



Microalgae-bacteria symbiosis in microalgal growth and biofuel production: a review

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Abstract

Photosynthetic microalgae can capture solar energy and convert it to bio-energy and bio-chemical products. In nature or industrial processes, microalgae live together with bacterial communities and may maintain symbiotic relationships. In general interactions, microalgae exude dissolved organic carbon that becomes available to bacteria. In return, the bacteria re-mineralize sulfur, nitrogen and phosphorous to support the further growth of microalgae. In specific interactions, heterotrophic bacteria supply B vitamins as organic cofactors or produce siderophores to bind iron, which could be utilized by microalgae, while the algae supply fixed carbon to the bacteria in return. In this review, we focus on mutualistic relationship between microalgae and bacteria, summarizing recent studies on the mechanisms involved in microalgae-bacteria symbiosis. Symbiotic bacteria on promoting microalgal growth are described and the relevance of microalgae-bacteria interactions for biofuel production processes is discussed. Symbiotic microalgae-bacteria consortia could be utilized to improve microalgal biomass production and to enrich the biomass with valuable chemical and energy compounds. The suitable control of such biological interactions between microalgae and bacteria will help to improve the microalgae-based biomass and biofuel production in the future.

Key words: Microalgae, bacteria, symbiosis, growth, biofuel

Introduction

Society's reliance on fossil fuels for this energy represents one of the major challenges to global environment sustainability and economic stability (Donohue & Cogdell, 2006). Photosynthetic microalgae can capture solar energy and convert it to bio-energy and bio-chemical products, without damaging the environment or disrupting food supply (Rittmann, 2008). Microalgae are attractive targets for industrial application because of their

relatively higher conversion rate of solar energy into biomass than terrestrial plants. In addition, microalgae can grow in saline or hypersaline environments, metabolize industrial waste streams such as CO₂ flue gases and further synthesize a range of diverse bio-chemicals (Barclay *et al.*, 2013). If the metabolism of photosynthetic microalgae is directed to generate high-energy chemicals such as alcohols, alkanes and fats, they could serve as feedstock for the bio-energy industries (Donohue & Cogdell, 2006).

In nature or industrial processes, there is evidence of microalgae and bacteria living together in complex microbial communities. In many cases, these microbial communities perform concerted activities that would not be possible in the absence of partners (Donohue & Cogdell, 2006). For example, many bacteria are known to influence the development of algal blooms in nature (Tang *et al.*, 2010); the biodegradable organic matter is removed by microalgae-bacteria consortia in wastewater treatment ponds (Subashchandrabose *et al.*, 2011); many hazardous pollutants such as polycyclic aromatic hydrocarbons, phenolics, and organic solvents are remediated by algal-bacteria interactions (Munoz & Guieysse, 2006). Obviously, interactions between microalgae and bacteria exist in natural habitats, and their disruption may be responsible for the failure of microalgae isolation in the laboratory (Santos & Reis, 2014). On the other hand, many algal culture collections, intentionally or not, maintain the symbiotic relationship between algal isolate and associated bacteria, so that an algal isolate often contains one or more species of bacteria (Park *et al.*, 2008; Baggesen *et al.*, 2014). Many of these bacteria, at least those described, are of the same genera as those found in natural algal environments (Baggesen *et al.*, 2014). Microalgae and bacteria synergistically affect each other's physiology and metabolism, although bacteria have often been considered as contamination in the algae culture. In the past few years, the perception has changed and the algae-bacteria interactions are considered promising for biotechnology, as many recent studies have shown a positive effect of algae-bacteria symbiosis on algal growth and flocculation processes, which are the essential

steps in algal biotechnology (Fuentes *et al.*, 2016). Algae–bacteria interactions cover the whole range of symbiotic relationships, which are mainly identified as mutualism, commensalism and parasitism (Ramanan *et al.*, 2015). In this review, we focus on the mutualistic relationship between microalgae and bacteria. We review both general and specific interactions between microalgae and bacteria, summarizing recent studies on the mechanisms involved in microalgae-bacteria symbiosis. Symbiotic bacteria involved in promoting microalgae growth are described and the relevance of microalgae-bacteria symbiosis for biofuel production processes is reviewed. The vision is that improved knowledge, ultimately facilitating the control of microalgae-bacteria symbioses, will help to improve the microalgal-based biomass and biofuel production in the future.

