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Chapter 6

Phycoremediation of Eutrophic Lakes Using Diatom Algae

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Abstract

Eutrophication as a result of human intervention has led to severe deterioration of fresh water habitats. Due to population growth, industrialization and uncontrolled use of fertilizers led to excess nutrient runoff entering into rivers and lakes; this has caused reduction in water quality and abnormal changes in ecosystem structure and function. A solution to this cultural eutrophication is an urgent necessity since nutrient accumulation renders controlling eutrophication more difficult over time. Using algae for reduction of nutrients is a unique technology, which utilizes the enormous potential of microalgae in restoring water quality. This has a huge potential in urban lakes where there is an urgent need to use such technologies in combination with existing ones to speed up the process to reduce the formation of hypereutrophic lakes and dead zones in oceans. In this book chapter, we explore the enormous potential of diatoms as cost-effective, efficient and eco-friendly remedy for complex problems related to eutrophication. We report the case studies on using diatom-based technology. This will give us a new insight into microalgae-based lake remediation strategies, which can significantly reduce the cost, manpower needed and negative environmental impacts involved in existing technologies.

Keywords: diatoms, eutrophication, phycoremediation, Nualgi

1. Introduction

Many human activities are polluting freshwater ecosystems, modifying the structure of aquatic communities and thereby disrupting the functional continuum of river and lake systems. Indeed, anthropogenic pollution of freshwater ecosystems by the addition of organic matter and

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nutrients is an increasing phenomenon that affects many lakes and rivers worldwide [1]. Physicochemical alterations caused by contaminated water and biological wastes often involve increases in inorganic nutrients (ammonium, nitrate, nitrite, phosphate) and suspended organic solids, a decrease in dissolved oxygen, and a settlement of suspended organic matter settling on the lake bottom. Therefore, there is an urgent need to explore new eco-friendly, cost-effective strategies to mitigate nutrient input into waterways and also to remove nutrients from waterways.

Nutrient contamination of surface waters has led to widespread excessive algae growth, a process known as eutrophication. Eutrophication can lead to fish kills through oxygen depletion or the growth of toxic dinoflagellates that produce neurotoxins harmful to fish and humans [2–4]. Eutrophication also can cause taste and odour issues that create expensive problems for municipalities that rely on surface water for their drinking water and individual households depending on groundwater [1].

Benthic diatoms are the dominant algal community in water bodies and they contribute significantly to nutrient removal and dissolved oxygen levels in water. They also form the basis of benthic food web in water bodies. Diatom algae contribute up to 40% of primary production in lakes and oceans, which is more than that of all the tropical rain forests on earth. Diatoms play an important role as a major carbon carrier to Deep Ocean to be one of the major contributors to the "biological carbon pump".

Diatoms are microscopic plants, which use nitrates and phosphates to grow along with other nutrients such as silica, iron, copper, molybdenum, etc.; they use CO_2 and produce O_2 and they can also accumulate heavy metals, so by triggering the growth of these algae, many problems related to lake pollution can be solved. Growth of diatoms also reduces the growth of harmful algae such as blue green algae (BGA) [5]. Diatoms are important primary producers in streams, lakes and wetlands [6]. The main source of energy in streams was once thought to be detritus from terrestrial origin but later research showed that primary production by algae was important in many streams. Diatoms are now predicted to be the primary energy source in many streams [7]. Diatoms are also known to be important sources of energy for invertebrates in some headwater streams and even dominant, primary producers in many shallow lakes and ponds. In wetlands, diatoms are significant primary producers because of their high turnover rate. In addition to primary producers, diatoms are chemical modulators in aquatic ecosystems [8]. They transform many inorganic chemicals into organic forms. Diatoms are primary harvesters of inorganic phosphorus and nitrogen in stream spiralling in lake littoral modulation of influxes and in wetlands. Diatoms on surface sediments and plants are considered to be important sinks for nutrients before release into the water.

