Summary

Biotic interactions underlie life’s diversity and are the lynchpin to understanding its complexity and resilience within an ecological niche. Algal biologists have embraced this paradigm, and studies building on the explosive growth in omics and cell biology methods have facilitated the in-depth analysis of nonmodel organisms and communities from a variety of ecosystems. In turn, these advances have enabled a major revision of our understanding of the origin and evolution of photosynthesis in eukaryotes, bacterial–algal interactions, control of massive algal blooms in the ocean, and the maintenance and degradation of coral reefs. Here, we review some of the most exciting developments in the field of algal biotic interactions and identify challenges for scientists in the coming years. We foresee the development of an algal knowledgebase that integrates ecosystem-wide omics data and the development of molecular tools/resources to perform functional analyses of
individuals in isolation and in populations. These assets will allow us to move beyond mechanistic studies of a single species towards understanding the interactions amongst algae and other organisms in both the laboratory and the field.

I. Introduction

Algae are key primary producers in aquatic environments and represent several emerging genetic model systems (Armbrust et al., 2004; Hopes et al., 2016; Nymark et al., 2016). They also play an increasingly important role in human nutrition (FAO, 2014). Algal photosynthesis provides about one-half of the oxygen that we breathe, and their genomes reveal the story of a tangled past that traverses the tree of life through the processes of endosymbiosis and horizontal gene transfer (HGT) (Price et al., 2012; Cenci et al., 2017). Biotic interactions between algae and other eukaryotes (e.g. Worden et al., 2015) are extremely widespread in aquatic and terrestrial ecosystems. The degree to which nature has experimented with these relationships is wide ranging, including interactions among organisms that maintain few functional associations, to those that have evolved a highly integrated suite of functions. In addition to the intracellular interactions described below, algae also engage in extracellular/surface interactions in the phycosphere, which is the ecologically and physiologically integrated neighborhood inhabited by the alga (Bell & Mitchell, 1972). Epibiosis (surface colonization of one organism, the basibiont, by other attached organisms, epibionts) will not be covered in great detail here, but occurs on all immersed surfaces in the aquatic environment, including those of micro- and macroalgae, and is of paramount importance in the marine environment (Wahl et al., 2012). Epibiotic interactions (e.g. alga–alga, alga–bacterium, alga–virus; see below) play key roles in nutrient acquisition and recycling, metabolic flux, energy flow and developmental processes. In parallel with herbivory, epibiosis represents one of the most important interactions that can determine the fate of an alga and has been shown to shape entire marine communities (Korpinnen et al., 2007).

In this review, we focus on research that has contributed some of the most exciting insights into the ways in which biotic interactions shape algal evolution and physiology. This perspective recognizes that ‘symbiomes’ or ‘holobionts’ are important targets of study to elucidate the overall capacity of genomes to interact with the environment. Here, symbiome refers to co-localized and co-evolving (i.e. under selection) taxa comprising a given consortium, whereas holobiont includes all physically associated taxa regardless of the nature of the biotic interaction (Boucaud et al., 2013; Bordenstein & Theis, 2015; Douglas & Wetten, 2016; Tripp et al., 2017). This revolution in understanding integrative ecosystem function has largely been driven by the occurrence of technological advances in fields such as genomics, proteomics and cell biology. It is clear, however, that we are on the cusp of far greater advances, as the concept of the symbiome informs our experimental approaches. Below, we discuss prominent examples of algal biotic interactions that have been selected to illustrate the importance of these relationships in a broad range of contexts, ranging from deep evolutionary time to processes of key relevance in the current context of global climate change. The review begins with a discussion of the origin of photosynthetic organelles based on endosymbiosis, and then examines algal interactions in the coral symbioses and the threat that climate change imposes on this association. Lastly, we examine the role of bacteria in algal biology, and the arms race associated with alga–virus interactions.