Microalgae-bacteria symbiosis mechanism

In aquatic systems, autotrophic phytoplankton can fix carbon in the form of CO₂, through the Calvin cycle, the formed organic carbon molecules are used for algal growth. Some of this carbon is eventually leaked into the surrounding water as dissolved organic matter (DOM), which includes dissolved organic carbon (DOC), dissolved organic nitrogen (DON), and dissolved organic phosphorous (DOP) (Buchan *et al.*, 2014). Fig. 1 shows a simplified diagram of general and specific interactions between photosynthetic microalgae and symbiotic bacteria. In general interactions, phytoplankton exudes DOM as sources of carbon, sulfur, nitrogen or phosphorus for bacteria. In return, bacteria re-mineralize these organic nutrients into inorganic forms and support the further growth of microalgae (Cole 1982, Buchan *et al.* 2014). In specific interactions, the bacteria supply B vitamins as organic cofactors to algae, while the algae supply the fixed carbon as DOC to bacteria in return (Croft *et al.*, 2005). Siderophores produced by some bacteria could bind iron that in this way become bio-available to microalgae, which is another aspect for microalgae-bacteria specific interaction through a “carbon for iron mutualism” (Amin *et al.*, 2009).

Dissolved organic carbon

Most algae-bacteria interactions are based on microbial degradation and transformation of organic matter produced by phytoplankton (Grossart *et al.*, 2005). One carbon source supplied for bacteria is in the form of extracellular polymeric substance (EPS), which is exuded by microalgae and especially from diatoms. EPS are metabolic products accumulating on the microbial cell surface to provide protection to the cells (Mishra *et al.*, 2011). During starvation, EPS could serve as carbon or energy reserves for microalgal cells (Mishra *et al.*, 2011). The secretion of EPS from diatoms, usually as polysaccharides of uronic or pyruvic acids, depends on the presence of individual bacteria, since the soluble substances produced from bacteria have an impact on diatom physiology (Borowitzka & Moheimani, 2013). Another specific source of DOC comes from microalgae is in the form of glycolate, released by microalgal cells. Glycolate is a small 2-carbon, water-soluble molecule produced by photoautotrophs as a by-product of photorespiration (Amin *et al.*, 2012b). Some bacteria contain a glycolate utilization gene, *gldD*, and therefore incorporate glycolate for cellular growth (Lau & Armbrust 2006). Specific bacterial species possessing the *gldD* gene could therefore benefit from association with glycolate-releasing phytoplankton (Lau & Armbrust 2006).

Nitrogen, phosphorus and sulfur

Symbioses between nitrogen (N₂) fixing prokaryotes and photosynthetic eukaryotes are important for nitrogen acquisition in nitrogen-limited environments (Thompson *et al.*, 2012). Some bacteria and archaea can fix nitrogen into biologically available ammonium for microalgae in the ocean (Canfield *et al.*, 2010). Many diatoms inhabit low-nutrient waters of the open ocean and live in close association with autotrophic nitrogen-fixing bacteria (cyanobacteria) (Thompson *et al.*, 2012). In the oceans, some of these associations are believed to be mutualistic, where nitrogen-fixing cyanobacterial symbionts provide nitrogen for the diatoms; in return, the bacteria benefit

from DOM released by the diatoms (Carpenter & Foster 2003, Foster *et al.*, 2011). Nitrogen-fixing cyanobacteria have been shown to supply fixed nitrogen to diatoms, likely in the form of ammonia or dissolved organic nitrogen (DON) (Foster *et al.*, 2011). The exchange of nitrogen between diatom and cyanobacterial symbionts is likely the primary interaction between these cell types, however, it is likely that other benefits such as metabolites, vitamins, and trace elements are also exchanged between these partners (Thompson *et al.*, 2012, 2013).

