The Diatoms: From Eutrophic Indicators to Mitigators

Aviraj Datta, Thomas Kiran Marella, Archana Tiwari, and Suhas P. Wani

1 Introduction

Human activities can bring negative effects in the environment. Any substance that can cause a negative effect in the environment is considered a pollutant which has to be controlled in order to reduce adverse impacts. Pollutants can come from different sources, although human activities like agriculture, change of land use, and others are one source of production of pollutants which can affect the environment (Gottschalk et al. 2011). Elevated nutrients contribute to poor lake ecosystem, which highlights the need for efficient nutrient removal strategies that enable us to protect or restore the water bodies from eutrophication. Biological elements, such as macroinvertebrate species, macrophytes, diatoms, and zooplankton, have been used to monitor nutrient changes (Lougheed et al. 2007). Diatoms are one of the most explored species for water quality assessment around the world, due to their sensitive time-dependent response (Stevenson 2014).

The main purpose of developing biological monitoring strategies is to enable researchers to assess water quality of lotic and lentic systems. This approach makes use of aquatic biota to evaluate complex and dynamic changes in water quality. Biotic communities are generally sensitive to inflow of chemicals and change in physical factors that bring about a change in their morphology and diversity which reflects the physiochemical conditions of the ecosystems. This approach uses biota to represent the general environmental conditions and assess environmental quality of the monitored ecosystem.

The biotic organisms of water include macroinvertebrates, phytoplankton, zooplankton, phyto-benthic macroinvertebrates, and the fish communities (De Pauw

A. Datta (✉) · T. K. Marella · S. P. Wani
International Crops Research Institute for the Semi-Arid Tropics, Patancheru, India

A. Tiwari
AMITY University, Noida, India

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et al. 2000). The ecological indicators are used based on species diversity of these organisms to monitor water quality, hydrology, and the overall health of a water body. Indicators species are used to monitor the levels of toxins, physicochemical parameters, and the overall nature of the water resource (Nixon 2009).

The role played by algae is crucial in all water ecosystems. They are identified as strands or filaments in rivers and along the lake shorelines and act as a link between the biotic and abiotic environments. The algal community assemblage and abundance change in response to water quality fluctuations, and this can be attributed to their direct reliance on making them sensitive to water quality changes. The sensitivity of algae to water quality changes makes them useful as bioindicators of the physical and chemical properties in water environments. Diatoms are single-celled organisms and basically the lone member group of algal organisms applied in aquatic studies until recent years (Ruhland et al. 2008). They are represented by over 100,000 species all over the world and are identified in rivers and from the lake shorelines as brown, slimy covering on submerged substrates such as mud, sand, macrophytes, or rocks. The benefits of diatoms used as bioindicators include the following: they are easily identifiable under a microscope, and they have cell walls with each species having specific shape and morphological structure. Diatom classification is well detailed and defined, as well as various species tolerance to environmental changes. The species cell walls composed of silica from silicon resist decomposition, and so can be preserved, thus providing a permanent record whereby short- or long-term changes can be assessed (Cox 1991). Moreover, historical conditions of water can be projected by use of the species cell walls preserved in sediments at the bottom of the lakes (Lavoie and Campeau 2010).

Diatoms play a major role in biomass production and sinking of atmospheric greenhouse gas in oceans. Diatoms are responsible for about 20% of the total photosynthetic CO_2 fixation, which is equivalent to the photosynthetic activity of all rainforests combined and approximately 40% of annual marine biomass production (Falkowski et al. 1998). Diatoms are exceedingly robust and can inhabit virtually all photic zones from the equator to arctic where they are extensively studied for their usefulness as indicators of changes in physiochemical conditions due to their rapid response to any slight changes. Thus, diatoms show high degree of flexibility in varied culture conditions that could be useful for their use in biotechnological applications despite challenging conditions. Diatoms are sensitive to changes in their aquatic environments and are reliable indicators of the water quality. The reason for this is their reproduction rate, which allows for significant increase in population of a given species under favorable conditions while other species concurrently decrease or disappear.

Diatoms are members of the heterokont class of algae, which are highly different, and have a more complex evolutionary history than green algae and vascular plants. The evolutionary age of diatoms has been estimated from molecular genetic data as 165–240 M·ya (Kooistra and Medlin 1996), which is in reasonable agreement with the fossil record. Diatoms are secondary endosymbionts and part of the heterokont group, which includes other silica-forming algae. Diatom genomes are a complex mixture derived from combination of higher organisms of both plant and animal origin. This unique combination gave diatoms a peculiar metabolic profile and process which is different from other algae (Armbrust et al. 2004). The evolutionary success of diatoms is also connected to their cell wall which is made up of silica which needs lower energy requirement to build when compared with (Raven 1983).

Diatoms are divided into four major groups based on their cell wall structure, radial centrics, bipolar and multipolar centrics, araphid pennates, and raphid pennates (Fig. 1). All these groups have evolved under decreasing CO_2 levels during the Mesozoic era (Armbrust 2009). This has led to an advanced carbon-concentrating mechanism making them highly adaptable to changing CO_2 levels.