II. Endosymbiosis

1. Complex biotic interactions explain plastid origin

Primary endosymbiosis in Archaeplastida

Algae originated as a consequence of primary plastid endosymbiosis, a process in which a mitochondrion-containing, single-celled eukaryote engulfed and retained a cyanobacterium that eventually became the photosynthetic organelle or plastid (Cavalier-Smith, 1982; Bhattacharya et al., 2004). The product of this c. 1.6 billion-yr-old endosymbiotic event (Yoon et al., 2004) eventually split into the three primary plastid lineages, the red algae, the glaucophyte algae and the green algae plus plants (together, the supergroup Archaeplastida) (Adl et al., 2012; Price et al., 2012). Algae from these groups were themselves frequently engulfed by other protists, giving rise to a rainbow of serially derived plastids distributed throughout the tree of life (Palmer, 2003; Gould et al., 2008) (Fig. 1a). The process of primary plastid capture has sometimes been depicted as a ‘hungry’ single-celled eukaryote engulfing a prokaryote, followed by the subsequent evolution of a functional organelle. This portrayal begs the obvious question: if the process is so simple, then why has the event been so rare, given that oceans and lakes are replete with phagotrophic protists that have been feeding on prokaryotes for hundreds of millions of years? In fact, there are only two bona fide primary endosymbioses known that gave rise to widespread organelles over the long history of eukaryotes; the event from which all plastids originated, as explained above, and a prior event that led to the evolution of mitochondria. Other more taxonomically limited cases of organelle origin are associated with the photosynthetic amoeba lineage Paulinella (see below), the nonphotosynthetic organelle of the trypanosomatids (Kostygov et al., 2016; Morales et al., 2016) and nitrogen-fixing spheroid bodies in the rhopalodiacean diatoms (Nakayama et al., 2014; Zehr et al., 2016). The rarity of primary endosymbiosis has fascinated scientists for many years and is usually attributed to the extensive innovations required for organelle establishment. These include: (1) events that lead to the protection of the nascent endosymbiont from host digestion; (2) tailoring of processes critical for the exchange of metabolites between the endosymbiont and host cell (Facchinelli & Weber, 2011); (3) the origin of an import system to move cytosolic proteins into the nascent organelle (Schleiff & Becker, 2011); (4) foreign gene acquisition through HGT and the
integration of the HGT-derived protein products into both host and newly developing organelle pathways (Cavalier-Smith, 2002; Karkar et al., 2015); and (5) movement of genes from the organelle to the host nucleus to escape Muller’s ratchet, that is, the accumulation of mutations in nonrecombining genomes (Felsenstein, 1974). Processes that would exacerbate the impact of Muller’s ratchet and make the relocation of genes from the organelle to the nuclear genome more imperative are the mutagenic effect of damaging reactive oxygen species (ROS) produced as a consequence of photosynthesis in the organelle (van Creveld et al., 2015), and as yet unexplained processes associated with greater damage of DNA in organelles than in their aerobic bacterial ancestors (Raven, 2015). Explanations of why organelle genomes are retained include the coordinated synthesis of complexes assembled in the organelle, and the regulation of transcriptional and post-transcriptional processes by the organelle redox state (van Creveld et al., 2015).

The most critical innovation listed above is the first, namely how a captured bacterial cell evades digestion by the host during the initial stages of plastid evolution. A potential answer to this question comes from recent work exploring the evolution of mitochondria. Current mitochondrial gene phylogenies indicate that this organelle originated from an anciently diverged environmental Rickettsiales-like pathogens with relatively large gene inventories (Wang & Wu, 2015; Ball et al., 2016a), whose descendants are now often found in association with protists (Martijn et al., 2015). However, these taxa are distinct from the highly specialized animal parasites with streamlined genomes, such as the typhus agent *Rickettsia prowazekii* (Zomorodipour & Andersson, 1999), which were initially proposed as the alphaproteobacterial candidates based on limited data collected over 10 yr ago (Emelyanov, 2003). The host of this mitochondrial endosymbiosis was likely to be a member of the recently discovered archaeal ‘Asgard’ superphylum (including the Lokiarchaeaota and Heimdallarchaeota), which is the most closely related prokaryote to the eukaryote nuclear lineage (Spang et al., 2015; Zaremba-Niedzwiedzka et al., 2017). Therefore, the increasingly widely accepted view is that an Asgard-like cell was infected by a relatively gene-rich Rickettsiales-like pathogen, thus laying the foundation for mitochondrial endosymbiosis and eukaryogenesis. By virtue of their existing ability to thrive in the intracellular environment, the ancestors of mitochondria were preadapted to switch from pathogenesis to endosymbiosis. These cells had evolved efficient solutions to deal with host innate immunity as a result of millions of years of co-evolution with the Asgard lineage. These findings suggest that, to become a successful proto-endosymbiont, the invading cell needs to evade host defenses, which is more likely to be achieved by an intracellular pathogen adapted to the cytosolic lifestyle (Ball et al., 2016b,c; Cenci et al., 2017).