Phosphorus is required by all living organisms to make DNA, RNA, ATP (energy molecules) and other essential organic compounds (Ruttenberg, 2001). Phosphorus demand is especially high for the growth of phytoplankton. In the ocean, the most abundant phosphorus exists in the form of phosphate (PO_4^{3-}) ions, which phytoplankton can utilize for growth. Under high phosphate conditions, a fraction of phosphorus assimilated by phytoplankton is released as dissolved organic phosphorus (DOP) to the seawater. Bacteria hydrolyse this organic phosphorus back to phosphate, and thus supply P to microalgae for further growth. However, under low phosphate conditions, phytoplankton and other marine organisms will utilize DOP by releasing phosphate from organic molecules using alkaline phosphatase (Yong *et al.*, 2014).

Some phytoplankton produce large amounts of the organic sulfur compound dimethylsulfoniopropionate (DMSP), which can be converted to dimethyl sulfide (DMS) by marine bacteria (Marlin, 2006). DMSP is produced in abundance by phytoplankton species such as in prymnesiophytes and dinoflagellates (Marlin, 2006). The breakdown of DMSP by marine bacteria is significant since it is a major source of organic sulfur in the world's ocean (Howard *et al.*, 2006). DMSP can be converted to DMS particularly by members of the *Roseobacter* clade, which could use DMSP as a sulfur resource (Geng and Belas, 2010, Azam & Malfatti, 2007). The *Roseobacter* can further synthesize a biological active secondary metabolite, tropodithietic acid (TDA),

which has antibacterial properties and prevents algicidal bacterial species from harming the phytoplankton (Brinkhoff *et al.*, 2004). Under this condition, the healthy algal host provides DMSP as an attachment surface for the *Roseobacter*, while the bacterial symbiont provides the antibacterial compound to the microalgae, thus forming a mutualistic symbioses relationship between the partners (Seyedsayamdost *et al.*, 2011)

Vitamin B and siderophores

Many culture- and field- based studies showed evidences of impact of available vitamins on phytoplankton growth and microbial community composition (Droop, 2007). The majority of eukaryotic phytoplankton is vitamin B auxotroph since they require exogenous B vitamins to grow. Many studies have identified algal species that require different combinations of three B vitamins: vitamin B₁₂ (cobalamin), vitamin B₁ (thiamine) and vitamin B₇ (biotin) (Croft *et al.*, 2005). Cobalamin is by far the most studied vitamin in relation to algal requirements. Vitamin B₁₂ is required in organisms lacking cobalamin-independent methionine synthase, Met E (Amin *et al.*, 2012b). Many algae either lack the synthetic pathways to produce cobalamin or utilize alternative cobalamin-independent pathways to bypass the need for the vitamin (Cruz-Lopez *et al.*, 2016). Thiamine was the first vitamin found to be an algal growth factor and acts as cofactor for a number of enzymes, which are involved in primary carbohydrate and branched chain amino acid metabolism (Croft *et al.*, 2005). Biotin is a cofactor for several essential carboxylase enzymes, including acetyl coenzyme A carboxylase, which is involved in fatty acid synthesis (Croft *et al.*, 2005). Each of these vitamins is synthesized in bacteria and may serve as part of synergistic interactions with microalgae. Available genomic sequences indicate the evidence of bacteria, archaea, and marine cyanobacteria as potential vitamin producers (Santos & Reis, 2014). In 306 species of microalgae investigated, more than half of the algal species was found to require cobalamin, 22% of the species required thiamine and 5% of the species required biotin (Croft *et al.*, 2005). About 52% of the surveyed heterokonts were unable to grow in

B₁₂-deficient medium, indicating the importance of vitamin B₁₂ for diatoms (Croft *et al.*, 2005). A more recent investigation on 332 microalgal species showed that 54% of the algal species required cobalamin, 27% of the species required thiamine, and 8% of the species required biotin (Tang *et al.*, 2010).

