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J. I. S. Khattar
D. P. Singh
Rupinder Pal Singh *Editors*

Industrial and Biotechnological Applications of Algae

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Preface

The realm of algae, often overshadowed by more conspicuous aspects of our natural world, has begun to emerge as a major player in the pursuit of sustainable and innovative approaches for food, health, energy, and environment sectors. As we navigate through an era marked by rapid technological and research advancements, the potential industrial and biotechnological applications of algae offer promising avenues for sustainable and environmentally safe solutions in these sectors. In recent years, algae have transcended their traditional roles as aquatic plants to become a central point of discussions about renewable energy, waste management, and biotechnological breakthroughs. Their versatility and high efficiency in photosynthesis, coupled with their ability to prosper in different and even harsh environments, have made them a potential player in the development of eco-friendly technologies and sustainable practices.

This book, *Industrial and Biotechnological Applications of Algae*, aims to provide a comprehensive and updated knowledge on the multifaceted roles that algae play/can play in modern science and industry. The contents of this book delve into their applications, ranging from biofuels and bioplastics to pharmaceuticals, foods, and environmental remediation. Each chapter presents a detailed analysis of current research, technological advancements, and real-world applications, underscoring the transformative potential of algae in addressing our most concerned global challenges.

The contributions in this book are crafted by leading and renowned experts in their field. Their insights reflect the dynamic and evolving nature of algal research and its implications for industrial processes and biotechnological innovations. By bridging the gap between theoretical research and practical applications, this book serves as both a reference and a source of inspiration for students, scientists, engineers, entrepreneurs, policymakers, and others.

As a leading editor and on behalf of our editorial team, we hope for an enthusiastic response from our readers and stakeholders. We are sure they will welcome this panorama of the algal world wholeheartedly.

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Patiala, Punjab, India	J. I. S. Khattar
Patiala, Punjab, India	D. P. Singh
Fatehgarh Sahib, Punjab, India	Rupinder Pal Singh

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Editors and Contributors

About the Editors

Yadvinder Singh is working as an Assistant Professor of Botany in the Department of Botany, Central Univtversity of Punjab, Bathinda, Punjab, India. He completed his Ph.D. in Botany at Punjabi University, Patiala. Dr. Singh is a recipient of numerous national and international awards and fellowships. In his 15 years of research career, he has published extensively on diverse topics, including extremophilic cyanobacteria and algae, the production of value-added products from cyanobacteria, freshwater limnology and ecology, the bioremediation potential of cyanobacteria, cyanobacterial systematics, and phylogenetics. He has published a number of research papers in journals of international repute and several book chapters, having more than 813 citations with 16 h-index and 19 i10-index. Dr. Singh has also been awarded the Early Career Research Award and is a lifetime member of various scientific societies as well as a reviewer for several peer-reviewed journals.

J. I. S. Khattar has recently retired as a Professor of Botany after serving at Punjabi University, Patiala, India, for 32 years. He has published over 70 research papers in reputable national and international journals, primarily in the fields of botany, with a focus on phycology, environmental microbiology, and cyanobacterial biotechnology, garnering more than 1505 citations and an h-index of 22. He has also edited a book on algal biology and biotechnology. His research focused on various aspects of algae, including the taxonomy, physiology, biochemistry, and biotechnology of cyanobacteria. He holds lifetime membership of various scientific bodies, including the Indian Botanical Society, the Indian Science Congress Association, the Association of Microbiologists of India, and the Biotech Research Society of India. He has successfully completed seven major research projects sponsored by various Indian funding agencies. He has supervised and mentored 27 Ph.D. and M.Phil. students. Besides his teaching and research activities, he also served as the Head of the Department of Botany and as the Dean of the Faculty of Life Sciences at Punjabi University, Patiala.