The application of this concept to the origin of plastids requires some modification because extant cyanobacteria are not intracellular pathogens and lack the inherent capacity to evade host defenses. We suggest two possible explanations for cyanobacterial survival. First, the Archaeplastida host of this endosymbiosis may have developed mutations that reduced the efficacy of its lytic/phagocytic functions. This provided the cyanobacterium with sufficient residence time within a host food vacuole to evolve a character(s) beneficial to the host (e.g. secretion of fixed carbon or reduced nitrogen compounds), which allowed the establishment...
and spread of a founder population. This scenario is more likely to have occurred in oligotrophic waters, which lacked abundant prey. An alternative explanation is that the cyanobacterium was protected by a third ‘player’ that could withstand host defenses. This latter idea receives support from the finding that there are several dozen genes of chlamydial origin present in the nuclear genome of algae and plants (Huang & Gogarten, 2007; Becker et al., 2008). Phylogenetic data suggest that these genes are from environmental strains with relatively large genomes, such as those that infect Acanthamoeba, and not the highly reduced human pathogens. In addition, many of the products of these nucleus-encoded genes are plastid-targeted and perform specialized functions not associated with cyanobacteria (Huang & Gogarten, 2007; Moustafa et al., 2008). These observations have led to the ‘ménage à trois’ hypothesis (MATH) to explain the origin of plastids. In this scenario, a Chlamydiales ancestor evolved from a pathogenic to symbiotic lifestyle, protecting the cyanobacterium in its inclusion vesicle (Ball et al., 2013; Cenci et al., 2017). Although the MATH remains controversial, largely as a result of issues associated with ‘deep time’ gene phylogenies and the unresolved role of HGT in eukaryote evolution (Dagan et al., 2013; Ball et al., 2016a), its complexity reflects well-established biotic interactions. As illustrated in Fig. 1b, it predicts that an elementary body (chlamydial infectious particle) escapes host defenses by remodeling the phagocytic membrane and by secreting chlamydial effector proteins that enable bacterial-specific metabolites of photosynthesis, such as ADP-glucose, to enter the host cytosolic glycogen stores. Both glucophytes and red algae store carbohydrates in their cytosol, suggesting that the glycogen/starch pool may have provided an opportunity to buffer the unsynchronized demand and supply of carbon of the cyanobiont and its host. Several observations support this idea: (1) enzymes involved in the manipulation of host carbohydrate metabolism are pathogen effectors secreted by the type-III secretion system (Gehrre et al., 2016); (2) pathogenic Chlamydiae synthesize extracellular storage carbohydrates within parasitophorous vacuoles using analogous nucleotide-sugars and nucleotide-sugar transporters (Gehrre et al., 2016); (3) nucleotide-sugar transporters of host origin are evolutionary ancestors of plastid carbon exporters in red and green algae, as well as in plastids of secondary or tertiary endosymbiotic origin (Moog et al., 2015); and (4) analysis of the tryptophan biosynthesis pathway in Archaeoplastida shows that one-half (4/8) of the genes encoding proteins in this pathway are putatively of chlamydial origin, as are the Escherichia coli tyr/mtr (tyrosine/tryptophan) transporter genes (Cenci et al., 2016, 2017).

Tryptophan starvation may have been a mechanism used by the host of the primary plastid to combat chlamydial infection (Bonner et al., 2014). Tryptophan is by far the most costly amino acid for cells to synthesize. In comparison with the eukaryotic host and the cyanobiont, the sensitivity of the chlamydial symbiont to tryptophan starvation would have been exacerbated by the energy requirements for its synthesis (Bonner et al., 2014). This biotic interaction would therefore have selected for movement of the chlamydial trp operon to the cyanobacterial endosymbiont genome to ensure high levels of gene expression. Cenci et al. (2016) posit that the chlamydial trp operon transfer occurred via conjugation during the co-localization of chlamydial and cyanobacterial cells in inclusion vesicles. At a later time, some trp genes were moved to the Archaeoplastida nuclear genome by endosymbiotic gene transfer (EGT) (Martin & Herrmann, 1998) from the cyanobacterial plastid forerunner. The MATH is reinforced not only by functional considerations, but also gene numbers. Chlamydiae HGTs are not scattered randomly among the organisms of the tree of life, but, rather, an outsized contribution (c. 30–50 genes, depending on the lineage being studied) is found when compared with other noncyanobacterial prokaryotic gene acquisitions in Archaeoplastida nuclear genomes (Huang & Gogarten, 2007; Deschamps, 2014). In addition, analysis of the plastid proteome shows that, despite having >50-fold more proteobacterial than chlamydial sequences in current genome databases (e.g. National Center for Biotechnology Information), Proteobacteria and Chlamydiae genes represent the largest contribution to plastid functions (46 and 24 genes, respectively, in Arabidopsis thaliana), with only 13 from alpha-Proteobacteria (Qiu et al., 2013). The MATH provides a testable model that can be used to study the steps that led to plastid origin. Beyond its specific predictions (Ball et al., 2013; Cenci et al., 2017), this theory highlights the complexity of biotic interactions that underlie endosymbiosis. In the future, the aim should be to develop systems in the laboratory to study the processes underlying endosymbiosis, so that we can move beyond trees and diagrams to allow the experimental elucidation of mechanisms underlying organellogenesis.

