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Review

Antibacterial activity of freshwater microalgae: A review

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The photoautotrophic microorganisms collectively termed 'micro-algae' (including micro-eukaryotes and cyanobacteria) are known to produce a wide range of secondary metabolites with various biological actions. They are known as well for their richness in bioactive compounds, with promising applications in pharmaceutical formulations. Their cell-free extracts have accordingly been tested as additives for food and feed formulation, in attempts to circumvent use of antimicrobial compounds of synthetic origin, or subtherapeutical doses of regular antibiotics. The increased use of antibiotics and chemotherapeutants for disease treatment leads to emergence of drug resistant forms. It also adversely affects the ecosystem. Microalgae are rich source of antimicrobial agents and provide a safer and cost effective way of treating bacterial infections. This article describes the antibacterial properties of some freshwater microalgae like, *Euglena*, *Microcystis*, *Chlorella*, *Chroococcus*, *Anabaena*, *Oscillatoria* and *Spirulina*.

Key words: Antibacterial, *Aeromonas hydrophila*, bioactive compounds, extraction, *Euglena viridis*.

INTRODUCTION

Microbial infections are one of the prominent causes of health problems, physical disabilities and mortalities around the world. There is a widespread belief that green medicines are healthier and harmless or safer than synthetic ones because of their limited side effects. Algae are now drawing a greater interest following the increase in demand for biodiversity in the screening programs seeking therapeutic drugs from natural products. Microalgae exhibit a notable biodiversity; they can in fact be found as individual cells, colonies or extended filaments. These microorganisms account for the basis of the food chain in aquatic ecosystems; they possess the intrinsic ability to take up H_2O and CO_2 that, with the aid of solar energy, are used to synthesize complex organic compounds, which are subsequently accumulated and/or secreted as primary or secondary metabolites. They are ubiquitously distributed throughout the biosphere, where they have adapted to survival under a large spectrum of environmental stresses for example, heat, cold, drought, salinity, photo-oxidation, anaerobiosis, osmotic pressure and UV

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exposure (Tandeau-de-Marsac, 1993); hence, they may grow essentially under all environmental conditions available, ranging from freshwater to extreme salinity, and can survive in moist, black earth and even desert sands and they have as well been found in clouds, being in addition essential components of coral reefs. This wide spectrum of ecosystems contributes to the myriad of chemical compounds that they are able to synthesize, thus accounting for their unique potential as stakeholders in blue biotechnology. As many as 30,000 distinct microalgal species might inhabit the earth and over 15,000 novel compounds have been chemically obtained from them (Cardozo et al., 2007; Rodr´ıguez-Meizoso et al., 2010). In addition, their importance as a source of novel compounds is growing rapidly, and researches have indicated that these compounds exhibit diverse biological activities (Wijesekara et al., 2010).

The problem of microbial resistance is growing and the outlook for the use of antimicrobial drugs in the future is still uncertain. The majority of clinically used antimicrobial drugs have drawbacks like toxicity, lack of efficacy, inhibiting cost and their frequent use leading to the emergence of resistant strains. Thus, there is an urgent need to develop alternative biodegradable agents, which should have minimal side effects. It is generally considered that natural compounds are biodegradable and so more environmentally acceptable. Commercial applications of microalgae derived compounds have, as yet, received little attention in the area of pharmaceuticals, antibiotics and other biologically active compounds. Some researchers have envisioned the enormous possibilities of algae and microalgae as potential source of bioactive compounds; particularly, some microalgae have been studied as a potential natural source of different functional compounds (Herrero et al., 2006; Rodrı´guez-Meizoso et al., 2008). They are able to produce a wide range of biologically active substances with antibacterial, antiviral, antifungal, enzyme inhibiting, immunostimulant, cytotoxic and antiplasmodial activities (Ghasemi et al., 2004).

Freshwater microalgae as source of antibacterial agents

Microalgae have a significant attraction as natural source of bioactive molecules, because they have the potential to produce bioactive compounds in culture, which are difficult to be produced by chemical synthesis (Borowitzka and Borowitzka, 1989; Goud et al., 2007; Kaushik and Chauhan, 2008). Most of those compounds are accumulated in the microalgal biomass; others are excreted during growth into the environment (Jaki et al., 2000, 2001). This review article describes some freshwater microalgae, which contains potential bioactive compounds for antimicrobial activity.