Diatoms as indicators of lake water quality were well studied by many researchers but diatoms also play a significant role in maintaining the water quality [9–11], so using diatom algae for nutrient removal is novel and cost-effective method of water treatment. The main bottleneck in using only diatom for nutrient removal is to trigger only diatom growth instead of other algae, so to solve this problem, the main solution is to use the silica as the nutrient, which is absolutely required by diatoms for their growth.

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Phycoremediation is defined as the use of algae to remove pollutants from the environment or to render them harmless [12]. Phycoremediation has evolved from the early work done by Oswald and Gotaas [13] for the use of microalgae for tertiary treatment of municipal wastewater to many other applications in which microalgae are cultivated and utilized for specific bioremediation needs. The use of microalgae for the treatment of municipal wastewater has been the subject of research and development for several decades.

Considering excess nitrate and phosphate as a resource and not as a pollutant is a key in unlocking the problems related with nutrient pollution. Solar energy can be harnessed to grow algal biomass on wastewater nutrients and this biomass can be a source on which fish can be grown; this could provide a holistic solution to nutrient management problems. Nutrient runoff from agriculture and sewage constitutes a major component of wastewater generated everyday in both developed and developing nations such as United States and India. Excess N and P fertilizers and untreated or partially treated sewage find its way to the water bodies resulting in enrichment of nutrients, leading to eutrophication and formation of dead zones. One of the highly significant and rapidly developing methods for nutrient removal is the use of photosynthesis by growing algae. The use of algae for municipal wastewater treatment in ponds is well established [14, 15]. Algae growth in wastewater treatment ponds contributes to treatment mainly through dissolved oxygen production and nutrient assimilation. The release of oxygen from water during photosynthesis provides aerobic microbiological waste oxidation and the absorption of carbon dioxide which also accompanies photosynthesis.

The use of microalgae for the treatment of municipal wastewater has been the subject of research and development for several decades. In the early1950s, the first research on using microalgae for wastewater treatment was started. It was demonstrated that algae-based wastewater treatment could remove the nutrients (e.g. N and P) from settled domestic sewage more efficiently than traditional activated sewage process, indicating a great potential of algae-based wastewater treatment system. The result of such effort is that some commercial technologies and processes are available in the market such as the Advanced Integrated Wastewater Pond Systems (AIWPS) Technology commercialized by Oswald and Green [16], LLC, in the United States. Sewage contains mainly N and P, so this technology of Oswald's will aptly suite for excess nutrient removal in any water body but the main obstacle is to grow a particular type of algae in a controlled way. This can be achieved by using our technology.

1.1. Phycoremediation

Phycoremediation in a much broader sense is the use of macroalgae for the removal or biotransformation of pollutants, including nutrients and xenobiotics from wastewater and CO_2 from air. Algae can fix carbon dioxide by photosynthesis and remove excess nutrients efficiently at minimal cost; in addition, photosynthetically produced oxygen can relieve biological oxygen demand (BOD) in waste water.

Phycoremediation comprises several applications such as oxygenation of the atmosphere, nutrient removal from municipal wastewaters and effluents rich in organic matter, nutrient and xenobiotic compounds removal by biosorption using algae, treatment of acidic and metal

wastewaters, CO_2 sequestration, transformation and degradation of xenobiotics and biosensing of toxic compounds by algae.

1.2. Diatoms and aquatic ecosystems

Phytoplankton community composition is highly dependent on the quantity and ratio of macro- and micronutrients in aquatic ecosystems. There are many examples of taxonomic shifts due to the relative supply of silica (Si) versus other nutrients (e.g. nitrogen (N) and phosphorus (P)). Bacillariophytes, or diatoms, are fast-growing phytoplankton that utilize dissolved silicate (SiO_4) to make their siliceous-armoured skeletal frustules [17]. In marine systems, diatoms require a particulate cell N/Si ratio of ~1 for balanced growth. Other phytoplankton species, such as dinoflagellates, cyanophytes, haptophytes and raphidophytes, do not utilize Si. If silicate is limiting, these other phytoplankton are capable of outcompeting diatoms despite generally slower growth rates [18]. Therefore, by 'fertilizing' waters those are depleted in Si relative to other macronutrients, such as with high Si-content solutions, the potential exists to shift the phytoplankton community to diatom dominance.