D. P. Singh is an esteemed botanist, researcher, and academician. He is a former Professor of Botany and Head of the Department of Botany at Punjabi University, Patiala, Punjab, India. Dr. Singh has made significant contributions to the understanding of cyanobacterial physiology, biochemistry, and biotechnology. He obtained his Master's in Botany and Ph.D. in Life Sciences from Punjabi University, Patiala. His research career has been marked by a prolific publication record, with more than 45 research papers published in national and internationally peer-reviewed journals, as well as several book chapters. These publications have garnered 1105 citations, according to Google Scholar. Prof. Singh is a lifetime member of various scientific societies, notably the Indian Botanical Society, the Association of Microbiologists of India, and the Biotech Research Society of India, among others. In addition to his research contributions, Prof. Singh has played a crucial role in supervising and guiding students. He has successfully mentored over 10 Ph.D. and M.Phil students, supporting them in their research projects and nurturing their scientific abilities. He has co-edited a book titled *Algal Biology and Biotechnology*.

Rupinder Pal Singh is currently working as an Assistant Professor and In-charge of the Department of Food Processing Technology at Sri Guru Granth Sahib World University, Fatehgarh Sahib, Punjab. He has 10 years of teaching and research experience. His area of research interest includes microbial and food technology, with a specialization in microbial and fermentation technology, which encompasses the production of industrial enzymes from microbial sources, applicable in the food and fermentation industries and also includes the extraction of biopolymers and bioactive compounds from agri-industrial wastes and the valorization of food agri-industrial wastes. He has published his research on the production, purification, and characterization of food-grade prebiotics, microbial production of bio-surfactants, and other food-grade bioactive compounds produced by microorganisms. Dr. Singh has published articles in leading peer-reviewed journals of international repute with good impact factors in the fields of microbiology, biotechnology, and food science. He is a recipient of MASHAV International Fellowship provided by the Ministry of Foreign Affairs of Israel. Currently, he and his team are working on the development of biodegradable food packaging materials from agricultural waste and waste from the fruit and vegetable industry.

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Microalgae-Based Wastewater Treatment and Biomass Production for Different Applications

1

Prabhkirat Kapahi, Esha Goyal, Tufail Fayaz, Sachitra Kumar Ratha, and Nirmal Renuka

Abstract

Phycoremediation is emerging as an innovative approach for the remediation of different types of wastewater rich in nutrients, dyes, pharmaceuticals, as well as other organic and inorganic pollutants. Different microalgal species (*Chlorella*, *Chlamydomonas*, *Scenedesmus*, etc.) have been employed for remediation of various kinds of wastewater, such as domestic, industrial, and municipal wastewater. Phycoremediation provides advantages over conventional treatment methods because it does not negatively impact the environment, and the complete removal of pollutants is also evidenced in many studies. However, several factors, like temperature, pH, light, etc., may impact the efficacy of the removal process, with the best removal found at optimum conditions, which may vary for each strain. Furthermore, the treatment process is accompanied by high-biomass production that can be used commercially in the biofuel, nutraceutical, pharmaceutical, and biofertilizers industries. This chapter, therefore, aims to provide insights into the bioremediation potential of microalgae with various value-added products linked with generated biomass. Microalgae-mediated contaminant removal from different wastewater, the impact of culture conditions on the remediation efficiency and the mechanisms (biodegradation, biosorption, bioaccumulation) governing remediation have been discussed. Potential commercial applications of the generated microalgal biomass are also discussed.

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Keywords

Bioremediation · Commercial applications · Mechanisms · Microalgae · Wastewater treatment