Euglena viridis

Euglena viridis is a unicellular flagellate algal protist, which occur both in freshwater and marine forms. The taxonomic position of this alga is Phylum Euglenozoa, Class Euglenida, Order *Euglenales* and Family *Euglenaceae* (Ehrenberg, 1830). It is usually a free swimming, fusiform, elongate, lanceolate, spindle-shaped, flexible unicellular mobile animal. It has one or rarely two flexible flagella issuing out of an anterior notch at the base of which is an oval aperture and distinctive red pigment spot known as eye-spot. *E. viridis* is characterised by a single stellate group of band shaped chloroplast and finely striated delicate periplast, varying from 40 to 150 µm in length. It exhibits positive phototaxis, determined by a photoreceptive spot in the wall of the contractile vacuole. All members contain chlorophyll a, b, β-carotene and xanthophylls (Cunningham and Schiff, 1986; Fiksdahla and Liaaen-Jensena, 1988). There is no cell wall and the reserve food material is paramylon and oil. *Euglena* forms red blooms in all type of water bodies when density is very high, characterised by formation of haematochrome during bright sunny days. The coloration is green in cloudy days (Biswas, 1949).

Microcystis aeruginosa

Microcystis is a type of blue green alga (also referred to as cyanobacteria) and are the dominant phytoplankton group in eutrophic freshwater bodies (Davidson, 1959; Negri et al., 1995). The unicellular, planktonic freshwater cyanobacterium belongs to Phylum Cyanobacteria; Class Cyanophyceae; Order Chroococcales and Family Microcystaceae (Kützing, 1846). Among all the species reported worldwide, *Microcystis aeruginosa* is one of the important cyanobacteria characterized by mucilage covering with a cell size of 3 to 4 µm and with varying colonies ranging from few to hundreds of cells (Biswas, 1949). It is one of more than 700 species of algae that may be found in water samples collected and usually blooms in mid to late summer. The extracellular covering of *M. aeruginosa* is divided into several layers: the cytoplasmic membrane or plasmalemma, the peptidoglycan layer, and the multilayered structure of the cell wall (Kim et al., 1997). These common bloom-forming algae are especially abundant in shallow, warm, nutrient enriched fresh waters and lower salinity estuaries. These are also found in polluted water low in oxygen and can grow to form thick scums that could colour the water (Stotts et al., 1993). *M. aeruginosa* produces vast number of peptides for example, aeruginosins, microginins, microviridins, aeruginoguanidins, aeruginosamides, kasumigamide, some of which are highly toxic (Ishida and Murakami, 2000; Ishida et al., 2002). They are producing two kinds of toxin, the cyclic peptide hepatotoxin and the alkaloid neurotoxin.

Chlorella vulgaris

The simple and common fresh water, single-celled green algae of the genus *Chlorella* (Beijernick, 1890), are placed under the order Chlorococcales and family Chlorellaceae (Hoek et al., 1995). Cells are spherical in shape tending to aggregate into colony; yellowish green, 4 to 8 µm in diameter, and is without flagella. *Chlorella* contains the green photosynthetic pigments chlorophyll-a and -b in its chloroplast often with one pyrinoid, more or less situated in the middle. Cell wall is smooth and thin hyaline. *Chlorella vulgaris* (*C. vulgaris*) is a genus of unicellular green algae containing high level of protein (50 to 70% of dry matter), lipid, vitamins and minerals (Phang, 1992). *Chlorella* is a nutrient-dense super food that contains 60% protein, 18 amino acids (including all the essential amino acids), and various vitamins and minerals. When dried, it is about 45% protein, 20% fat, 20% carbohydrate, 5% fiber, and 10% minerals and vitamins (Phang, 1992; Khatun et al., 1994). The nucleus of *Chlorella* cell contains a unique nucleotide-peptide complex known as *Chlorella* growth factor (CGF) which actually promotes cell growth in the body, stimulating tissue repair and healing to an extent. It is also a reliable source of essential fatty acids that are required for many important biochemical functions, including hormone balance. *Chlorella* also contains high levels (ranging from 3 to 5%) of chlorophyll, beta-carotene and RNA/DNA (Gouveia et al., 1996). Chlorophyll is one of the greatest food substances for cleansing the bowel and other elimination systems, such as the liver and the blood. More than 20 vitamins and minerals are found in *Chlorella*, including iron, calcium, potassium, magnesium, phosphorous, pro-vitamin A, vitamins C, B1, B2, B5, B12, E and K, inositol, folic acid (Milner, 1953).