Diatoms are a widespread, diverse group of microalgae found in all aquatic systems. They represent a major component at the base of the marine food web, responsible for up to 50% of total lake and oceanic primary production [7, 19] and 25% of all oxygen produced on the planet. It also absorbs 23.5% of carbon dioxide generated on the planet. Diatoms can be found from the poles to the tropics, vary in size (2–200 μm), shape (centric, pennate), and can exist as single cells, colonies or chains [20]. Diatoms are opportunistic, generally exhibiting high growth rates and blooming rapidly when nutrient and light conditions are favourable [21]. Similarly, blooms can end as quickly when the diatoms have utilized all available nutrients and are either grazed upon (supporting higher trophic levels) or sink rapidly (driving the carbon pump). Diatoms require less light than other algae [22]; since their silica shells are transparent, they grow even on cloudy or rainy days. Diatoms can even dominate under nutrient-limiting conditions; in one example, a diatom species was shown to outcompete non-N-fixing cyanobacteria under low nitrate concentrations in a eutrophic lake [23]. These factors make most species of diatoms effective nutrient "sponges". This combination of the diatom's abilities makes them an ideal organism for water remediation practices.

1.3. Diatom algae for nutrient removal

Growing microalgae/phytoplankton such as diatom algae in the sewage will enable the nutrients in the sewage to be consumed and the oxygen produced will satisfy the BOD and chemical oxygen demand (COD) and provide oxygen to fish. Phytoplankton is the natural food for fish and diatoms are the best group of phytoplankton. Thus, polluted lakes will become clean and have plenty of fish. About 50% of the photosynthesis on Earth takes place in water—lakes and oceans and diatom algae account for about 50% of the algae in water bodies. Treated water with BGA and green algae cannot be released into public water bodies. Diatoms assimilate a significant amount of nutrients because they require high amounts of nitrogen and phosphorus for the synthesis of proteins (45–60% of microalgal dry weight),

nucleic acids and phospholipids. Nutrient removal can also be further increased by NH_3 stripping or P precipitation due to the rise in the pH associated with photosynthesis.

1.4. Advantages of treating waste water with diatoms

Diatom algae are the most prolific microalgae in nature and they grow in all ponds, lakes, rivers, oceans, aquariums, etc. They are responsible for about 20–25% of all photosynthesis on Earth [24]; this is more than the share of tropical rain forests (~18%) and agriculture (8%). Diatoms can consume N and P faster than other algae. Diatoms can consume all forms of N such as nitrate, nitrite, urea and ammonia. Diatoms are best sequesters of CO_2, so they can release more oxygen.

1.5. Is diatom-based phycoremediation important?

Diatoms have an absolute requirement for significant amounts of silicon. Total algal biomass cannot be limited by Si, but its availability will shape phytoplankton communities. Ryther and Officer [25, 26] suggest that the eutrophication of waters by domestic wastes relatively poor in Si could lead to Si depletion and the elimination of diatoms from the phytoplankton communities. This process has been documented in the Laurentian Great Lakes [27]. Centric diatoms have been classed as the most desirable phytoplankton in coastal and fresh waters because they are important in aquatic food chains, they do not form noxious surface blooms and they are not toxic. Marine diatoms often have high growth rates and some freshwater diatoms have been shown to outcompete other algal groups for both N and P when adequate Si is available [5, 28, 29]. Therefore, the availability of Si can make diatom consume N and P at a faster rate than other undesirable species such as blue green algae and flagellates. Dissolved silica becomes available in waters primarily through the weathering of silicate rocks. Domestic wastes have low concentrations of Si relative to N or P [25], and the relative proportion of Si to either N or P is very low compared to the requirements of diatoms and the relative abundance of these elements in natural water. The proportion in which nutrients are loaded to a system can exert a strong influence on which algal species will thrive [30]. In this regard, the apparent growing preoccupation with nitrogen and phosphate in the literature may be counterproductive. A balanced approach emphasizing the interplay of various nutrients including the trace metals in shaping phytoplankton communities and their response to enrichment is required. If we can manage the species composition of eutrophic systems to promote the growth of algal species such as diatoms that, in turn, increase the secondary productivity of valuable food species, then we will have solved the important nutrient limitation riddle.