Origin of the Paulinella chromatophore The concept of multiple microbes contributing to plastid evolution in the Archaeoplastida may also explain the maintenance and evolution of the plastid (termed the chromatophore) in Paulinella chromatophora (Marin et al., 2005; Nowack et al., 2008; Yoon et al., 2009). In this case, there is currently no evidence for clamydial-facilitated organelle origin. However, over 200 bacterium-derived HGTs have been found in the nuclear genome of this species that complement gene losses from the chromatophore genome. Specifically, many missing components of critical endosymbiont pathways, such as for amino acid and peptidoglycan biosynthesis and DNA replication, have been compensated for by the acquisition of a variety of prokaryotic donor genes via HGT (Nowack et al., 2016). Access to these foreign genes was probably facilitated by phagotrophic uptake of bacteria by the host amoeba, followed by HGT of DNA to the amoeba nuclear genome. Once activated, nucleus-encoded gene products were relocated to the chromatophore, possibly by trafficking through the secretory system, where they could replace components of the pathways encoded on the chromatophore genome (Nowack & Grossman, 2012). It should be stressed that a response to Muller’s ratchet acting on the chromatophore genome (leading to genome reduction) in P. chromatophora is certainly expected, but, surprisingly, it is not primarily EGT and the rerouting of host proteins in this relatively ‘young’ endosymbiosis (i.e. 90–140 million yr old;
Delaye et al., 2016) that facilitates this process, but, rather, the repurposing of environmental DNA as a result of biotic interactions.

2. Complex biotic interactions explain the symbiosis between algae and corals

Maintenance of the symbiosis Corals are the structural and trophic foundation of coral reefs, which support c. 30% of all described marine species (Wilkinson, 2004). Critically, reef-building corals are a symbiosis between the coral animal per se and photosynthetic dinoflagellates in the genus Symbiodinium (Figs 2a,b). Symbiodinium are also key algal symbionts in a wide range of coral reef animals, including sea anemone, sponges, jellyfish and clams. The coral–Symbiodinium association is one of relaxed specificity: individual corals can harbor alternative and multiple symbiont types simultaneously, and a Symbiodinium type may associate with a range of coral hosts (Silverstein et al., 2012). On acquisition of Symbiodinium by host gastrodermal cells (within which the algal cells reside), the Symbiodinium are physically separated from the cytoplasm by a host-derived vacuole known as the symbiosome (Roth et al., 1988). Exposure to competent Symbiodinium cells triggers an initial stress response in the coral Acropora digitifera, resulting in transient suppression of protein synthesis and mitochondrial metabolism (Mohamed et al., 2016). This finding supports the hypothesis that the symbiosome is a phagosome that has undergone early arrest (Shinzato et al., 2011; Mohamed et al., 2016).

Coral reefs thrive in nutrient-poor waters. In return for shelter (e.g. from ultraviolet radiation, predation), Symbiodinium photosynthesis may provide >90% of the fixed carbon requirement (Muscatine & Porter, 1977) of their hosts. A critical limitation of photosynthesis is access to dissolved inorganic carbon. As they have no direct access to ambient seawater, Symbiodinium cells depend on the host for the delivery of inorganic carbon (\(C_i\), \(CO_2\) or \(HCO_3^-\)) (see Fig. 2c). When net photosynthesis takes place, some \(C_i\) is generated via respiration, but, in corals, the predominant \(C_i\) supply to photosynthesis is its accumulation within host tissue from external sources (Shinzato et al., 2011). The concentration of \(C_i\) in the host tissue can be c. 70-fold that of seawater, which represents a steeper gradient than is observed for most organisms that use a carbon-concentrating mechanism (CCM) (Shinzato et al., 2011). The host also appears to play an active role in regulating photosynthesis in the symbionts (Barott et al., 2015; Bhattacharya et al., 2016).

The algae of the holobionts also accumulate \(C_i\) (Walker et al., 1980; Barott et al., 2015). This is probably related to the fact that dinoflagellates, such as Symbiodinium, have Form II ribulose-1,5-bisphosphate carboxylase-oxygenase (RuBisCO), which shows low \(CO_2 : O_2\) selectivity and, probably, a low affinity for CO2 (Leggat et al., 2000). Other symbioses have different roles for the animal in \(C_i\) supply to Symbiodinium; for example, in tridacnid giant clams, where the symbionts are extracellular, the hemolymph is the immediate source of \(C_i\). The \(C_i\) concentration in the hemolymph in the light is lower than that in seawater (Muscatine & Porter, 1977); thus, there is no evidence for the accumulation of \(C_i\) to higher concentrations than in seawater during influx through the gill epithelium. In this case, the accumulation of \(C_i\) by the photobiont presumably plays an even more vital role in algal primary production.