Spirulina platensis

Spirulina is a photosynthetic, filamentous, spiral-shaped, multicellular blue green microalga, generally found in fresh water. The two most important species are *Spirulina maxima* and *Spirulina platensis*. It contains carotenoid, chlorophyll, and major phycocyanin pigment. It belongs to Cyanophyceae class, Oscillatoriaceae family; this cyanobacteria is characterized by spiral chains of the cells enclosed in a thin sheath. It is rich in nutrients, such as proteins, vitamins, minerals, carbohydrates and γ-linolenic acid (James et al., 2006). It consists of 60 to 70% protein in dry weight. The protein elements consist of 18 types of amino acids, several vitamins, such as vitamins A, B, E, and K, minerals, and fatty acids necessary for the body. It is gaining more and more attention, not only for the food aspects but also for the development of potential pharmaceuticals (Quoc and Pascaud, 1996). *Spirulina* contains a whole spectrum of natural mixed carotene and xanthophyll phytopigments which, together with phycocyanin, seem to be related to its antioxidant activity (Pineiro et al., 2001). Basic constituents of different freshwater microalgae were represented in Figure 1.

Antibacterial compounds from freshwater microalgae

Microalgae constitute one of the commercially important living and renewable resources. They contain more than sixty trace elements including minerals, proteins, iodine, bromine and many bioactive substances. To date, many chemically unique compounds of fresh water origin with various biological activities have been isolated, and some of them are under investigation and some are being used to develop new pharmaceuticals. Algae are a very interesting natural source of new compounds and many of them possess antioxidant, antimicrobial, and antiviral activities (Plaza et al., 2010; Rodr´ıguez-Meizoso et al., 2010). These organisms live in habitats exposed to extreme conditions, and therefore they must adapt rapidly and efficiently, and as a consequence, produce a great variety of biologically active secondary metabolites that participate in the natural defense mechanisms (Rodr´ıguez-Meizoso et al., 2010). These defense strategies can result in a high level of structural and chemical diversity of compounds, originating from different metabolic pathways. Microalgae can biosynthesize, metabolize, accumulate and secrete a great diversity of primary and secondary metabolites, many of which are valuable substances with potential applications in the food, pharmaceutical and cosmetics industries (Yamaguchi, 1997). A large number of algal extracts have been found to have antimicrobial activity (Mao and Guo, 2010; Plaza et al., 2010). Major group of antimicrobial agents found in freshwater microalgae are fatty acids, lipids, pigments, polyphenols, carbohydrates, simple hydrocarbons and some other derivatives (Table 1).

Much attention has been focused on the microalgae as sources of novel, biologically active compounds such as phycobiline, phenols, phenolic glycosides, saponins and phytoalexins terpenoids, steroids and polysaccharide (Li et al., 2007). The important compounds also identified as antimicrobial are fatty acids, acrylic acid, halogenated aliphatic compounds, terpenes, sulphur containing hetero cyclic compounds, carbohydrates and phenols (Kannan et al., 2010). Many of the structures identified as fatty acids and hydroxyl unsaturated fatty acids, glycolipids, steroids, phenolics and terpenoids. lauric acid, palmitic acid, linolenic acid, oleic acid, stearic acids are known to be potential antibiotics (MacMillan et al., 2002; Shanab, 2007; Tan, 2007). Algal lipids are composed of glycerol, sugars or bases esterified to saturated or unsaturated fatty acids (12 to 22 carbon atoms). Among all the fatty acids in microalgae, some fatty acids of the ω3 and ω6

Table 1. Antibacterial compounds from different algae and their target bacterial pathogens.