1.6. Mechanism to trigger diatoms in open waters

Growing one type of algae in open waters is a complicated process, but diatoms have a distinctive advantage because of their absolute requirement for silica. Therefore, by using this advantage we can trigger diatoms in open waters especially in fresh water ecosystem where silica concentration is less. Taking this advantage into consideration, we have developed a nanosilica-based micronutrient mixture called "Nualgi", which has nanosilica as its major

constituent along with iron and nine other trace metals. The silica becomes both the carrier for other nutrients and the nutrient by itself. It is in a water-dispersible particulate form. Nualgi because of its nanosize is able to pervade very small spaces in the subsurface and remain suspended in water, allowing the particles to travel farther than larger, macro-sized particles there by increasing the bioavailability of the nutrients for easy absorption by microalgae and achieve wider distribution. In water, Nualgi causes diatom algae to bloom, though any pond, lake, estuary or coastal water has many species of organisms, only diatoms require silica and they consume Nualgi rapidly and bloom. In laboratory experiments with pure marine diatom cultures, highest biomass concentration and biomass productivity were attained in both *C. clostridium* and *C. fusiformis* in cultures grown in Nualgi containing medium; these values were almost double than in the popular f/2 medium [31] (**Figure 1**).

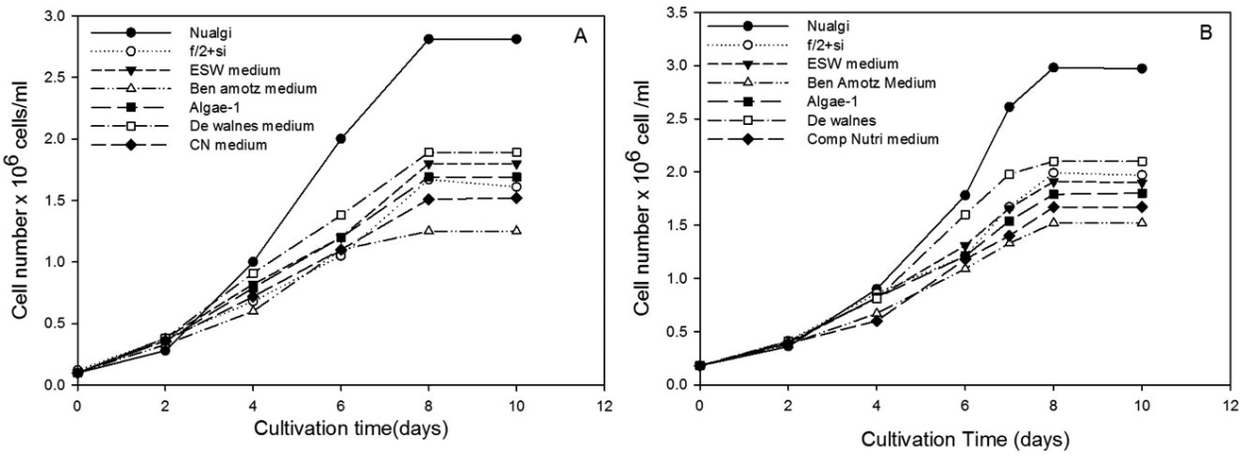


Figure 1. Effect of six different mediums on the growth of *C. clostredium* (A) and *C. fusiformis* (B) when compared with Nualgi-containing medium.