The symbiotic relationship between Symbiodinium and its coral hosts determines not only the rate of coral reef growth (calcium carbonate deposition), but also how corals respond to environmental stress (Voolstra et al., 2015). A modest episodic period of increased temperature of the ocean surface (e.g. a few days at 1–2°C above the mean summer minimum) can set off a cascade of photoinhibition, the decoupling of carbon flow between the symbiont and host (breakdown of symbiosis), oxidative damage and physical loss of symbiont cells (Wooldridge, 2013). This process, known as coral bleaching, leaves the coral host at risk of starvation, disease and death unless the symbiosis is soon re-established (Hoegh-Guldberg, 1999). In this way, algae are essential for the survival and maintenance of coral reef ecosystems. The impact of current environmental change on the health of the symbiotic association is particularly alarming, especially in recent years. For example, > 90% of the 911 reefs surveyed in 2015–2016

Fig. 2 Dinoflagellate symbionts in corals. (a) Acropora millepora and (b) A. tenuis showing tentacles associated with individual coral polyps and tissue color (with individual cells visible in A. tenuis) associated with a high abundance of Symbiodinium within the coral gastrodermis tissue layers (photo credits: Jean-Baptiste Raina). (c) Metabolic exchange and nutrient trafficking between the coral animal and its Symbiodinium symbionts and extracellular microbes. \(C_i\), inorganic carbon.
at the Great Barrier Reef (the world’s largest continuous reef system) showed signs of severe bleaching (Albright et al., 2016).

**Omnics perspective on the coral symbiosis** The cellular and molecular processes of symbiosis that are actively being explored include recognition, capture of the symbiont in the symbiosome, proliferation of symbionts in host tissue, loss of symbionts from the host tissue, and metabolic exchange and nutrient trafficking between *Symbiodinium* and the host across multiple membranes. There is much to be learned about these topics from a broader genomic and molecular evolutionary perspective. *Symbiodinium* is classified into nine clades based on phylogenetic markers, although they represent a highly divergent group of dinoflagellate species (LaJeunesse et al., 2005; Wham & LaJeunesse, 2016) and may include > 100 species capable of forming symbiotic associations with corals. Hurdles in studying the genomic and molecular aspects of the coral system include difficulties associated with the establishment of axenic cultures of the various *Symbiodinium* types, their slow growth, and the size and complexity of their genomes. The dinoflagellate nuclear genome can be massive, up to 250 Gbp in size (LaJeunesse et al., 2005; Lin, 2011), and exhibits unusual features, including noncanonical nucleotides, atypical intron–exon splice signals (Lin, 2011) and RNAs that are trans-spliced (Zhang et al., 2007). RNA editing of transcripts has been described in mitochondrial and plastid genomes (Lin, 2011; Jackson & Waller, 2013; Mungpakdee et al., 2014), whereas the plastid genome comprises distinct DNA minicircles, each containing a gene, a few genes or, in some cases, no genes (Zhang et al., 1999; Howe et al., 2008). The nuclear genomic features are set against a backdrop of gene or genome fragment duplications, and abundant noncoding repetitive elements (McEwan et al., 2008; Shoguchi et al., 2013). Three genome sequences of *Symbiodinium* from distinct clades have been published recently (Shoguchi et al., 2013; Lin et al., 2015; Aranda et al., 2016), and their estimated genome sizes of 1.1–1.5 Gbp are smaller than the earlier estimates of 3–5 Gbp (LaJeunesse et al., 2005). In addition, these genomes share little sequence similarity; that is, < 1% of total sequenced reads from *Symbiodinium kawagutii* mapped onto the genome assembly of *Symbiodinium minutum*, and vice versa (Lin et al., 2015).
results indicate a high level of genome divergence among distinct Symbiodinium clades.

Further complexity in corals comes from a three-way functional complementarity between the coral host, the dinoflagellate and the associated microbiome of bacteria and viruses (Ziegler et al., 2017). For example, the incomplete cysteine biosynthesis pathway in the coral Acropora digitifera (Shinzato et al., 2011, 2014) is compensated for by Symbiodinium (Shoguchi et al., 2013; Lin et al., 2015), whereas bacteria probably play a key role in regulating the availability of nitrogen to the coral host and algae and in resistance to thermal stress (Rädecker et al., 2015; Ziegler et al., 2017). Given the diversity of Symbiodinium species, a ‘one-reference-genome-fits-all’ assumption will not be possible for the study of the coral–dinoflagellate symbiosis and interactions, and additional genome data from the different species types/clades will be necessary. An effective approach would be to integrate multi-omics data from the coral and the associated Symbiodinium and microbiome, that is, the holobiont (Bordenstein & Theis, 2015), to tease apart the individual contributions of each component in sustaining a healthy holobiont. The availability of additional data from free-living dinoflagellates will help to address key questions, including the evolutionary events and functional innovations that lead to the transition from a free-living to a symbiotic lifestyle. At the same time, tractable laboratory model systems are being developed (Shapiro et al., 2016) that will enable the study of cellular mechanisms that underlie the response to elevated temperature and pathogens. Findings from such studies will inform strategies for the conservation of and risk mitigation for reef ecosystems.