Antibacterial compound	Microalgae	Target bacterial pathogens	References
Pigments	*Anabaena cylindrical Chlorococcum humicola, Spirulina platensis, Nostoc*	*E. coli, S. typhimurium*, *K. pneumoniae*, *V. cholerae*, *S. aureus*, *B. subtilis, Streptococcus* sp., *Pseudomonas* sp., *Bacillus* sp., *Staphylococcus* sp., *E. coli, Enterobacteria aerogens*	Jaya Prakash et al. (2007), Bhagavathy et al. (2011), Muthulakshmi et al. (2012) and Fan et al. (2013)
Fatty acids and Lipids	*Dunaliella salina*, *Haematococcus pluvialis*, *Phaeodactylum tricornutum, Chaetoceros muelleri, Spirulina platensis*	*Escherichia coli, Staphylococcus aureus,* MRSA, *Listonella anguillarum, Lactococcus garvieae, Vibrio* spp	Xue et al. (2002), Herrero et al. (2006) and Santoyo et al. (2009)
Carbohydrates	*Anabaena sphaerica*, *Chroococcus turgidus*, *Oscillatoria limnetica, S. platensis, Porphyridium cruentum*	*E. coli, S. typhimurium, S. faecalis*	O'Doherty et al. (2010) and Abdo et al. (2012)
Polyphenols	*Anabaena sphaerica*, *Chroococcus turgidus*, *Oscillatoria limnetica* and *Spirulina platensis*	*Salmonella typhi, Streptococcus*, *E. coli* and *Staphylococcus aureus*	Gao and Zhang (2010), Klejdus et al. (2010), Shu et al. (2011), Abdo et al. (2012) and Hetta et al. (2014)

families are of particular interest (Spolaore et al., 2006). Many authors have found that antibacterial activities of micro algae are due to fatty acids (Kellam et al., 1988). Antibacterial activity of unsaturated and saturated long chain fatty acids of chain length more than 10 carbon atoms induced lysis of bacterial protoplasts.

Naviner et al. (1999) reported inhibition of various marine bacteria using fatty acids of the alga, *Skeletonema costatum*. *Chlamydomonas reinhardtii* has unsaturated fatty acids like linolenic, linoleic, oleic and palmitoleic acid and saturated fatty acids such as palmitic, stearic, myristic acid. Crude fatty acid extracts of *C. reinhardtii* showed antibacterial activity (Sudalayandi et al., 2012). *Spirulina* lipids were investigated as a natural source of functional bioactives because of its usefulness for human health (Ramadan et al., 2008). Specifically, the antimicrobial activity of the methanolic extract of *S. platensis* was explained by the presence of γ-linolenic acid (Demule et al., 1996), an antibiotically active fatty acid present in a high concentration in this alga (Xue et al., 2002). A bioactive modified peptide, aeruginosamide has been isolated from the cyanobacterium *Microcystis aeruginosa* (Lawton et al., 1999).

Carbohydrates in microlagae can be found in the form of starch, glucose, sugars and other polysaccharides. Most microalgae produce polysaccharides and some of them could have industrial and commercial applications, considering the fast growth rates and the possibility to control the environmental conditions regulating its growth. Aqueous extract of five freshwater algal species, *Anabaena sphaerica*, *Chroococcus turgidus*, *Oscillatoria limnetica*, *Spirulina platensis* and *Cosmarium leave* contains polysaccharides which showed antibacterial activity when tested against *Escherichia coli, Salmonella typhimurium* and *Streptococcus faecalis* (Abdo et al., 2012). The major carbohydrate of *Euglena*, the glucose polymer paramylon, and its role in the biogenesis of chloroplasts have been investigated widely during the past 15 years (Dwyer and Smillie, 1970; Manners and Sturgeon, 1982). The most promising microalga for commercial purposes is the unicelular red alga *Porphyridium cruentum*, which produces a sulphated galactan exopolysaccharide that can replace carrageenans in many applications (Gouveia et al., 2008). Certain highly sulphated algal polysaccharides also present pharmacological properties acting on the stimulation of the human immune system (Pulz and Gross, 2004). Among many different algal polysaccharides, the most important are galactans, fucoidan, laminarin and alginates (Ferreira et al.,