From Laboratory trails with eutrophic fresh water from Hussain Sagar Lake, Nualgi triggered diatom growth by not only triggering an increase in the number of diatom cells but also increasing diatom diversity. In samples without Nualgi, six different species of diatoms were identified in which *Cyclotella meneghiniana*, *Gomphonema lanceolatum* and *Nitzschia palea* were the abundant species, whereas in samples with Nualgi addition *Achnanthidium exiguum, Navicula cryptocephala, Cymbella tugidula, Navicula gracilis* and *Pleurosigma elongatum* were the dominant species with a total of 30 different diatom species. The dominant species in samples with no Nualgi addition were identified as pollution-tolerant species and in Nualgi-added samples dominant species are less pollution-tolerant species; this change in diatom species diversity clearly indicates the effect of Nualgi addition in nutrient reduction. In field conditions with Nualgi, N-removal percentage was 95.1% and P removal of 88.9% was achieved over a period of 10 days. COD and BOD reduction was also significant with 91 and 51%, respectively (Thomas, unpublished). Nualgi as an efficient tool to trigger diatom growth is well proven in both laboratory and field trials. Therefore, by using this product we want to propose a new

strategy to mitigate excess nutrients by growing diatom algae in large fresh and brackish water bodies [32].

1.7. Bioremediation versus phycoremediation

There is a general understanding that bioremediation is akin to phycoremediation. In actual, they are very different and bioremediation is at best a subprocess of phycoremediation.

In bioremediation, bacteria are dosed into the water bodies. These break down the organic matter in the sewage, dead algae and weeds. Bacteria consume oxygen, so aerators may have to be used to provide the oxygen required. The organics are removed to a certain extent but the dead bacteria sediment and accumulate. Thus, most of the nutrients remain in the lake. Bacteria are cultured and dosed periodically. This is quite expensive. In nature, bacteria help in the digestion of food even in human digestive system and in sewage treatment plants (STPs). This is being copied. Mechanical aerators result in the release of CO_2 at the power plants or from the diesel engines. Similarly, STPs produce sludge, which is difficult to dispose off.

In phycoremediation, nutrient enrichment with Nualgi causes the native diatoms present in all water bodies to grow. The oxygen produced by diatoms causes the native bacteria to grow and these work in the same way as the bacteria dosed in bioremediation solutions. Diatoms release pure oxygen during photosynthesis, resulting in increased DO levels; this will lead to cascading improvement in water quality, aquatic life and biodiversity. Diatoms are consumed by zooplankton and fish and thus exit the water as fish biomass. Very few diatoms die and fall into the lake bed. Diatoms grow with a small dose nutrients and since fish consume them, the income from the sale of fish will recover most of the cost of Nualgi used. The cost and quantum of dosage are much less than the dosing of bacterial strains under bioremediation. Phycoremediation restores the natural food chain in the lake and this is the best way to remove nutrients. The native bacteria break down the nutrients and diatoms help remove them from the water. Diatoms consume CO_2 and nutrients to release O_2. There is no waste generation.

1.8. Case study: phycoremediation of Indira Park Lake

1.8.1. Lake location and area

Indira Park is having 76 acres of area and it lies on the lower Tank Bund road downstream of Hussain Sagar, Hyderabad, India. The total lake area is around 1.875 hectare with an average depth of 8 m with a total water volume of 150,000,000 lL approximately with a daily inflow of approximately 500,000 L.

1.8.2. Lake condition before treatment

The lake is heavily contaminated with blue green algae (BGA) mainly *Microcystis* sp. BGA is present as a suspension in the water column all over the lake (**Figure 2**). Inlet water coming from Hussain Sagar Lake is also contributing to BGA input. BGA mat is removed manually on a daily basis. Foul smells emanate from the lake and also from places were lake water is used for horticulture. Lake water contains total nitrogen (TN) and total phosphate (TP)

concentration of 35.44 and 1.45 mg/l, respectively COD and BOD were also high at 323 and 64 mg/l, respectively.



Figure 2. Lake condition before treatment at different locations.