III. Biotic interactions within the phycosphere

1. Alga–bacterium biotic interactions

Interactions between algae and bacteria are likely to be universal in the environment. Many notable examples are species-specific, such as the green seaweed Ulva mutabilis, which relies on different bacterial strains for successful morphogenesis (Spoerner et al., 2012). In the laboratory, rather than forming the typical blade- or tube-like morphology, axenic gametes of U. mutabilis develop into callus-like aggregates of undifferentiated cells with abnormal cell walls. These findings suggest the existence of chemical signaling between bacteria and the alga, and, potentially, the complementarity of metabolic pathways. Similar interactions have also been found between the bacterium Sulfibacter pseudonitzschiae and the diatom Pseudo-nitzschia multiseries (Amin et al., 2015), and bacteria have been shown to facilitate the acclimation of the brown seaweed Ectocarpus siliculosus to a freshwater environment (Dittami et al., 2016). Another striking example of this phenomenon is the ‘Jekyll-and-Hyde’ (named by the authors) relationship between Phaeobacter gallaeciensis, a biofilm-forming roseobacter, and the bloom-forming haptophyte alga Emiliania huxleyi (Seyedsayamdost et al., 2011). Under normal growth conditions, P. gallaeciensis secretes antibiotics and growth phytohormones (e.g. the auxin indole-3-acetic acid) that appear to benefit the alga. However, as the algal population ages, the bacteria shift their small molecule biosynthesis pathways to the production of algalicides, and act as E. huxleyi pathogens (Seyedsayamdost et al., 2011; Segev et al., 2016). A different type of biotic interaction involves capture and ‘farming’ of the cryptophyte alga Teleaulax amphioxeia by its host ciliate, Mesodinium rubrum, to extract nutrients from the intact alga (Qiu et al., 2016).

More general interactions are seen with bacteria that play a key role in providing micronutrients to algae. Examples are essential organic compounds, such as thiamine (vitamin B₁) and cobalamin (vitamin B₁₂). These compounds are required as enzyme cofactors, but many phytoplankton species are unable to synthesize them. Only prokaryotes (and, only then, a subset of both Eubacteria and Archaea) can synthesize cobalamin de novo (Warren et al., 2002), and levels free in the aquatic environment are generally too low to support algal growth (Sanudo-Wilhelmy et al., 2012). Direct provision of the vitamin from bacteria to algae has been demonstrated in the laboratory (Croft et al., 2005; Wagner-Döbler et al., 2010; Kazamia & Smith, 2014; Durham et al., 2015), and evidence that similar exchanges occur in the natural environment comes from correlations observed between the presence of B₁₂-producing bacteria and algal blooms (Gobler et al., 2007; Bertrand et al., 2015). There is specificity in this interaction, demonstrated by the fact that, although cyanobacteria are B₁₂ producers, they make a variant known as pseudocobalamin which is considerably less bioavailable to eukaryotic algae than cobalamin, the variant produced by many heterotrophic bacteria (Hellwell et al., 2016). Thus, provision of photosynthate from the algae may provide the signal to attract and retain cobalamin producers within the phycosphere. Similar to B₁₂, recent studies have demonstrated that bacteria can also provide either thiamine (vitamin B₁) or its precursors to phytoplankton (McRose et al., 2014; Paerl et al., 2015) and, because thiamine is also often limiting (Sanudo-Wilhelmy et al., 2012), phytoplankton blooms may similarly be limited by thiamine-producing bacteria or other microbes.

The specificity and extent of such algal–bacterial interactions in the natural environment remain to be determined, however. One exciting development that will enable a better understanding of the diverse and multifaceted ways in which algal cells interact with their biotic and abiotic environments is the explosion of metagenomics and metatranscriptomics information that is being produced by projects such as the TARA Oceans Expedition (Bork et al., 2015). Current analyses of the ‘interactome’ in the photic zone have revealed novel partnerships and unexpected factors controlling community structure (Lima-Mendez et al., 2015). Together with mechanistic examinations of algal physiology and biochemistry in laboratory conditions (e.g. Durham et al., 2015), these omics-enabled analyses will fundamentally change our views of how algal sense and survive in the current world, and how resilient they may be to fluctuating conditions wrought by climate change.