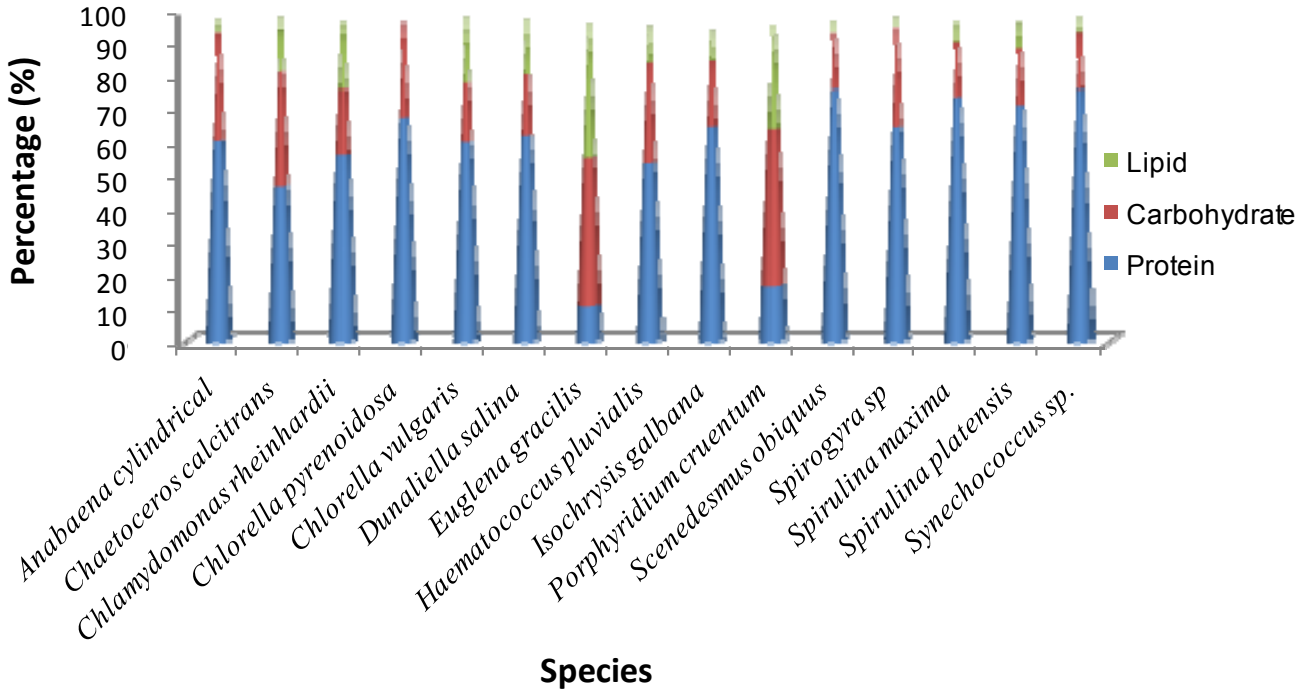

Figure 1. Basic constituents of different freshwater microalgae in percentage.
Source: Gouveia et al. (2008).

2012). Laminarin is one of the major polysaccharides found in brown algae with antiviral and antibacterial properties (O'Doherty et al., 2010). Fucoidan is a sulfated polysaccharide present in the cell wall matrix of brown algae that has been demonstrated to inhibit certain enveloped viruses with low toxicity (Elizondo-Gonzalez et al., 2012).

In some instances it appears that water soluble chlorophyll derivatives have antibacterial activity (Mowbray, 1957). Smith states that "chlorophyll is not strictly bactericidal but that it does exert a definite bacteriostatic and even a bactericidal effect under suitable environmental conditions." Nevin and Bibby (1954) showed that chlorophyll inhibited the growth of staphylococci, streptococci and lactobacilli. Chlorophyll inhibited the growth of the tested oral bacteria, particularly *Porphyromonas gingivalis* and *Fusobacterium nucleatum* (Saeki et al., 1993). Jaya et al. (2007) evaluated the antibacterial activity of chlorophyll a and b from certain fresh water micro-algae from the sriver Godavari (India).