1.1 Introduction

Water is a fundamental and vital requirement for the sustenance of life of all organisms. Thus, water must be safe, readily available, sufficient, and uncontaminated in all respects (Madhav et al. 2020). Water pollution and its remediation have become a significant global concern as most residential and industrial operations generate wastewater containing unwanted and harmful pollutants (Crini and Lichtfouse 2019; Rashid et al. 2021). Worldwide wastewater generation currently stands at about 380 trillion liters annually (Pratap et al. 2023). Unfortunately, just 24% of the wastewater produced is processed properly before it is released into rivers or utilized again for agricultural use (Pratap et al. 2023). Current wastewater treatment technologies utilize a combination of physical, chemical, and biological procedures (Crini and Lichtfouse 2019). Physical processes involve aeration, filtration, sedimentation, and screening, while chemical procedures rely on chemical interactions. Serious drawbacks to these procedures include high-energy input, high cost, changing basic qualities of water, and disruption of native vegetation. In contrast, biological wastewater treatment uses microbes to reduce contaminants (Ahmad et al. 2020; Kannaujiya et al. 2019). This method exhibits greater potential due to their ease of application, lower cost, energy-saving features, and enhanced ecological security (Al-Tohamy et al. 2022). Phycoremediation uses algae to remove harmful contaminants like pesticides, hydrocarbons, radioactive materials, and heavy metals from wastewaters (Koul et al. 2022). It aims to develop a sustainable alternative to traditional activated sludge while reducing production costs of algae cultivation (Li et al. 2019). Many microalgal species, such as *Chlamydomonas reinhardtii*, *Chlorella salina*, *C. vulgaris*, *C. miniata, C. sorokiniana*, *Scenedesmus quadricauda*, *S. abundans*, *Monoraphidium braunii*, *Spirogyra* spp., *Botryococcus*, *Phormidium*, *Navicula* sp., etc. have been studied for this purpose (Ankit et al. 2022; Sakarika et al. 2020). Some studies have shown that employing microalgae has resulted in the removal of approximately 100% of total nitrogen, iron, and ammonium nitrogen (Hassanien et al. 2023). *Chlorella vulgaris,* in three different wastewater types (primary, secondary, and petroleum effluent), can remove 78–83% of total nitrogen and 100% of total phosphorus (Znad et al. 2018). *Chlorella sorokiniana* showed 95% reduction of COD, 62% of N, and 95% of P in municipal wastewater under autotrophic conditions. Under mixotrophic metabolism, the reductions were 89% for COD, 76% for N, and 93% for (Kotoula et al. 2020). The biomass productivity is closely associated with the removal efficiency (Nguyen et al. 2022). Some microalgal strains are resilient to the extreme conditions of industrial and municipal wastewater, making them suitable for treating such wastewater (Abdelfattah et al. 2023). Additionally, the presence of carbohydrates, proteins, fats, carotenoids, and

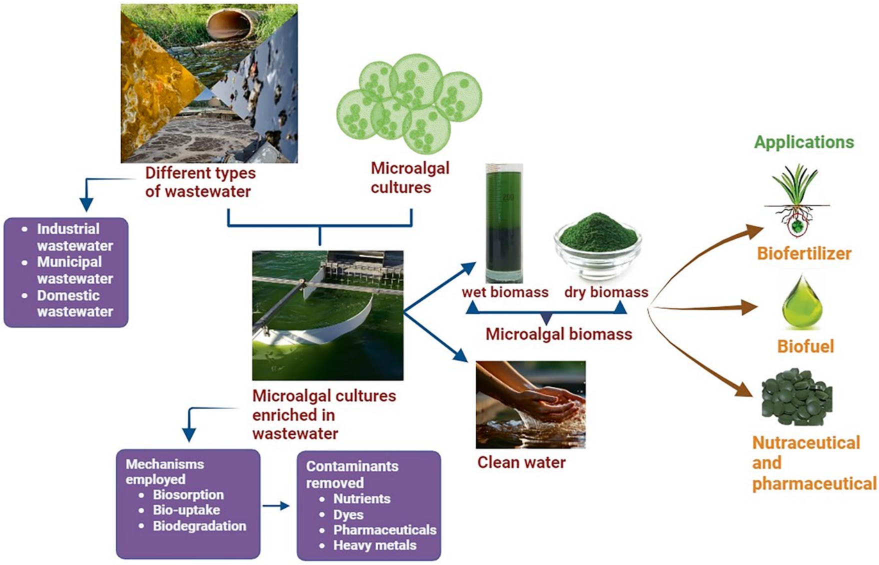

Fig. 1.1 Microalgae enrichment in wastewater for remediation and other commercial applications

other bioactive compounds in their biomass makes them suitable for various industries, including food, animal feed, cosmetics, pharmaceuticals, biofertilizer, and biofuel production (Priyadharshini et al. 2021).