Silica cell wall gives diatom algae an advantage of enhanced sinking rate which results in increased carbon burial in shallow seas and continental margins (Smetacek 1999; Falkowski et al. 2005) and are known to be primary contributors to present nascent petroleum reserves.

Diatoms are useful indicators of water quality because of their diversity in varied environments, species richness, and dynamic response to changes in physicochemical conditions of surrounding ecosystem (Dixit et al. 1992; McCormick and Cairns 1994). Diatoms play a significant role in controlling and biomonitoring of organic pollutants, heavy metals, hydrocarbons, PCBs, pesticides, etc. in aquatic ecosystems. Although diatoms are extensively studied for their role as indicators of different kinds of water pollution, their application in phycoremediation of polluted water bodies has just started. In this chapter we explore the potential of diatoms as indicator species for pollution and their implications on wastewater treatment.

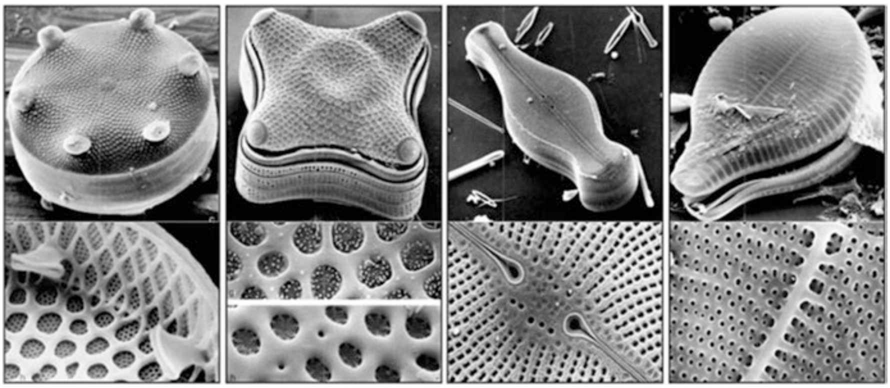

Fig. 1 Different silica frustule shapes and intrinsic frustule designs of diatoms *Aulacodiscus* sp., radial centric; *Amphitetras* sp., polar centric; *Didymosphenia* sp., raphid pennate; and *Podocystis* sp., araphid pinnate (Kröger and Poulsen 2008)

1.1 Why Diatoms as Bioindicators, Sensitivity of Diatoms to Physiochemical Changes

It is paramount to understand the biological, chemical, and physical processes of any water body in order to determine the mass balance of pollutants into and out of the system. The pollution of fresh water bodies from excess nutrients and hazardous chemicals is one of the greatest environmental issues of the developing world. For successful mitigation to these issues, along with treatment efficient monitoring approaches are needed. Ecosystem monitoring employs physical-, chemical-, and biological-based methods for routine monitoring. Although chemical and physical methods provide instant results, they do not provide us with information on previous dynamic changes of the ecosystem, but with biological monitoring we can get information on long-term effects on the ecosystem by different physicochemical fluxes. Therefore, complementing biological monitoring with physicochemical monitoring is the right way to monitor water quality.

In order to be considered as bioindicators, the species which are being monitored should show a strong correlation with a physiochemical parameter, should have a narrow tolerance range to that parameter, and should be commonly found in the sample. Diatoms meet all these criteria which make them ideal for biomonitoring water quality. Diatoms are present in all aquatic ecosystems due to which same species can be compared for assessment of different habitats like lakes, wetlands, oceans, streams, etc. Diatoms grow as attached biofilm on solid substrates so they can be monitored by sampling these substrates even when the water body is dry. Due to their faster growth rate compared to other species, they can give us an early warning to impeding pollution and water quality restoration. Diatom-based monitoring is cost-effective when compared with other methods, and they give an added advantage of retaining the samples for longer times for long-term studies. These attributes make diatom-based biomonitoring of water habitats an important parameter for habitat assessment in many countries worldwide.

Diatom-based water quality indices have been developed for monitoring water quality in many geographic areas. Nutrient influxes along with some physicochemical parameters are key factors which influence diatom growth and survival. Diatoms respond to nutrient influx by changing their community structure in terms of species response, where specific diatom species dominate nutrient-rich waters, whereas others prefer nutrient-depleted conditions. This dynamic response makes diatoms ideal indicators of nutrient enrichment. Physical and chemical monitoring methods where water samples are picked at one defined time cannot provide this dynamic nutrient influx data. Monitoring nonpoint source pollution of inorganic nutrients like phosphorus is quite difficult even with multiple sampling efforts due to its sudden fluctuations. With diatom-based monitoring, when diatom communities are exposed to cumulative nutrient, diatoms respond by changing their community structure leading to better monitoring efficiency (Table 1).