The main carotenoids produced by microalgae are β-carotene from *Dunaliella salina* and astaxanthin from *Haematococcus pluvialis*. Carotenoids were reported to have antibacterial activity (Mahanom et al., 1990). The protective effect of β-carotene from green algae, *Chlorococcum humicol* has also been reported (Bhagavathy and Sumathi, 2010). The main natural resources of phycobiliproteins are the cyanobacterium *Spirulina* (*Arthrospira*) for phycocyanin (blue) and the rhodophyte *Porphyridium* for phycoerythrin (red) chromophoric prosthetic groups, named phycobilins. Phycobiliproteins are water soluble pigments produced by cyanobacteria (blue-green algae), red algae and crypto-monads (Mihov et al., 1996). Cyanobacteria, Rhodophyta and Cryptomonads algae contain phycobiliproteins, deep colored water-soluble fluorescent pigments, which are major components of a complex assemblage of photosynthetic light-harvesting antenna pigments, the phycobilisomes (Glazer, 1994). In *Spirulina*, phycocyanin is a phycobiliproteins; it is used against many bacterial infections and has anti-inflammatory, antioxidant and antiviral properties. It is effectively active against human pathogens such as *Streptococcus* sp., *Staphylococcus* sp., *E. coli*, *Bacillus* spp., and *Pseudomonas* spp. (Muthulakshmi et al., 2012; Murugan, 2012). Large quantities phycocyanin was isolated and partially purified from *Anabaena cylindrical* and filamentous fresh water cyanobacterium *Westiellopsis* spp., which was tested against Gram positive and Gram negative bacteria (Sabarinathan et al., 2008; Abdo et al., 2012).

It was reported that five freshwater microalgae, *Anabaena sphaerica*, *Chroococcus turgidus*, *Oscillatoria limnetica*, *Spirulina platensis* and *Cosmarium leave* from an Egyptian water station contain quercetin with antibacterial activity (Rattanachaikunsopon and Phumkhachorn,

2010; Abdo et al., 2012). Three isoflavone compounds were found in freshwater algae and cyanobacteria (Klejdus, et al., 2010). It was also reported that the phenolic content are active as antibacterial against different types of microorganisms like *Salmonella typhi* (Ouattara et al., 2011) and the flavonoids are active against several strains like *Streptococcus* (Shu et al., 2011); *E. coli* and *Staphylococcus aureus* (Gao and Zhang, 2010).

Many phytochemicals not mentioned have been found to exert antimicrobial properties. This review has attempted to focus on reports of chemicals which are found in multiple instances to be active. It should be mentioned, however, that there are reports of antimicrobial properties associated with microalgal-derived oxylipins, the antibacterial activities of polyunsaturated aldehydes deserve a special mention. Such compounds are synthesized by diatoms, for example *S. costatum* and *Thalassiosira rotula*. One illustrative example is decadienal – probably derived from (the polyunsaturated) arachidonic acid (C20:4 n-3), which exhibits a strong activity against such important human pathogens as multi resistance *S. aureus* (MRSA) and *Haemophilus influenza* as well as against *E. coli* and *Pseudomonas aeruginosa*, and *S. aureus and S. epidermidis* (Smith et al., 2010). Some volatile components and various extracts of *Spirulina* also showed antibacterial activities (Ozdemir et al., 2004; El-Sheekh et al., 2014).

Antibacterial activity

Microalgae have for long been used with therapeutic purposes; their systematic screening for biologically active principles began in the 1950s. The first investigation on the antibiotic activity of algae was carried out by Pratt et al. (1944). However, in the last decade microalgae have become the focus of extensive research efforts, aimed at finding novel compounds that might lead to therapeutically useful agents (Mendes et al., 2003; Cardozo et al., 2007). Pratt et al. (1944) isolated the first antibacterial compound from a microalga, *Chlorella*; a mixture of fatty acids, viz. chlorellin, was found to be responsible for that inhibitory activity against both Gram+ and Gram- bacteria. The methanolic extract of cyanobacteria has been investigated for *in vitro* antimicrobial activity against *Proteus vulgaris, Bacillus cereus, E. coli, P. aeruginosa, Aspergillus niger* and *A. flavus* using agar cup plate method (Prashantkumar et al., 2006). Although microalgae can synthesize a few useful products, search for novel antibiotics is still incipient; illustrative examples are presented in Table 2.