1.2 Applications of Microalgae for Wastewater Treatment

The decline in water quality over the past few decades has been a persistent issue driven by human intervention (Kumar et al. 2023). The three main domains of wastewater are domestic, industrial, and agricultural (Manasa and Mehta 2020). Bioremediation using microalgae and cyanobacteria offers an eco-friendly solution for treating contaminated water and enables wastewater recycling while providing raw materials for energy and material usage (Fig. 1.1).

1.2.1 Domestic Wastewater

Municipal wastewater, a blend of human waste and domestic water, is rich in carbon, nitrogen, and phosphorus, making it an ideal nutrient source for microalgae growth (Do et al. 2022; Ostermeyer et al. 2022). *Scenedemsus obliquus* demonstrated significant nutrient removal efficiency when tested for bioremediation of municipal wastewater, with both phosphorus and nitrogen removal rates above 99% (Ling et al. 2019). Additionally, the microalgal consortium of *Chlorella* sp. and *Scenedesmus* sp. in diluted wastewater showed higher nutrient removal capacity

(78–98%) and higher lipid content (24.5–34.8%) with greater biomass concentration (0.95–1.78 g L^{-1}) (Silambarasan et al. 2021). In primary-treated municipal wastewater, three native microalgae, i.e., *Nostoc muscorum, Navicula veneta*, and *Chlorella vulgaris,* were capable of significantly lowering wastewater's total nitrogen, total phosphorus, and chemical oxygen demand levels (Sisman-Aydin 2022).

1.2.2 Industrial Wastewater

Wastewater from various industries, including textile, automotive, petrochemical, leather, pharmaceutical, agricultural, paper, pulp, oil, brewing, food, and beverage, provides an extensive overview of industrial wastewater and disposal issues (Sathya et al. 2022). Several microalgal species, such as *Spirulina platensis*, *Scenedesmus quadricauda*, *Chlorella tenuis*, *Chlorella pyrenoidosa*, and *Chlorella vulgaris*, have distinct capacities to remove nitrogen, dyes, carbon, heavy metals, and other harmful substances found in wastewater (Sharma et al. 2022). It has been shown that *C. vulgaris* can remove up to 91.9% COD, 95.9% ammonium, 100% nitrate, and 98.8% phosphate from industrial effluents (Yadav et al. 2019). *Chlorococcum humicola* removed 44%, 38%, and 17% of copper, sodium, and cadmium from textile industry wastewater after 6 days of treatment (Borah et al. 2020). These studies indicate that microalgae may be an alternative to conventional treatment methods for remediating industrial wastewater.

1.2.3 Agriculture Wastewater

This wastewater, originating from agricultural activities, contains toxic chemicals from irrigation, herbicides, pesticides, antibiotics, and veterinary medicines (Ali et al. 2021; Plöhn et al. 2021). Reportedly, algae and cyanobacteria can biodegrade pesticides and remediate other pollutants from agricultural runoff. A consortium of cyanobacteria and microalgae, including *S. quadricuda*, *C. vulgaris*, and *S. platensis*, effectively removed heavy metals (nickel, cadmium, and lead) and pesticide malathion from wastewater (Abdel-Razek et al. 2019). *C. vulgaris,* after a two-stage process, showed 83.16%–94.27% of ammonium (NH_4^+-N), 91.24–92.17% COD and 90.98–94.41% of total phosphorus (TP) removal from undiluted cattle farm wastewater (Lv et al. 2018). Three strains of microalgae (*Acutodesmus nygaardii*, *Coelastrella* sp., and *C. sorokiniana*) were used to clean raw piggery wastewater without dilution. Within 6 days, 90% and 92% of the pollutants (ammonia and COD) were successfully eliminated (Lee et al. 2021). Thus, microalgae emerged as an efficient approach for treating agricultural wastewater with simultaneous high biomass productivity.