Nutrient monitoring based on diatoms is used widely since they are the major primary producers with an ability to strongly reflect their ecosystem nutrient

Table 1 Diatom-based monitoring of different parameters from varied ecosystems reported in literature

Ecosystem	Monitoring parameter/impact	References
Rivers, streams	Eutrophication	Lobo et al. (2004)
	Heavy metal contamination	Leguay et al. (2016)
Lakes	Eutrophication	Poulíčková et al. (2004)
	Heavy metal contamination	Cantonati et al. (2014)
Marine benthos	Various environmental parameters	Weckström and Juggins (2006)
Marine biofilm	Eutrophication	Cibic and Blasutto (2011)

concentrations by their community structure. Diatoms are useful for monitoring nutrient influx into lakes due to their relative abundance and richness which can provide a sensitive index for physicochemical changes (Black et al. 2011). Some diatom-based models measure interaction between diatom community dynamics, and nutrients can provide nutrient concentration information which will be useful to develop efficient management practices. Macroinvertebrates and fishes have also been used as biomonitors (Hering et al. 2006), but diatoms have an advantage due to their increased sensitivity (Leira and Sabater 2005). Benthic diatoms are known to be influenced more by local factors like major nutrients, pH, etc. than large-scale factors like climate and geology (Stevenson and Pan 1999; Leland 1995). Benthic diatoms also respond well to hydro-morphological modification and nutrient enrichment (Hering et al. 2006; Rott et al. 2003).

1.2 Taxonomy of Indicator Species from Different Environments

Diatoms are ecologically diverse and extensively distributed in both fresh and saline habitats. There are diatom species that are very tolerant with a wide ecological valence, yet other species have tolerance levels that are distinct and narrow optima for many environmental variables; these attributes enable them to be remarkably applied in quantifying environmental features with great precision (Dixit et al. 1992). Excess nutrient loading and organic contamination have been regularly monitored using diatoms and indices of various types developed to quantify the quality of water (Rott et al. 2003). Some of the extreme pollution-tolerant species are *Navicula atomus*, *Nitzschia palea*, *Gomphonema parvulum*, *Navicula cryptocephala*, and *Navicula minima*, and species sensitive to extreme pollution are *Achnanthes biasolettina*, *Cocconeis placentula*, and *Gomphonema minutum*. Heavy metal pollution can result in cell wall deformities and loss of diversity caused which are useful indicators to monitor heavy metal pollution (Walsh and Wepener 2009).

Intrinsic silica patterns on diatom frustule make diatoms unique in terms of taxonomic identification up to strain level compared to other algal species. Species diversity and biomass in terms of bio-volume are the two main criteria which are based solely on diatom-based monitoring. Sampling habitat plays a significant role in effectiveness of biomonitoring. Sampling of rocks and hard surfaces is recommended in the European Union (Kelly et al. 1998), whereas in US programs random sampling of any available substrate is recommended (Weilhoefer and Pan 2007). Species composition and biomass in terms of cell bio-volume are two of the key parameters on which diatoms can be differentiated from other algae and microbes (Table 2).

1.3 Diatom-Based Water Quality Indices

Water chemistry significantly influences diatom assemblage communities. Diatom development and structure respond extensively to eutrophication, organic pollution, fluctuations in conductivity and pH, and elevated levels of sediments suspended in water. Most researches have documented relationships between concentration of nutrients and assemblage of diatom communities and likened a high amount of the community difference proportionally to the recorded nutrients in the water bodies

Table 2 Indicator species of diatoms for different physiochemical parameters of wastewater

Species	Indicator for
Nitzschia palea	Organic pollution
Nitzschia fonticola	Organic pollution
Fragilaria capucina	Heavy metal
Achnanthidium minutissimum	Heavy metal
Fragilaria ulna var. *acus* (*Kütz.*) *Lange-Bert*.	High conductivity
Eunotia sp.	Eutrophication
Diatoma vulgare	Eutrophication
Eunotia exigua, *Gomphonema angustum*	pH
Amphora veneta, *Gomphonema rautenbachiae*	pH
Melosira nummuloides	Salinity
Melosira sp.	Flow rate
Cocconeis sp.	Flow rate
Gomphoneis herculeana, *Achnanthidium* sp., *Achnanthes subhudsonis* var. *kraeuselii*	Eutrophication
Gomphoneis herculeana, *Achnanthidium* sp., *Achnanthes subhudsonis* var. *kraeuselii*	Low total phosphate (TP)
Luticola goeppertiana, *Navicula recens*, *Nitzschia inconspicua*, *Nitzschia palea*, *Rhopalodia* sp., *Eunotia* sp.	High TP
A. minutissimum, *Cocconeis placentula*, *Surella construens*, *Sorella pinnata*	Nitrogen (autotrophic)
Gomphonema parvulum, *Eolimna minima*, and *Nitzschia palea*	Nitrogen (heterotrophic)
Navicula sp., *Nitzschia* sp., *Surirella* sp.	Silt tolerant
Eunotia sp., *Pinnularia* sp.	Acidic pH

(Torrisi et al. 2010). Others, on the other hand, have noted significant correlations among type of substrate, dissolved oxygen, and alkalinity (Blinn and Herbst 2003).

Lake classification based on algae is well documented in the literature (Stoermer 1978), which are listed in Table 3. Many classification systems employ diatoms to assess the water quality (Hecky and Kilham 1973; Carpelan 1978).