Antibiotics are typically less effective against Gram-negative bacteria because of their complex and multilayered cell wall, which makes it more difficult for the active compound to penetrate (Ördög et al., 2004); this justifies why the antibacterial activity of the supernatant (and methanolic extracts) is more potent against Gram-positive than Gram-negative bacteria (Ghasemi et al., 2004, 2007). Hexadecatrienoic acid isolated from *P. tricornutum* displays activity against (the Gram-positive pathogen) *S. aureus*. Pressurized (liquid) ethanol extracts from *Haematococcus pluvialis* in its red stage possess antimicrobial activity against a Gram-negative bacterium, *E. coli*, and a Gram-positive bacterium, *S. aureus;* this was once again associated with the presence of short-chain fatty acids, namely butanoic and methyl lactic acids (Santoyo et al., 2009). The antimicrobial potential of fresh water microalgae viz., *Oscillatoria sancta* (*O. sancta*) (Kuetz), *Lyngbya birgei*, *Oedogonium echinospermum*, *Spirogyra decimina* (Muller), *Spirogyra grantiana*, *Spirogyra crassa*, *Spirogyra biformis* and *Spirogyra condensata* (Vaucher) against human bacterial pathogens were screened. Antimicrobial study was carried out by disc diffusion method against the pathogens viz., *E. coli* (ATCC 35218), *S. aureus* (ATCC 6538), *S. typhi* (MTCC 733), *Proteus vulgaris*, *Proteus mirabilis* and *Streptococcus pyogenes* (Prakash et al., 2011).

Mechanism of action of antibacterial agents

Antimicrobial agents act selectively on vital microbial functions with minimal effects or without affecting host functions. Different antimicrobial agents act in different ways. The understanding of these mechanisms as well as the chemical nature of the antimicrobial agents is crucial in the understanding of the ways how resistance against them develops. Broadly, antibacterial agents may be described as either bacteriostatic or bactericidal (Salvador et al., 2007). However, the mechanism of action of antimicrobial agents can be categorized further based on the structure of the bacteria or the function that is affected by the agents. The modes of action of different antibacterial agents are represented in Table 3.

The exact mechanism of action of fatty acids remains unknown: they may act upon multiple cellular targets, even though cell membranes are the most probable ones as membrane damage will likely lead to cell leakage and reduction of nutrient uptake, besides inhibiting cellular respiration; conversely, Desbois et al. (2009) claimed a peroxidative process. Furthermore, compounds synthesized by *Scenedesmus costatum*, and partially purified from its organic extract, exhibited activity against aquaculture bacteria because their fatty acids are longer than 10 carbon atoms in chain length which apparently induce lysis of bacterial protoplasts. Since the fatty acid analysis indicated the presence of other fatty acids that had been also reported to have some antimicrobial activity, specifically palmitoleic and oleic acids, it was hypothesized that lipids kill microorganisms by leading to disruption of the cellular membrane (Lampe et al., 1998).

Table 2. Antibacterial features of selected compounds from microalgae.