1.8.3. Treatment using Nualgi

Lake treatment was started on May 20, 2014 (Day 01), with the addition of 4 l of Nualgi lakes with a subsequent addition of 2 l on days 6, 14 and 23, respectively. Nualgi was added along the sides of the pond by using a boat, and water samples for testing were collected before addition. Water condition on the day of addition was heavily contaminated with BGA growth and the BGA layer is formed towards the east-side bridge, and as the air turbulence is from west to east, BGA layer was forming on the bridge side (**Figure 2**).

1.8.4. Change in visible water condition and physiochemical parameters

Water quality of the pond changed considerably when we investigate at visible changes of pond surface (**Figure 3**) and also the change in water quality parameters (**Table 1**) tested before and 1 week after treatment a significant reduction in total dissolved solids (TDS) was observed from the initial reading of 864–474 mg/l; similarly, COD and BOD were also reduced from 350 to 212 and 56 to 14 mg/l, respectively. Nutrient levels also reduced with nitrate reducing from 1.94 to 0.86 mg/l and phosphate reducing from 1.12 to 0.88 mg/l and Total Kjeldahl Nitrogen (TKN) also reduced from 16 to 10 mg/l over a period of 7 days after the addition of Nualgi.

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Figure 3. Visible change in water colour and BGA layer during treatment with Nualgi.

S. No	Parameter	Inlet	Day 01	Day 06	Day 14	Day 23	Percentage of reduction
	Nualgi dosage		4 l	2 l	2 l	2 l	
1	pH	7.61	6.56	6.98	6.99	7.01	
2	Conductivity, ms/cm	1021	932	856	824	839	
3	TDS, mg/l	935	864	474	948	1079	
4	TSS, mg/l	33	14	<10	<10	<10	
5	COD, mg/l	323	350	212	101	32	94%
6	BOD, mg/l	64	56	14	27	10	89%
7	DO, mg/l	0.3	0.6	0.2	1.2	0.8	
8	TKN, mg/l	33	16	10	18	6	83%
9	Nitrate, mg/l	2.42	1.94	0.86	0.78	0.58	82%
10	Phosphate, mg/l	1.45	1.12	0.88	0.54	0.38	80%
11	Faecal coliform	21	18	8	12	12	

Table 1. Water quality parameters tested with percentage of reduction for inlet water from Hussain Sagar, Indira Park Lake water, during treatment with Nualgi and Nualgi dosage pattern.

1.8.5. Change in phytoplankton diversity

From phytoplankton analysis, BGA was the dominant phytoplankton type present in the lake before Nualgi addition and *Microcystis* sp. was the dominant species along with *Spirulina* sp. but after Nualgi addition slowly the phytoplankton dominance shifted from BGA to diatoms (**Table 2**) with *Nitzschia* sp., *Navicula* sp., *Cocconeis* sp., *Gymphonema* sp. and *Gyrosigma* sp. as dominant species along with *Cyclotella* sp. during the initial phase of treatment. From this analysis, Nualgi clearly triggered diatom growth and in turn diatoms acted as catalysts in the improvement of water quality parameters and reduction of BGA growth.

Phytoplankton	Day 01	Day 07	Day 16	Day 24
Blue green algae (BGA)	122	63	43	41
Diatoms				
Pennate	11	34	54	71
Centric	2	13	16	16

Table 2. Changes in phytoplankton concentration and diversity during lake treatment using Nualgi.

2. Conclusions

Using algae for the reduction of nutrients is a unique technology, which utilizes the enormous potential of microalgae in restoring water quality. Diatoms have the ability to simultaneously tackle more than one problem, which is not capable by conventional chemical processes. Growing microalgae using waste water can provide a viable alternative of tertiary biotreatment coupled with simultaneous production of value-added biomass with various benefits such as production of biofuels, antioxidant, anti-cancerous, anti-obesity, anti-viral, antibacterial compounds, aqua, poultry, animal feed additives, etc. Phycoremediation is cost-effective as it saves power and many chemicals, and it has a potential for CO_2 sequestration—a solution for the threat of global warming.

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