Iron is an essential element for photosynthesis and respiration in microalgae, whose growth is often limited due to the poor solubility and exceedingly low concentration of iron in the ocean (Tortell *et al.*, 1999). To alleviate the limitation of this key micro-nutrient, many marine heterotrophic bacteria produce siderophores, the small organic molecules that tightly bind to iron and thereby increase its solubility (Vraspir *et al.*, 2009). The heterotrophic bacteria can then take up siderophore-bound iron via outer-membrane transporters that are specific for different groups of siderophores (Amin *et al.*, 2009). Microalgae were neither found to produce siderophores nor to carry genes involved in siderophore biosynthesis in their genomes. They are, however, able to access iron from strong chelates complexed by the siderophores in the seawater (Hopkinson & Morel 2009). Rather than direct internalization of siderophores and other iron chelates, phytoplankton primarily use uptake pathways involving a reduction step to free bound iron, which is closely coupled with iron transport into the cell (Hopkinson & Morel 2009). Genomic evidences in both diatoms and green algae showed that they could via ferrireductases and adjacent Fe(II) transporters to access iron from siderophores or other chelates (Kustka *et al.*, 2007).

Symbiotic bacteria in microalgal growth promotion

Many microalgal cultures are maintained in co-culture with bacteria in the culture collection. In many cases, the bacteria living in the algal culture were found to stimulate or promote microalgal growth through different symbiosis factors. Table 1 lists a number of symbiotic bacteria associated with microalgal growth promotion.

Halomonas species isolated from xenic algal culture were found to support the growth of both *Amphidinium*

operculatum and *Porphyridium purpureum*, by synthesizing vitamin B₁₂ de novo to the same extent as exogenous vitamin B₁₂ (Croft *et al.*, 2005). The bacterium *Halomonas* sp. showed increased production of vitamin B₁₂ when the culture was provided with fucidin, a commercial algal extract, suggesting the exchange of vitamin for fixed carbon in the bacteria (Croft *et al.*, 2005). Cruz-Lopez (*et al.*, 2016) showed that both vitamin B₁ and B₁₂ required by a marine dinoflagellate *Lingulodinium polyedrum* can be provided by its associated bacterial community in the same culture. One heterotrophic bacterium *Mesorhizobium* sp. was found to support the growth of another vitamin B₁₂-dependent green alga, *Lobomonas rostrata* (Kazamia *et al.*, 2012). *Chlamydomonas reinhardtii* does not require vitamin B₁₂ for growth, because it encodes a B₁₂-independent methionine synthase (MetE). Co-culturing of *C. reinhardtii* with *Mesorhizobium* sp. however results in reduced MetE expression, demonstrating that the bacterium can deliver vitamin B₁₂ to B₁₂-independent algae as well (Kazamia *et al.*, 2012).

Keshtachere-Liebson (*et al.*, 1995) reported for the first time that a halophilic and oligotrophic bacterium, *Halomonas* sp., improves the availability of iron to an alga called *Dunaliella bardawil*. The *Halomonas* sp. was found to produce siderophores and increase the solubility of Fe, thereby making it more available to algae and facilitating their growth under Fe-deficient conditions. Amin (*et al.*, 2009; 2012) reported an algal-associated, heterotrophic bacterium belonging to the genus *Marinobacter*, which could release a siderophore called vibrioferrin to chelates Fe(III). Fe(III) was later assimilated by both the siderophores-producing bacterium and its algal partner (Amin *et al.*, 2009). A representative dinoflagellate alga, *Scrippsiella trchoidea*, could utilize iron from Fe(III)-vibrioferrin chelates released by *Marinobacter* sp. in the dark (Amin *et al.*, 2009; 2012a). In exchange, the algal cells produced dissolved organic matter to support bacterial growth and ultimately fueled the bio-synthesis through a “carbon for iron mutualism”. Baggesen (*et al.*, 2014) isolated three bacterial species, *Marinobacter* sp., *Halomonas* sp. and *Pelagibaca* sp. from one xenic culture of *Dunaliella salina*. These bacteria