2. Host–virus arms race during algal blooms

Viral control of algal blooms Many algal species exhibit the phenomenon of ‘blooms’, for example ‘red tides’, where there is a massive increase in cell numbers over a short period, frequently as a result of changing environmental conditions, such as agricultural run-off or ocean upwelling. In some cases, these can pose threats to
human health (so-called harmful algal blooms; HABs) as a result of the toxins that are produced by the algae and/or associated bacteria (Petitpas et al., 2014). Such blooms are ephemeral events of exceptionally high primary productivity that regulate the flux of nutrients and metabolites across aquatic food webs. These large-scale events also contribute to global net primary production, one-half of which is provided by oceanic phytoplankton (Behrenfeld et al., 2006). Several key biotic interactions can control the extent and fate of phytoplankton blooms in the ocean, including top-down regulation by grazers, interactions with algidic bacteria and viral infection (Bidle, 2015). Viruses play a key role in this process because they infect many marine algal species, such as the major ‘brown tide’ alga Aureococcus anophagefferens (Moniruzzaman et al., 2016), resulting in the cessation of phytoplankton blooms. Viruses are the most abundant biological entities in the marine environment and are considered to be major ecological, evolutionary and biogeochemical drivers of marine microbial life (Suttle, 2007). Moreover, they enhance the diversity and composition of the microbial communities by facilitating HGT among their hosts.

Recent reports have highlighted a novel inventory of auxiliary metabolic genes found in the genomes of marine viruses that were previously thought to be restricted to the genomes of their hosts (Enav et al., 2014; Rosenwasser et al., 2016), with functions including photosynthesis, the pentose phosphate pathway, phosphate regulation, sulfur metabolism, polysaccharide synthesis, sphingolipid metabolism and DNA/RNA processing. These genes can expand metabolic capabilities within infected phototrophs and affect the flux of metabolites and infochemicals to the phycosphere. The viruses that infect terrestrial plants are typically small RNA viruses that possess few genes, and therefore their life cycle is tightly integrated with and dependent on the cellular processes of their host plants (Roossinck, 1997). By contrast, viruses that infect eukaryotic algae can have a high burst size (i.e. number of viruses released from each infected cell), and have genomes of 160–560 kbp that encode up to 600 proteins (Wilson et al., 2009). Thus, these viruses require substantial resources, such as fatty acids, amino acids, nucleotides and energy to facilitate replication and assembly. Nevertheless, there is still no fundamental understanding of how such large viruses rewire the metabolism of their photosynthetic host to support their unique life cycle.

Although the ecological importance of host–virus interactions is well recognized, our ability to assess their functional/ecological impact is limited to current approaches that focus mainly on the quantification of viral abundance, gene content and diversity (Brum & Sullivan, 2015). The development of laboratory-based model systems for ecologically relevant algal–virus interactions, coupled with a molecular toolbox and genomic and post-genomics resources (Fig. 3), have deepened our mechanistic understanding of these interactions and their ecological impact (Read et al., 2013).