1.3 Types of Pollutants Remediated by Microalgae

Recently, microalgae have gained attention as the bioremediators of nutrients, heavy metals, pesticides, herbicides, and pharmaceutical products by metabolizing, removing, or absorbing them (Jais et al. 2017; Fayaz et al. 2024; Sutherland and Ralph 2019; Hena et al. 2021). Remediation can be combined with microalgal biomass productivity and value-added product synthesis (Znad et al. 2018). Different contaminants removed by microalgae are discussed below.

1.3.1 Nutrients

Different types of wastewater are rich in nutrients like nitrogen, phosphorus, calcium, magnesium, sodium, sulfate, ammonium, and orthophosphate, which make it unfit for use and may affect water life (El-Sheekh et al. 2016; Fallahi et al. 2021; Mohammadi et al. 2018). Several studies have been conducted to evaluate the potential of microalgae in nutrient removal (Table 1.1). *Oocystis minuta*, *C. vulgaris,* and *S. obliquus* have been reported to efficiently remove phosphate, nitrate, and sulfate (Ajala and Alexander 2020). *Chlorella variabilis* TH03, when cultured in domestic wastewater, removed 74.8–89.8%, 93.8–96.1%, and 97.1–99.9%, of COD, total nitrogen, and phosphorus, respectively (Tran et al. 2021). Likewise, *Scenedesmus* grown in secondary effluent wastewater showed high nutrient recovery with an activated sludge system, leading to high methane production (Arias et al. 2018). Thus, microalgae efficiently remove various nutrients, which can be combined with value-added product synthesis to enhance their utilization and cost-effectiveness.

1.3.2 Heavy Metals

Heavy metal removal using microalgae is an emerging approach that has been researched by various researchers (Singh et al. 2021). Metals, namely cadmium, lead, and copper were remediated by *Spirulina* with a removal efficiency of 92.76%, 94.09%, and 80.75%, respectively, with 5–100 mg L^{-1} concentration of each heavy metal (Sayadi et al. 2019). Biochar of *Scenedesmus quadrifolia* reflected 100% removal of 1–10 mg L^{-1} hexavalent chromium from wastewater (Daneshvar et al. 2019). Another report demonstrated more than 80% Zn^{2+} and 60% Cd^{2+} removal after 3 days of incubation with 20 mg L^{-1} and 4 mg L^{-1} of Zn^{2+} and Cd^{2+}, respectively with *C. vulgaris* JSC-7 (Alam et al. 2015). *Tetraselmis marina* AC16-MESO exhibited 40–90% removal of Cu^{2+}, 100% removal of Fe^{3+}, and 20–50% for Mn^{2+} within 3 days (Cameron et al. 2018). A comparative study on heavy metal removal by microalgal species demonstrated a maximum removal efficiency of 99.4% for manganese by *C. vulgaris*, 91.9% for zinc by *Chlorophyceae* spp., 38.6% for boron by *S. almeriensis,* 40.7% for arsenic by *S. almeriensis,* 88% for copper by *Chlorophyceae* spp. (Saavedra et al. 2018).