were found to promote the growth of *Dunaliella salina* under iron-limited conditions. The physical contact between algae and bacteria was found not required for this growth promoting effect. All three bacterial strains were able to produce siderophores, which could facilitate iron uptake in *Dunaliella salina* (Baggesen *et al.*, 2014). The presence of a group of bacteria, *Alteromonas sp.* SY007 and *Muricauda sp.*, enhanced the assimilation of ammonium and thereby promote the growth of *Dunaliella sp.*, under nitrogen-limited condition (Chevanton *et al.*, 2013). These symbiotic bacteria could be considered as helpers for iron or nitrogen assimilation in *Dunaliella sp.* cells.

Some microalgae exhibit close physical and physiological relationships with symbiotic bacteria. The *Roseobacter* clade of marine bacteria is often found associated with dinoflagellates as one of the major producers of DMSP (Geng and Belas, 2010). One member of the *Roseobacter* clade, *Silicibacter sp.*, developed biofilm on the surface of the dinoflagellate *Pfiesteria piscicida* (Alavi *et al.*, 2001). The axenic dinoflagellate fails to grow and ultimately dies without the presence of associated bacteria; adding back the bacterium could restore the growth of *P. piscicida* (Alavi *et al.*, 2001). Another study revealed that an algal-produced senescence signal from *Emiliania huxleyi* could elicit the production of roseobactin by its bacterial symbiont in the *Roseobacter* clade, *Phaeobacter gallaeciensis* (Seyedsayamdost *et al.*, 2011). The algal host provides the bacteria with a solid surface for biofilm formation, in return, the bacteria produce antibiotics to protect the algal host from pathogen invasion, which is beneficial to both algal and bacterial growth. The diatom, *Phaeodactylum tricoratum*, secretes EPS and thereby promotes biofilm formation for *Alphaproteobacterium sp.* on its surface, in return, both bacterial culture or the spent medium could promote growth of this diatom (Bruckner *et al.*, 2011).

Microalgal-bacteria symbiosis in biofuel production

Microalgae are capable of synthesizing a range of biofuels as lipids and carbohydrates represent the major energy storage molecules in the microalgae. In contrast, proteins in microalgae are generally not considered as substrates for biofuel production, but rather for both food and feed use in human and animal nutrition (Hayes et al. 2017). The stored carbohydrates in microalgal cells may produce hydrogen while re-direction of photosynthate to lipids may be converted into diesel fuels (Beer *et al.*, 2009). The residual biomass of microalgae could be further fermented to ethanol or biogas by yeasts or anaerobic bacteria (Zhu 2013). Table 2 lists both symbiotic bacteria and fermentative bacteria involved in microalgal biofuel production. As illustrated in fig. 2, microalgal-bacteria-based growth systems could be explored for microalgal biomass and biofuel production.

Biohydrogen

Microalgal-based hydrogen production represents a novel combination of fermentative and photolytic hydrogen-generating processes. It is well known that hydrogen production by microalgae depends on a hydrogenase enzyme activity that is highly sensitive to oxygen (Melis & Happe, 2001). Therefore, strict anaerobic conditions are necessary for efficient production of hydrogen by microalgae. The bacterial symbionts, *Brevundimonas* sp., *Rhodococcus* sp., and *Leifsonia* sp., were found to enhance the hydrogen production in the microalga *Chlamydomonas* (Lakatos *et al.*, 2014). This is due to oxygen elimination by efficient bacterial respiration, which is essential for the activation of a Fe-dependent hydrogenase in *Chlamydomonas* (Lakatos *et al.*, 2014). This phenomenon was not limited to natural associations between microalgae and bacteria, since it can also be achieved by artificial algal-bacterial communities. The highest hydrogen yield was obtained when a hydrogenase deficient *Escherichia coli* was utilized as an artificial symbiotic bacterium to *Chlamydomonas* (Lakatos *et al.*, 2014). Hence, with the help of bacteria that consume the oxygen evolved, the algae can capture

light energy and produce hydrogen simultaneously without further manipulation of the system. The solar energy can also be converted into electricity by synergistic cooperation between photosynthetic microalgae and heterotrophic bacteria in the form of microalgal fuel cells (MFC), without external input of exogenous organics or nutrients (He *et al.*, 2009).