1.4 Studies on Water Quality Monitoring Using Diatoms

Anthropogenic pollution of surface waters in many countries has led to increased stress on water ecosystems. To understand and monitor its effect, we need to place more emphasis on developing trophic variables. In some European countries, several diatom-based indices have been employed and are being used routinely (Prygiel et al. 1999). The European Water Framework Directive (WFD) (Bennion and Battarbee 2007) has encouraged the application of ecological studies to understand the impact of anthropogenic pollution on fresh water ecosystems (Muxika et al. 2007). The WFD mandates the use of ecological monitoring of rivers and lakes based on biological indicators like microalgae, fish, invertebrates, macrophytes, etc. of which diatoms are most commonly used species (King et al. 2006). In Latvia, Furse et al. (2006) reported that diatom-based diversity indices correlated strongly with environmental variables when compared with macrophytes and fish. With conversion of community response to a particular gradient into a continuously monitored variable by using diatoms, we can simplify ecological monitoring of water bodies. Studies related to effects of eutrophication have shown that diatom metrics detect eutrophication more efficiently than other metrics studies (Hering et al. 2006). All these studies providing strong evidence of usefulness of diatoms as

Table 3 Diatom-based water quality indices

Abbreviation	Nomenclature	Reference
CEE	Commission for economical community metric	Descy and Coste (1991)
DESCY	Descy's pollution metric	Descy (1979)
EPID	Pollution metric based on diatoms	Dell'Uomo (1996)
IBD	Biological diatom index	Prygiel et al. (1999)
IDG	Generic diatom index	Prygiel et al. (1996)
IDAP	Indice Diatomique Artois-Picardie	Prygiel et al. (1996)
L&M	Leclercq and Maquet's pollution index	Leclercq and Maquet (1987)
ROOT	Trophic metric	Rott et al. (1999)
SLAD	Sla'decek's pollution index	Sládeček (1986)
TDI	Trophic diatom index	Kelly and Whitton (1995)
WAT	Watanabe et al. pollution metric	Watanabe (1988)
ABSS	Abundance of reference taxa	Delgado et al. (2010)

bioindicators resulted in their increased use as tools for efficient monitoring of water quality (Gómez and Licursi 2001; Wu and Kow 2002).

Although several authors (Stoermer and Yang 1970; Tilman et al. 1982) showed that diatoms are useful indicators of water quality, still the development of new indices is necessary for many geographic locations before their widespread application in monitoring studies. The development and wide use of software packages, such as Omnidia, which facilitates calculation of indices, is quite helpful (Eloranta and Soininen 2002; García et al. 2008). In North America, the use of diatom metrics based on sensitive and tolerant species is more widespread (Fore and Grafe 2002; Passy et al. 2004).

2 Role of Diatoms as Bioindicator in the Performance Evaluation of Constructed Wetland

Constructed wetlands are ecological systems which are influenced by a combination of physicochemical and biological processes. In order to maintain them, a balance between these processes is paramount. In a constructed wetland, ecological food web consists of planktonic and benthic algae, bacteria, and other higher trophic organisms, but the majority of the primary productivity is fueled by sunlight and nutrients available in influent wastewater. Algae are a part of any wet habitat, and they are an integral part of any wetland ecosystem. Many different species of algae inhabit CWL depending on the type of vegetation. In CWL many types of vegetation are promoted depending on the design like free floating, rooted floating, submerged aquatic, emergent aquatic, and shrubs. All these different vegetation techniques are used in the presence of wastewater at different depths; this leads to a congenial environment for benthic diatom algae which grow as colonies on submerged substrates and include epiphytic, epipsammic, epipelic, and epilithic forms. Structure and productivity of benthic diatom community is influenced by nutrient loading into CWL (Gaiser et al. 2014). So by monitoring the dynamics of diatoms on submerged and emerging plants and other substrates, we can access water quality and treatment efficiency of a CWL, and it can be a suitable alternative for physiochemical analysis to evaluate wetland performance and evaluation.

2.1 Assessment of Wastewater Characteristics Through Diatom Species Diversity

Using diatoms as indicators of wastewater quality can be attributed to their presence in diverse ecosystems, sensitivity to changes in nutrient and environmental conditions, and easiness to access their diversity. Their significance in water ecosystems is linked to their primary role in aquatic food webs and biogeochemical cycle (Lamberti 1996; Mulholland et al. 2008).

Diatoms are diverse species which are present in all wetland ecosystems throughout the world. They show dynamic sensitivity response to different range of water pollution. Their fast growth rate enables them to inhabit new habitats in rapid time which makes their species monitoring ideal for studying their response to environmental change. Their fast response in terms of species diversity and abundance gives them a competitive advantage over physicochemical sampling, where sudden spike in a parameter can lead to ecological significant fluctuations which cannot be monitored over time. Benthic diatoms are attached to substrates so they are confined to particular habitats with specific physiochemical characters which make them ideal for biomonitoring of those environments (De la Rey et al. 2004). The species-specific response of diatom to varied conditions can be studied by their increase in biomass and species diversity (Patrick 1961). Benthic diatoms have been increasingly used to monitor physiochemical changes such as pH, conductivity and organic nutrients, eutrophication, and global warming problems.