Table 1.1 Microalgal enrichment in different wastewater for pollutant removal

Microalgal species	Type of wastewater	Cultivation method/ system	Biomass yield (g L^{-1})	Removal efficiency	Cultivation days/period	Reference
Tetraselmis suecica	Aquaculture wastewater	Mixed bubble column photobioreactors	0.6 ± 0.06	N removal—94.4 ± 1.0% P removal—>90%	7 days	Andreotti et al. (2017)
Dunaliella tertiolecta	Aquaculture wastewater	Mixed bubble column photobioreactors	0.38 ± 0.06	N removal - 95.4 ± 0.3% P removal - >90%	7 days	
Scenedesmus obliquus	Urban wastewater	Vertical tubular photobioreactor	0.4	N removal—95% P removal −92% COD—63%	13 days	Gouveia et al. (2016)
Consortium (*Chlorella, Chaetophora, Scenedesmus, Navicula*)	Urban wastewater	Vertical tubular photobioreactor	0.90	N removal—98% P removal −100% COD—64%	13 days	Gouveia et al. (2016)
Dunaliella salina	Municipal wastewater	Erlenmeyer flasks	0.1695	Nitrate-84.2% Ammonia-71% Phosphorus-47.5%	6 days	Liu and Yildiz (2018)
Chlorococcum humicola	Textile mill effluent	500-mL conical flasks	NA	NH_4–N reduced-81% BOD reduced −46% COD reduced −37% Fe removal- 74.47% Co removal - 44% Na removal −38% Cd removal −17%	6 days	Borah et al. (2020)
Monoraphidium	Aquaculture wastewater	Tubular photobioreactors	3.79	TN removal- 82.62% TP removal- 99.06% Nitrate-85% Nitrite-78% Orthophosphate-100%	10 days	Hawrot-Paw et al. (2020)

1.3.3 Pharmaceuticals

Microalgae-based technology is an emerging technique for remediating wastewater containing pharmaceuticals and drugs (Fayaz et al. 2023; Xiong et al. 2021) (Table 1.2). An alkaline solution-treated biomass of *S. quadrifolia* depicted 91% removal of tramadol within 45 min (Ali et al. 2018). A comparative analysis revealed better pharmaceutical removal by *Tetradesmus* with 40% removal of paracetamol and 93% removal of salicylic acid compared to 21% and 25% removal by *Chlorella* sp. (Escapa et al. 2017). In addition to paracetamol, florfenicol (47 mg L^{-1}) was removed by *Chlorella* L38 with 96% efficiency (Song et al. 2019). Antibiotic triclosan remediation was studied employing three microalgae with a 99.7% removal rate in a day by *S. obliquus* culture, 100% removal efficiency in 7 days by *Desmodesmus* culture, and 69.3% removal by *C. pyrenoidosa* culture (Wang et al. 2018).

1.3.4 Dyes

Wastewater with dyes, if disposed of untreated, leads to their accumulation in the water bodies with adverse impacts on the aquatic life, flora, and fauna (Premaratne et al. 2021). Various studies have demonstrated the successful removal of dyes using microalgae (Ihsanullah et al. 2020; Majumdar et al. 2022) (Table 1.2). A study on *S. obliquus* demonstrated 98.14% removal of orange 2RL Azo dye (20 ppm) when supplemented with glucose at pH 11 (Hamouda et al. 2022). This species is also effective in bioremediating methyl red and congo red dyes with removal efficiencies

Table 1.2 Microalgal-based pollutant removal studies

<table>
<tr><th>Microalgae</th><th>Pollutant</th><th>Type of pollutant</th><th>Removal mechanism</th><th>Removal (%)</th><th>Reference</th></tr>
<tr><td>Chlorella vulgaris</td><td>Methylene blue</td><td>Synthetic dye</td><td>Surface adsorption</td><td>83.04%</td><td>Chin et al. (2020)</td></tr>
<tr><td>Scenedesmus sp.</td><td>Acid blue 161</td><td>Dye</td><td>Surface adsorption</td><td>76.65%</td><td>da Fontoura et al. (2017)</td></tr>
<tr><td>C. pyrenoidosa</td><td>Triclosan</td><td>Emerging contaminant</td><td>Bioaccumulation</td><td>69.30%</td><td>Wang et al. (2018)</td></tr>
<tr><td>Nostoc muscorum</td><td>Malathion</td><td>Pesticide</td><td>Biodegradation</td><td>91%</td><td>Ibrahim et al. (2014)</td></tr>
<tr><td>Pseudanabaena sp.</td><td>Cypermethrin</td><td>Pesticide</td><td>Biodegradation</td><td>98%</td><td>Bano et al. (2021)</td></tr>
<tr><td rowspan="2">Scenedesmus dimorphus</td><td>17α-estradiol</td><td rowspan="2">Emerging contaminants</td><td rowspan="2">Biodegradation</td><td>85%</td><td rowspan="2">Bano et al. (2021)</td></tr>
<tr><td>17β-estradiol</td><td>95%</td></tr>
<tr><td rowspan="3">C. sorokiniana</td><td>Ibuprofen</td><td rowspan="3">Emerging contaminants</td><td rowspan="3">Biodegradation, photolysis</td><td>99%</td><td rowspan="3">de Wilt et al. (2016)</td></tr>
<tr><td>Paracetamol</td><td>99%</td></tr>
<tr><td>Metoprolol</td><td>38%</td></tr>
</table>