Biodiesel

Microalgal produced triacylglycerol (TAG) and other lipids can be transesterified into fatty acid methyl esters (FAMES) and are of substantial interest as biodiesel precursors (Scott *et al.*, 2010). Many studies have screened and identified microalgal species for bio-diesel production based on their high lipid contents (Griffiths *et al.*, 2011). *Chlorella vulgaris* was identified as of high lipid contents and considered as major species in the scale-up of algal biodiesel production (Griffiths *et al.*, 2011, Chisti, 2013). Different species of *Pseudomonas* were associated with *C. vulgaris* (Sapp *et al.* 2007; Guo & Tong, 2013). The correlation between *Pseudomonas* populations with *C. vulgaris* throughout the open pond growth may suggest the symbiotic association (Bell *et al.*, 2016). Co-culturing of the green microalga, *Auxenochlorella protothecoides*, with *E. coli* led to 2–6 fold increase in algal growth and a doubling of the neutral lipid content as compared to axenic growth, which may also indicate a symbiotic relationship between the partners (Higgins & VanderGheynsta, 2014). A later study revealed that a positive effect may be traced to *E. coli*'s provision of thiamine derivatives and degradation products to *A. protothecoides* (Higgins *et al.* 2016). The marine microalga *Tetraselmis striata* was selected as a candidate for biodiesel production due to its high lipids content and fast growth (Chisti, 2007). Both bacterial strains, *Pelagibaca bermudensis* and *Stappia* sp., isolated from mass cultivation of *T. striata*, showed growth promoting effects on this microalgae (Park *et al.*, 2017). These microalgae symbionts could be used for industrial biodiesel production.

The residual biomass of microalgae after collection or extraction of useful products can be further fermented to ethanol or converted to biogas. The biochemical composition of microalgae grown under normal conditions, i.e. without nutrient limitation, is approximately composed of 30–50% proteins, 20–40% carbohydrates and 8–15% lipids (Hu, 2004). Moreover, microalgae can produce up to 40% dry weight of starch granules, which various bacteria can utilize to produce ethanol (Ramanan *et al.* 2015). Saccharification of marine microalgae as proven to produce ethanol by utilizing amylase from the marine bacterium *Pseudoalteromonas undina* (Matsumoto *et al.*, 2003). Enzymatic hydrolysis and fermentation of the microalga *Chlamydomonas reinhardtii* could be achieved with amylase from the bacterium *Bacillus licheniformis* followed by fermentation of a brewer's yeast *Saccharomyces cerevisiae* (Silva *et al.*, 2016). Bio-ethanol production from algal-bacterial co-cultures is however a new area yet to be fully explored for biotechnological application.

Biogas can be produced from microalgal residue biomass by a process of anaerobic digestion. The biochemical composition of microalgae, including trace elements of iron, cobalt, and zinc, meets the general nutrient requirements of anaerobic microbiota (Grobbelaar, 2007). The incubation of microalgae residual biomass with anaerobic microbes did also stimulate methanogenesis (Sialve *et al.*, 2009). The amount of biogas produced depended on the microalgal species used, since the relative proportion of proteins, carbohydrates, and lipids in the cells can influence the activities of anaerobic bacteria and archaea (Illman *et al.*, 2000). Another factor that may affect the methanogenic potential of microalgae is the protease resistance of their cell walls, which limits the effectiveness of the primary microbial degradation in the anaerobic digesters (Angelidaki *et al.*, 2004). Nine bacterial strains with endoglucanase activity were able to degrade the cell walls in the microalgae *Botryococcus braunii* and *Nannochloropsis gaditana* (Muñoz *et al.*, 2014). Bio-augmentation with a cellulolytic and

hydrogenogenic bacterium *Clostridium thermocellum* improved the degradation of *Chlorella vulgaris* biomass, resulting in higher levels of methane and hydrogen production, thus increasing the overall biogas yield and efficiency (Lü *et al.*, 2013, Munoz *et al.*, 2006).