2.2 Importance of Diatoms in Water Quality Management and Natural Food Production in Aquaculture

Microalgae are major contributors to nutrition in natural marine and fresh water ecosystems and in aquaculture. Being major primary producers in oceans, diatoms are major food for many marine invertebrates. Diatoms also contribute as natural food in intensive aquaculture systems by forming the base of aquatic food web. Dissolved oxygen (DO) is one of the major factors influencing aquatic animal's metabolism and growth. Decreased dissolved oxygen in intensive aquaculture systems is a serious concern. Artificial aeration using electrical aerators adds additional costs and risks. By growing diatoms we can increase the DO levels in ponds very rapidly, and as diatoms move in the water column depending on light requirement, DO increase will be achieved even in the middle and bottom of the ponds which is not the case when we use mechanical aeration.

In aquaculture ponds, some phytoplankton species are considered undesirable, especially blue-green algae (BGA), sometimes called cyanobacteria, and are particularly troublesome. Due to their higher light requirement, they always grow as mats on top of the water column resulting in decreased mixing of atmospheric gases into the water leading to less DO. BGA also produce smelly compounds which give off odor to cultured organisms which can result in poor meat taste. Some BGA species like *microcystis* produce toxins that can kill fish and shrimp. By growing diatoms we can efficiently reduce the problems associated with BGA growth. BGA mainly dominate the ponds when there is high nutrient content and high pH; by growing diatoms which can utilize nutrients much faster than BGA and also help in lowering pH by maintaining water quality, we can eradicate BGA growth in aquaculture ponds.

Aquaculture ponds contain bacteria and viruses that can infect cultured organisms and thus potentially devastate aquaculture farms. These same Bacteria and viruses can also infect diatoms, so diatoms have developed self-defense mechanisms to protect themselves like secretion of compounds that inhibit bacterial growth or viral attachment so by transferring this compounds to feeding animals and also some species of microalgae especially diatoms grow on surface of the fish and shellfish there by it induces immunity to many harmful water born bacterial and viral pathogens in shrimp, fish and shell fish.

Diatoms possess many advantages as natural food in aquaculture. Their size and shape are ideal for ingestion and easy digestion; their biochemical composition is ideal for culture species and zooplankton with the right amount of carbohydrates, proteins, and fats. Diatoms also provide many phytonutrients like PUFAs – e.g., EPA, arachidonic acid (AA), and DHA.

3 Potential to Treat Effluents from Constructed Wetlands

The role of algae as primary producers and in nutrient cycling of wetlands is well established (Wu and Mitsch 1998). Diatom assemblages are increasingly used in lake bio-assessment and paleolimnological studies (Dixit et al. 1992). Weilhoefer and Pan (2007) found that diatoms growing on submerged macrophytes, sediment surface, and in the water column in wetlands are ideal for diatom-based wetland bio-assessment. Although much research is focused on using diatom-based biomonitoring of wetlands, their importance in mitigation of eutrophication through excess nutrient removal and natural oxygenation through photosynthesis is not well explored.

Phycoremediation using microalgae was considered as one of the effective ways to deal with water pollution because it causes no secondary pollution and has high efficiency and low cost (Olguın 2003). Furthermore, microalgae have the ability to use inorganic nutrients (N and P), metabolize organic compound, and remove heavy metals and toxic organic compounds, which were then converted into biomass (Renuka et al. 2015). The biomass may be harvested and used in various applications, which then assist in purification of the wastewater, besides reducing the biochemical oxygen demand (BOD), resulting from biodegradation of the dead cells in the treated water.

Wetlands are ideal environments to mitigate nutrient-enriched surface waters. Denitrification process in wetlands was known to happen at sediment interphase (Payne 1991). But recent research has shown that maximum denitrification rate was achieved in upper 3–5 cm of wetland sediment which is dominated by periphyton attached to natural substrate especially dominated by diatoms (Eriksson and Weisner 1997). Ishida et al. (2008) studied the potential relation between the algal community structure and bacterial cell densities and denitrification rates and found that elevated denitrification rates were found in periphyton with high relative diatom concentration but not with green or blue-green algae. Diatoms also contributed to increase in bacterial density; this might be due to specific relationship between diatom and bacterial community structure.

3.1 Effect on Dissolved Oxygen Concentration

Dissolved oxygen is one of the key factors which influence the survival rate of not only cultured organism but also aerobic bacteria in aquatic ecosystems. Due to high amount of oxygen in the atmosphere (21%–300 mg L^{-1} of air), terrestrial organisms rarely experience its depletion. But in aquatic environments, solubility of oxygen is less than 1% of its solubility in the air. This amount of oxygen solubility in water depends on factors like pH, temperature, and surface area. At an atmospheric pressure of 1, saturated DO concentrations can reach a maximum of 9 mg L^{-1} at 20 °C which is much less when compared with its concentration in the air (Wetzel 1981).

The significant O_2 generation from algal photosynthesis can offset the cost incurred by wastewater treatment plants and aquaculturists for mechanical aeration (Mallick 2002). Oxygenation due to algal photosynthesis in oxidation ponds facilitates enhanced breakdown of organic and inorganic compounds by aerobic bacteria (Munoz and Guieysse 2006). Algal photosynthesis provides dissolved oxygen for aerobic bacteria, while the bacteria provide carbon, nitrogen, and phosphorus needed by algae for growth. The technique has been widely utilized in treating agricultural, municipal, and industrial wastewater. Algae growth especially blue-green algae which grow on water surface can hinder light penetration and gaseous exchange in ponds with submerged vegetation leading to hindered growth and lower DO levels.