Emiliania huxleyi–EhV – an important host–pathogen model system The cosmopolitan coccolithophore E. huxleyi is a unicellular alga that forms massive oceanic blooms covering thousands of square kilometers (Tyrrell & Merico, 2004). The intricate calcite exoskeleton of E. huxleyi accounts for approximately one-third of total marine CaCO3 production (Monteiro et al., 2016). Emiliania huxleyi is also a major producer of dimethyl sulfide (DMS), a bioactive gas with a significant climate-regulating role that enhances cloud formation (Alcolombri et al., 2015). Therefore, biotic interactions that regulate the fate of these blooms play a profound role in determining atmospheric conditions and nutrient cycling in the ocean. Annual E. huxleyi spring blooms are frequently terminated by infection with a specific large dsDNA virus (EhV) (Schroeder et al., 2002) that belongs to the Coccolithoviruses group within the monophyletic Phycodnaviridae, a family of nucleocytoplasmic large DNA viruses. This model host–virus interaction spans >10 orders of spatial magnitude, from the individual cell (c. 10^-6 m) to mesoscale oceanic eddies (c. 10^5 m) (Lehahn et al., 2014). The system is physiologically well characterized and there is a wealth of genomic information from the alga (Read et al., 2013) and for specific viral strains with different degrees of virulence. Analysis of the EhV genome revealed a cluster of putative sphingolipid biosynthetic genes (Wilson et al., 2005). The production of glycosphingolipids is strongly induced during viral infection. These lipids are major constituents of EhV membranes and can induce host programmed cell death (PCD) during lytic infection in cultures and during natural blooms (Vardi et al., 2012). Indeed, during lytic infection, EhV triggers hallmark PCD responses, including the production of ROS (Vardi et al., 2012; Sheyn et al., 2016), the induction of caspase activity, metacaspase expression and compromised membrane integrity (Bidle et al., 2007). Viral infection also induces remodeling of the host antioxidant gene network and redox metabolism through the co-induction of glutathione and H2O2 synthesis, both essential for successful viral replication (Sheyn et al., 2016). Viral infection ‘engineers’ the sphingolipid metabolism of the host by causing downregulation of host sphingolipid biosynthesis genes, whereas the viral genes are highly upregulated (Rosenwasser et al., 2014), resulting in altered substrate specificity of serine palmitoyl-CoA transferase activity (Ziv et al., 2016). The viral enzymes have different substrate specificities from those of the host and regulate the production of virus-specific glycosphingolipids composed of unusual hydroxylated C17 sphingoid-bases (t17:0) (Ziv et al., 2016). These virus-specific sphingolipids are essential for assembly and infectivity by the virion. Combined transcriptomic and metabolomic analyses over the course of an E. huxleyi viral infection revealed major, rapid transcriptome remodeling that elicited elevated de novo fatty acid synthesis to support viral assembly and a high demand for viral internal lipid membranes (Rosenwasser et al., 2014). Remodeling of lipid metabolism was mediated by the accumulation of distinct lipid droplets containing highly saturated triacylglycerols (TAGs) (Malitsky et al., 2016). Stored TAGs may serve as energy and lipid reservoirs that are catabolized for viral assembly during later stages of infection. These approaches, which involved rigorous quantification of the rewired metabolism during algal–virus interactions, have provided fundamental insights into the strategies employed during their biochemical ‘arms race’. The identification of the specific metabolites synthesized during these interactions may yield biomarkers for sensitive detection of active viral infection in the marine environment (Vardi et al., 2009).

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IV. Future prospects

As described in this review, there has been significant progress in studies of the algal symbiome that stress the primacy of biotic factors in algal growth and productivity in the environment. These analyses have provided significant mechanistic insight into emerging systems across the algal tree of life. Nevertheless, there still remain many gaps in our knowledge and approaches. For example, most studies at the functional level have focused on ‘pairs’, such as bacteria–microalgal or viruses–microalgal, whereas these are likely to be much more complex in the natural environment. Similarly, although studies of microbial communities during annually reoccurring phytoplankton blooms provide clues about microalgal–bacteria interactions at the community level and in relation to changing environmental conditions, including those driven by global change (e.g. Needham & Fuhrman, 2016), few address specific interactions. These specific interactions are important because short-term fluctuations of environmental parameters (e.g. diurnal fluctuations) may be buffered by biotic interactions and are therefore invisible to the investigator, which would lead to the conclusion that they are not important, even though they might have an impact over a longer time scale. Furthermore, most environmental studies do not look beyond correlations based on co-occurrence networks, which, although providing useful preliminary data on who interacts with whom, do not provide insights into the biological processes that orchestrate these interactions.

To tackle these challenges, future studies should include detailed biochemical analyses of metabolites in both environmental samples in situ and under controlled laboratory conditions, using either natural or synthetic communities. The combined analyses of natural and synthetic communities and the use of microbial mutants that impact specific pathways will help to determine the activities associated with ecosystem function. Genome editing applied to model microalgae and bacteria, in combination with biochemical analyses of processes that govern their interactions, will provide a step change in understanding the significance of these interactions in relation to abiotic drivers of biological diversity, such as temperature, nutrients, seasonality and solar irradiance. By studying communities across global-scale environmental gradients, such as coastal–open sea, surface–deep ocean or polar–tropics, it should be possible to identify commonalities between taxonomically distinct, yet functionally equivalent, communities.

Finally, metagenomics data are of vital importance to this field, but need to be combined with functional studies. We are now presented with an overwhelming amount of genomics and meta data, and the time has come to start ferreting out the biological ‘meaning’ of this information using algal model systems, genetic tools and functional genomics to understand gene function and cellular mechanisms and to connect these insights with in-depth studies of physiology, metabolism and life cycle phenotypes. The addition of the new dimension of single-cell analysis is another emerging area that will probably fundamentally change how we interpret algal diversity, behavior and acclimation strategies. With these integrative approaches, we may even be able to provide key insights into how global change not only impacts the diversity of specific taxa, but the complex interacting communities of species in the ocean that underpin marine ecosystem services responsible for the health and well-being of human societies.

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