Conclusion and perspective

Bacteria have long been considered as contaminants in algal cultures, thus efforts have been paid to obtain axenic algal mono-cultures for developing biomass and biochemical production processes. It is nowadays realized that microalgae-bacteria consortia may be utilized to improve algal biomass production and to enrich the biomass with valuable chemical and energy compounds. The chemical complexity of microalgal-bacteria interactions includes a wide variety of exchanged metabolites, molecular signals, and transporters, whose functions still needs to be further investigated. Most molecular signals, genes, and enzymes involved in microalgal-bacteria symbiosis remain to be elucidated. A greater insight at molecular level with respect to the regulation of microalgal-bacterial interaction will be useful (Fuentes *et al.*, 2016). Compared to the correct integration of genes and the proper expression of enzymes in a single organism within the right regulatory/metabolic network, using mixed microbial communication and their ability for “the division of labour” could be an easier way to process optimization (Subashchandrabose *et al.*, 2011). We propose the control of such biological interaction as efficient tool to increase biomass yield and reduce cultivation costs in microalgae production systems. With the increased demand of microalgal biomass for industrial applications, one of the key challenges will be the controlled integration of specific bacteria in the specific microalgal production processes. Hence, the recognition of symbiotic microalgal-bacteria interaction is likely to have profound implications for future exploitation of microalgae, both as an energy source and for biotechnological application. Potential use of bacterial symbiont in microalgal cultivation will have commercial and environmental positive impacts in the future.

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Conflict of interestt

None declared

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Table 1. Symbiotic bacteria involved in microalgal growth promotion.

Microalgae	Symbiotic bacteria	Symbiosis factor	References
<i>Amphidinium operculatum</i>	<i>Halomonas</i> sp.	Vitamin B ₁₂	Croft <i>et al.</i> , 2005
<i>Porphyridium purpureum</i>	<i>Halomonas</i> sp.	Vitamin B ₁₂	Croft, <i>et al.</i> , 2005
<i>Chlamydomonas nivalis</i>	<i>Mesorhizobium</i> sp.	Vitamin B ₁₂	Kazamia <i>et al.</i> , 2012
<i>Lobomonas rostrata</i>	<i>Mesorhizobium</i>	Vitamin B ₁₂	Kazamia <i>et al.</i> , 2012
<i>Dunaliella salina</i>	<i>Marinobacter</i> sp., <i>Halomonas</i> sp., <i>Pelagibaca</i> sp.	Siderophore	Baggesen <i>et al.</i> , 2014
<i>Dunaliella bardawil</i>	<i>Halomonas</i> sp.	Siderophore	Keshatacher <i>et al.</i> , 1995
<i>Scippsiella trochoidea</i>	<i>Marinobacter</i> sp.	Siderophore	Amin <i>et al.</i> , 2009, 2012
<i>Hemiaulus membranaceus</i>	<i>Richelia intracellularis</i>	Nitrogen	Foster <i>et al.</i> , 2011
<i>Rhizosolenia clevei</i>	<i>Richelia intracellularis</i>	Nitrogen	Foster <i>et al.</i> , 2011
<i>Chaetoceros</i> spp.	<i>Calothrix rhizosoleniae</i>	Nitrogen	Foster <i>et al.</i> , 2011
<i>Dunaliella salina</i>	<i>Alteromonas</i> sp., <i>Muricauda</i> sp.	Nitrogen	Chevanton <i>et al.</i> , 2013
<i>Pfiesteria piscicida</i>	<i>Roseobacter</i> sp.	Dimethylsulfoniopropionate	Alavi <i>et al.</i> , 2001
<i>Alexandrium fundyense</i>	<i>Roseobacter</i> sp.	Dimethylsulfoniopropionate	Ferrier <i>et al.</i> , 2002
<i>Emiliania huxleyi</i>	<i>Phaeobacter gallaeciensis</i>	Roseobacticides	Seyedsayamdost <i>et al.</i> , 2011

Table 2. Symbiotic and fermentative bacteria involved in microalgal biofuel production.