3.2 Residual Nutrient Removal Efficiency

Diatom algae can dominate under nutrient-limiting and excess conditions; it is shown that diatom species outcompete non-nitrogen-fixing cyanobacteria under low nitrogen concentration in a eutrophic lake (Amano et al. 2012). Enhanced carbon fixation ability and concomitant nutrient removal capability increase the applicability of diatoms for CO_2 mitigation and wastewater treatment. Diatom algae produce oxygen during photosynthesis which acts as stimulant for heterotrophic bacterial growth which in turn can enhance bacterial degradation of organic pollutants (de Godos et al. 2010). Growth of benthic diatom *Nitzschia* sp. has resulted in enhanced aerobic bacterial activity in sediment layer which can lead to accelerated decomposition of organic matter (Yamamoto et al. 2008). Phthalate acid esters (PAEs) are commonly occurring priority pollutants and endocrine disruptors. Marine benthic diatom *Cylindrotheca closterium* has shown increased PAE removal rate in surface sediments. In bottom sediment it helped in increase of aerobic bacterial growth by photosynthetic oxygen, thereby resulting in a combination of bacteria-diatom-dependent PAE removal (Li et al. 2015). Diatom *Stephanodiscus minutulus* under optimum nutrient availability has shown increased uptake of PCB integer 2,2′,6,6′-tetrachlorobiphenyl (Lynn et al. 2007). Polyaromatic hydrocarbon (PAH) phytoremediation has limited success rate due to their high toxicity, but diatoms *Skeletonema costatum* and *Nitzschia* sp. have shown accumulation and degradation of phenanthrone (PHE) and fluoranthene (PLA), two typical PAHs (Hong et al. 2008). Diatom algae-produced O_2 can help in bacterial degradation of PAHs, phenolics, and organic solvents in benthic environments. Diatom *Amphora coffeaeformis* is known to accumulate herbicide mesotrione (Valiente Moro et al. 2012). The potential of diatom algae in biodegradation and accumulation of pollutants is enormous, but till date little research is done in this field.

3.3 Role in Pathogenic Bacteria Removal

Phytoplankton and bacteria have coexisted in the environment for millions of years. There exists a positive and negative allelopathic interaction between the both. Microalgal photosynthesis can enhance pathogen removal by changing the water physical parameters like increased pH, dissolved oxygen, and temperature (Ansa et al. 2011). Diatoms develop natural defense mechanism to protect against bacteria which can harm them and are often harmful to humans and animals also. Effective control of some harmful bacteria can be achieved, if we can grow natural diatom populations in wastewaters which have innate defense mechanism to control their growth. Diatoms secrete volatile and nonvolatile substances like fatty acids, esters, and polysaccharides as antibacterial compounds to control their growth (Lebeau and Robert 2003). Many of these hydrophobic molecules act as deterrents to bacteria by disrupting their cell signaling mechanisms during their adhesion to diatom cells. In a study on diatom *Navicula delognei* by Findlay and Patil (1984), fatty acids and sterols have shown strong antibacterial effect against pathogens like *Staphylococcus epidermidis*, *Salmonella enterica*, etc. Diatom *Phaeodactylum tricornutum*-produced eicosapentaenoic acid (EPA) has shown to inhibit gram-positive bacteria (Desbois et al. 2009). The same diatom has also shown inhibitory effect on multiresistant staph aureus (MRSA) (Desbois et al. 2009). *Chetoceros* sp. a marine planktonic diatom when maintained at higher concentration in the aquaculture ponds has lead lowered pathogenic bacteria like *Vibrio vulnificus* and simultaneously reduced propagation of viruses in shrimp production system. Diatom *Skeletonema costatum* was shown to inhibit *Vibrio*, a pathogen of fish and shellfish (Naviner et al. 1999). Many pathogenic bacteria are anaerobes which cause many respiratory, digestive, and urinary tract infections which are waterborne. Walden and Hentges (1975) have shown that anaerobic pathogenic intestinal bacteria growth was inhibited in the presence of oxygen; so to counter this, many anaerobes grow at oxygen-deficient zones especially in the sediment layer of wastewater ponds. Mechanical aeration cannot provide enough oxygen to these zones leading to proliferation of harmful bacteria. This can be reversed if we can promote diatom growth in these ponds with high sediment accumulation as benthic diatoms are known to produce high amount of oxygen even inside the sediment leading to aerobic zones.

4 Diatom-Based Excess Nutrient Removal from Eutrophic Water Bodies

Diatoms can be grown using agricultural and municipal wastewater. Wastewater contains macronutrients like nitrate, phosphate, silica, and other trace metals which are essential for algal growth. Hence growing algae in wastewater can be economically and environmentally beneficial as it can lead to decreased water treatment cost with an option of generating value added (Oswald 1988). The combination of three roles of microalgae in CO_2 mitigation, wastewater treatment, and biofuel production has the potential to decrease the use of fresh water for biofuel production and on climate change through CO_2 removal; however many crucial challenges like isolation of algal strains with high growth and nutrient uptake, integration of algal growth system with wastewater treatment systems, improved algal harvesting, and life cycle analysis are to be further explored to maximize the enormous potential of algal biofuels. Benthic diatoms are the dominant algal community in wastewater bodies, and they contribute significantly to nutrient removal and primary productivity in water.