Microalgae	Target bacterial pathogens		References
	Gram +ve	Gram -ve	
Euglena viridis	-	*Pseudomonas* sp., *Aeromonas* sp., *Vibrio* sp., *E. coli*, *Edwardsiella tarda*	Das and Pradhan (2010)
Spirulina platensis	*Streptococcus* sp., *Bacillus* sp., *Staphylococcus* sp.	*Pseudomonas* sp., *Aeromonas* sp., *Vibrio* sp., *E. coli*, *E. tarda*	Mar et al. (2008), Kaushik and Chauhan (2008), Ranga Rao et al. (2010), Muthulakshmi et al. (2012), Pradhan et al. (2012) and Helen et al. (2014)
Chlorella vulgaris	*S. aureus*, *S. epidermidis*, *B. subtilis*	*E. coli*, *Pseudomonas* sp., *Aeromonas* sp., *Vibrio* sp., *E. tarda. Salmonella typhi*, *Klebsiella pneumoniae*	Pratt et al. (1944), Matusiak et al. (1965), Ghasemi et al. (2007) and Vishnu and Sumathi (2014)
Microcystis aeruginosa	*B. subtilis, S. aureus, S. mutans*	*Pseudomonas putida, P. aeruginosa, P. fluorescens, E. coli, Aeromonas hydrophila,, Vibrio* sp., *Edwardsiella tarda*	Ishida et al. (1997), Madhumathi et al. (2011) and Silva-Stenico et al. (2014)
Haematococcus pluvialis	*Staphylococcus aureus*	*Escherichia coli,*	Jaime et al. (2010)
Nostoc	*Staphylococcus aureus*, *Bacillus subtilis*	*P. aeruginosa*, *E. coli*, *Salmonella*	Zornitza et al. (2000), Goud et al. (2007), Jaya Prakash et al. (2007) and Silva-Stenico et al. (2014)
Phaeodactylum tricornutum	*Listonella anguillarum, Lactococcus garvieae*	*Vibrio* spp.	-
Chroococcus turgidus	*Streptococcus faecalis, Salmonella*	*E. coli*	Abdo et al. (2012)
Anabaena sphaerica	*Streptococcus faecalis, Salmonella*	*E. coli, Salmonella*	Goud et al. (2007) and Abdo et al. (2012)
Oscillatoria limnetica	*Streptococcus faecalis, S.mutants, Staphylococcus aureus, B. subtilis*	*E. coli, K. pneumonia, P. aeruginosa*	Goud et al. (2007), Abdo et al. (2012) and Helen et al. (2014)

The susceptibility of Gram-negative bacteria to killing by lipids was notable (Bergsson et al., 2002) and is probably due to the differences in the outer membrane or the cell wall of bacteria.

Conclusion

The problem of microbial resistance is growing and the outlook for the use of antimicrobial drugs in the future is still uncertain. The majority of clinically used antimicrobial drugs have drawbacks like toxicity, lack of efficacy, inhibiting cost and their frequent use leading to the emergence of resistant strains. Thus, there is an urgent need to develop alternative biodegradable agents, which should have limited side effects. It is generally considered that natural compounds are biodegradable and so more environmentally acceptable. Commercial applications of microalgae-derived compounds have, as yet, received little attention in the area of pharmaceuticals, antibiotics and other biologically active compounds. So this search prompted the exploration of natural algal products.

Scientists from divergent fields are investigating algae anew with an eye to their antimicrobial usefulness. A sense of urgency accompanies the search as the pace of species extinction continues. Laboratories around the world have found literally thousands of phytochemicals which have inhibitory effects on all types of microorganisms *in vitro*. More of these compounds should be subjected

Table 3. Mechanism of action of some antimicrobial agents.

Antimicrobial agent	Mechanism of action	References
Carotenoids	Digestion of cell wall by lysozyme enzymes.	Cucco et al. (2007)
Flavonoids	Increase in permeability of the inner bacterial membrane and a dissipation of the membrane potential	Mirzoeva et al. (1997)
Polyphenols	Binds to adhesins, enzyme inhibition, substrate deprivation, complex with cell wall, membrane disruption	Amaro et al. (2011)
Polysaccharide	Inhibition of hyaluronidase.	Amaro et al. (2011)
Fatty acids and Lipids	Disruption of the cellular membrane	Lampe et al. (1998) and Desbois et al. (2009)

to animal and human studies to determine their effectiveness in whole-organism systems, including particular toxicity studies as well as an examination of their effects on beneficial normal microbiota. It would be advantageous to standardize methods of extraction and *in vitro* testing so that the search could be more systematic and interpretation of results would be facilitated. Also, alternative mechanisms of infection prevention and treatment should be included in initial activity screenings.

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