Microalgae	Bacteria	Biofuel	References
<i>Chlamydomonas</i> CC124	<i>Brevundimonas</i> sp., <i>Rhodococcus</i> sp., <i>Leifsonia</i> sp.	Hydrogen	Lakatos <i>et al.</i> , 2014
<i>Chlorella vulgaris</i>	<i>Pseudomonas</i> sp.	Triglyceride	Bell <i>et al.</i> , 2016; Guo & Tong, 2013
<i>Tetraselmis striata</i>	<i>Pelagibaca bermudensis</i>	Triglyceride	Park <i>et al.</i> , 2017
<i>Auxenochlorella protothecoides</i>	<i>Escherichia coli</i>	Triglyceride	Higgins <i>et al.</i> , 2014, 2016
Green microalgae NKG 120701	<i>Pseudoalteromonas undina</i>	Ethanol	Mastumoto <i>et al.</i> , 2003
<i>Chlamydomonas reinhardtii</i>	<i>Bacillus licheniformis</i>	Ethanol	Silva <i>et al.</i> , 2016
<i>Chlorella vulgaris</i>	<i>Clostridium thermocellum</i>	Hydrogen, methane	Lü <i>et al.</i> , 2013
<i>Botryococcus braunii</i>	<i>Aeromonas</i> sp., <i>Raoultella</i> sp.	Methane	Munoz <i>et al.</i> , 2014
<i>Nannochloropsis gaditana</i>	<i>Raoultella ornithinolytica</i>	Methane	Munoz <i>et al.</i> , 2014

Figure legends

Figure 1. A simplified diagram of general and specific interactions between microalgae and symbiotic bacteria. In general interactions, microalgae exude dissolved organic matter (DOM), which becomes available for bacteria. In return, the bacteria re-mineralize sulfur (S), nitrogen (N), and phosphorus (P) to support further growth of the microalgae. In specific interactions, the bacteria supply B vitamins as organic cofactors or produce siderophores to bind iron, which becomes bio-available for microalgae, while the microalgae supply dissolved organic carbon (DOC) in return for the bacteria.

Figure 2. Microalgae-bacteria-based growth system for biomass and biofuel production. Symbiotic bacterial interaction with microalgae are through exchanged metabolites or elements such as dissolved organic matter (DOM), vitamin (Vit), iron (Fe), sulfur (S), nitrogen (N), and phosphorus (P). The stored carbohydrates in microalgal cells may produce hydrogen while re-direction of photosynthate to triglyceride may be converted into diesel fuels. The residual biomass of microalgae can be further fermented to ethanol or biogas by yeast or anaerobic bacteria. Most molecular signals, genes and enzymes involved in microalgal-bacteria symbiosis remain to be elucidated.

Fig. 1.

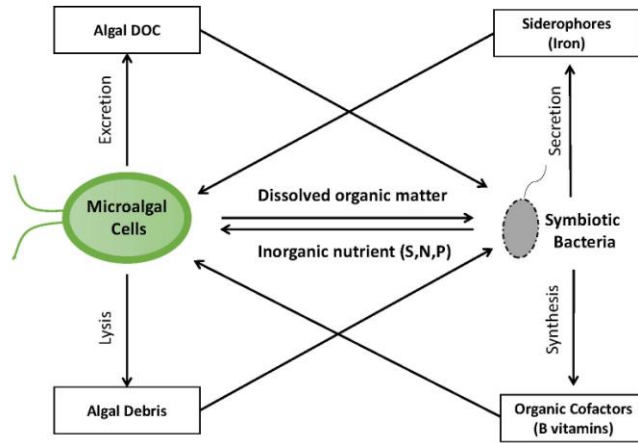


Fig. 2.