Any wastewater treatment plant had to remove high concentrations of N and P present; if not treated this will cause eutrophication to downstream waterbodies. P is very difficult to remove in a conventional STP as there are very few phosphorus-removing bacteria present than nitrate-removing bacteria, so it is primarily removed by chemical precipitation which cannot be recycled. Algae-based treatments are more efficient in removing excess P from wastewater than chemical treatments. Microalgae especially diatoms are efficient in utilizing N and P along with other metals present in wastewater for their growth through photosynthesis and play a significant role in excess nutrient mitigation. Furthermore, an algae-based bioremediation is more environmentally amenable and sustainable as it does not generate additional pollutants such as sludge; resultant algae biomass rich in nutrients can be used as low-cost fertilizer or as animal feed (Munoz and Guieysse 2006).

4.1 Diatom Physiological and Morphological Advantages for Efficient Nutrient Removal

Silica cell wall plays a significant role in carbon-concentrating mechanism (CCM) with diatom bio-silica acting as an effective pH buffer enabling increased carbonic anhydrase activity near cell surface which enables conversion of bicarbonate to CO_2

(Milligan and Morel 2002). Silica cell wall gives diatom algae an advantage of enhanced sinking rate which results in increased carbon burial in shallow seas and continental margins (Falkowski et al. 2005) and are major contributors to nascent petroleum reserves.

Diatoms possess larger storage vacuole compared to other algae which is one of the main factors for their dominance in oceans (Raven 1987). Nutrient utilization which is an important factor influencing growth is dependent on surface to volume ratio were smaller cells have an advantage but diatoms with their large storage vacuole can store nutrients inside the cell thus nullifies this factor even with large surface area. Thus in nutrient replete conditions, diatoms store nutrients, this enables them to perform several cell divisions even in deplete conditions, and this will further influence their dominance by preventing other algae to grow. Diatom algae consistently achieved growth rates in the range of two to four divisions per day which is much higher than other algae tested with the same size (Furnas 1990). Diatom algae can dominate other eukaryotic algae even under high turbulence, and mixing this makes them ideal for mass culturing under varied mixing regimes.

Diatom carbon fixing ability is greater than other algal groups in terms of productivity per unit of carbon. In comparison with *Chlorella vulgaris*, diatom *Phaeodactylum tricornutum* has shown two times more efficiency in converting light energy into biomass. This shows that diatoms have higher photosynthetic efficiency in low light conditions when compared with green algae (Smetacek 1999).

Diatoms lack α-carotene biosynthetic pathway which enables them to produce photo-protective and light harvesting pigments from the same precursors (Wagner et al. 2006). Diatom can perform both C3 and C4 biochemical fixation with a complete urea cycle (Armbrust et al. 2004). Diatoms store carbohydrate in the form of chrysolaminarin which is a soluble form of carbohydrate, whereas other classes of algae store in the form of starch in chloroplast. Although diatoms are not efficient in storing carbohydrates, relative energy required to utilizing soluble carbohydrate stored in CV to unsoluble carbohydrate stored in chloroplast is less (Hildebrand et al. 2012).

Diatoms synthesize their frustules with silica. The source of silica for diatoms is dissolved silicic acid which is absorbed in low quantities by silicic acid transporter proteins. The energy required to build silica cell wall is much less when compared with lignin or polysaccharide cell wall; this will also help in carbon saving as carbon in cell wall is replaced by silica and the carbon replaced is used for other cellular functions (Raven 1983). All these significant differences in cell structure and function might have contributed to the dominance of diatoms.

4.2 Studies on the Use of Diatoms for Different Wastewater Treatment

Integrating municipal wastewater treatment with microalgal cultivation can be a sustainable option for the existing STPs as it can reduce the high-maintenance costs and input cost for civil construction. Municipal wastewater contains ammonia, phosphate, and other essential nutrients which are required for microalgal growth. Over the past decade, many studies have been done on growing microalgae on different types of wastewaters like domestic wastewater, agricultural runoff, dairy wastewater, and industrial and municipal waste streams, and the success of these studies was dependent on biotic and abiotic factors. Majority of these studies concentrated on the use of green and blue-green algae, but in recent times, diatoms are increasingly recognized for their phycoremediation potential (Table 4).

In the 1950s, Oswald designed large-scale algae-based open pond systems called high-rate algal pond (HRAP). Algae photosynthesis was used to fulfill the oxygen demand to treat domestic wastewater which was a very efficient system for wastewater treatment (Olguın 2003). HRAP are shallow open ponds; under optimum

Table 4 Nitrogen and phosphorus removal efficiency of microalgae grown using different wastewaters

