. W., Millero, F. J., Taylor, J. R., Walsh, P. J., Christensen, V., Jennings, S., and Grosell, M. (2009). Contribution of fish to the marine inorganic carbon cycle. *Science* **323**(5912), 359–362. doi:10.1126/SCIENCE.1157972

Wu, Y., Campbell, D. A., Irwin, A. J., Suggett, D. J., and Finkel, Z. V. (2014). Ocean acidification enhances the growth rate of larger diatoms. *Limnology and Oceanography* **59**(3), 1027–1034. doi:10.4319/LO.2014.59.3.1027

Handling Editor: Michael Ellwood