Algal strain	Wastewater source	Treatment time	Removal efficiency (%)		Reference
			Total nitrate (TN)	Total phosphate (TP)	
Chlorella vulgaris	MWW[a]	09	78	87	Ruiz-Marin et al. (2010)
Scenedesmus dimorphus	IWW[b]	08	70	55	González et al. (1997)
Scenedesmus obliquus	MWW	08	79	47	Ruiz-Marin et al. (2010)
Arthrospira platensis	IWW	15	96	87	Phang et al. (2000)
Oscillatoria sp.	MWW	14	100	100	Craggs et al. (1997)
Phaeodactylum tricornutum	MWW	14	100	100	Craggs et al. (1997)
Mixed culture	DWW[c]	15	96[d]	99	Woertz et al. (2009)
Mixed (diatom dominance)	MWW	23	82	88	Marella et al. (2015)
Diatom consortium	MWW	07	91	88	Marella et al. (2018)

[a]MWW – municipal wastewater
[b]IWW – industrial wastewater
[c]Dairy wastewater 25% dilution
[d]Total ammoniacal nitrogen (TAN)

conditions BOD removal rates were as high as 3500 mg m^2 d^{-1} with hydraulic retention time of 4–10 days. A modified version of this was advanced integrated wastewater pond systems (AIWPS) which are a series of facultative, settling, and maturation ponds. Diatoms can be harnessed for tertiary treatment for enhanced nitrogen and phosphorus removal. Diatoms utilize N and P thorough biotic and abiotic process. Diatoms incorporate N and P into their biomass in the form of protein, nucleic acids, and phospholipids, whereas the increased pH due to their photosynthesis will enhance ammonia and phosphate volatilization and precipitation.

In aquaculture, artificial feed and fish waste enrich the water with excess nutrients leading to unwanted BGA blooms which are detrimental to culture organism growth. Diatom *P. tricornutum* has shown 30–100% removal of ammonium and orthophosphate in batch and continuous modes using diluted effluent (Craggs et al. 1995). Diatom-dominated biofilms grown on artificial substrates in shrimp ponds led to 33% phosphate removal. The diatom-dominated biomass from these treated biofilms can be used as natural feed for filter feeding fish and bivalves. This fish- and bivalve-based aquaculture system could be effective to reduce cost of water treatment with simultaneous production of natural feed.

5 Other Applications

In spite of their dominance in world's oceans combined with their tremendous diversity and tropic flexibility compared with other algae, they are the least explored species for biotechnological applications. Most studies have focused on polyunsaturated fatty acids like eicosapentaenoic acid (EPA) and decosahexanoic acid (DHA) which is used for pharmaceutical applications. Applications for other molecules like amino acids for cosmetics, antioxidants, antibiotics, and antiproliferative agents are at the early stage of development (Lebeau and Robert 2003).

Diatom algae contain very interesting bioactive compounds which are highly sought after in pharmaceutical and nutraceutical industries. Diatoms are rich source of pigments, lipids, sterols, hydrocarbons, phenolic compounds, polysaccharides, alkaloids, and toxins with high bioactivity. Although diatoms contain a variety of active compounds, previous literature is predominantly dedicated to PUFA especially EPA.

Fucoxanthin is a major light harvesting pigment and carotenoid present in seaweeds and diatoms. Fucoxanthin is known to show strong antioxidant, anti-inflammatory, anti-obesity, antidiabetic, anticancer, and antihypertensive activities (Abidov et al. 2010). At present the main commercial source for fucoxanthin is seaweeds, but they have major drawbacks like slow growth, less fucoxanthin content, and contamination by heavy metals. In comparison diatoms contain fucoxanthin in the range of 0.2–2% of dry weight which is 100 times more than that of brown seaweed which is a primary industrial source (Kim et al. 2012).

EPA which is an omega-3 polyunsaturated fatty is de novo synthesized in diatoms. These are the richest primary sources of EPA. The major dietary sources of EPA and DHA for humans are fatty fishes, but advantage of diatom-derived EPA is that it will be a vegetarian source of nutritional fatty acid. Pennate diatom *Phaeodactylum tricornutum* which can accumulate high levels of EPA is presently explored as a potential source for its industrial production.

Microalgal fatty acids are an integral part of animal nutrition; as higher organism cannot synthesize polyunsaturated fatty acids; they can only acquire them through food (Yongmanitchai and Ward 1989). EPA (20:5 (n-3)) and DHA (22:6 (n-3)) are the two main PUFAs required by marine animals to maintain good growth and survival (Renaud et al. 1991).

Microalga as a source of fuel is gaining popularity. Every single microalgal cell can act as a lipid factory which is not the case with terrestrial plants which produce specialized oil-bearing organelles like seeds. Due to this unique ability of microalgae, they are targeted organisms for large-scale funding and scientific studies for biomass and bioenergy production.

6 Conclusions

Algae culture can be integrated within the present wastewater treatment facilities with no or little change to existing infrastructure. This approach will enable reduced capital, maintenance cost, and scalability issues with enhanced treatment efficiency. Although there is much research done on this aspect, research lacuna still exists in areas like photobioreactor design, harvesting technology, drying methods, and other downstream processes which if worked on can lead to effective commercial exploitation of this environmental energy-efficient technology.

Microalgal biotechnology especially for wastewater treatment has received more attention in recent years as a viable alternative to conventional wastewater treatment systems. Algal biomass produced during this process is a sustainable bioresource for biofuel, nutraceutical, biofertilizer, animal feed, poultry feed, and aqua feed industries. In spite of its attractiveness, there are still some obstacles to be solved for its mass-scale exploitation.

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