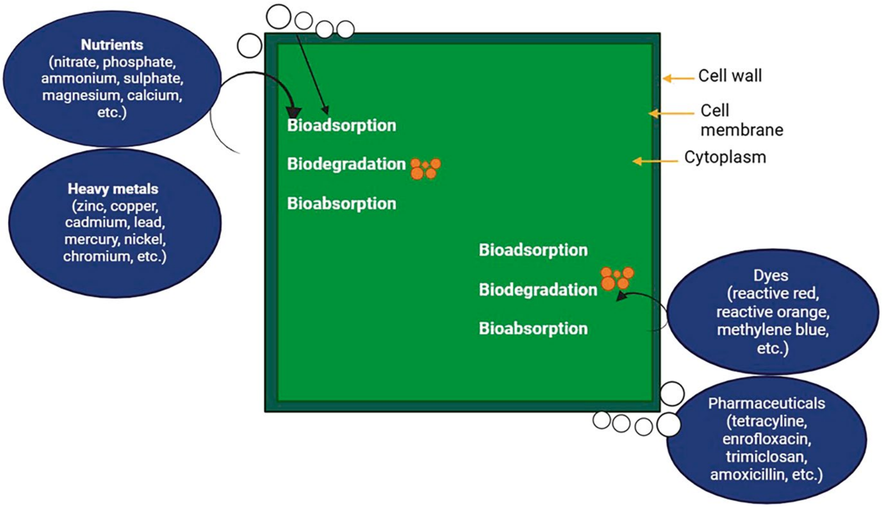

Fig. 1.2 Mechanisms governing contaminant uptake from wastewater by microalgae

of up to 48.60% and 41.15% in 10 days (Abou-El-Souod et al. 2020). *Arthrospira platensis* showed 75.7% and 61.11% removal of ismate violet 2R dye with dry and lipid-free biomass, respectively, at 10 mg L^{-1} of initial dye concentration (Alprol et al. 2021). After 7 days of treatment, *Oscillatoria* and *Scenedesmus* mixture removed azo dyes reactive orange and reactive red with 98.5% and 97.5% removal efficiency, respectively (El-Sheekh et al. 2021). Thus, microalgae are an attractive option for remediating dyes from wastewater and utilizing the harvested biomass for other commercial applications.

1.4 Mechanisms of Microalgal Bioremediation

Microalgae can eliminate various pollutants through biosorption, bioaccumulation, or biodegradation (Fig. 1.2). These mechanisms are discussed in the following sections.

1.4.1 Biosorption

Biosorption is the ability of active/inactive, living/non-living materials of biological origin to bind and collect metal ions from aqueous solutions (Sathya et al. 2022). Microalgae biosorption is a passive, metabolically independent activity involving complexation, precipitation, adsorption, absorption, and ion exchange (Yaashikaa et al. 2021). Algal cell walls consist of lipids and hetero-polysaccharides with negatively charged groups such as phosphate, carboxyl, hydroxyl, and amino, interactively binding with metal ions (Ramesh et al. 2023). Other heavy metals like lead and nickel were sequestered by microalgae *Rhizoclonium hookeri,* showing a maximum adsorption capacity of 81.7 mg g^{-1} of lead and 65.81 mg g^{-1} of nickel (Suganya et al. 2017). The ability of *Scenedesmus* sp. and *Synechocystis* sp. to adsorb diclofenac was examined. Diclofenac may be recovered from biomass by *Scenedesmus* sp. at a rate of 28 mg g^{-1} and by *Synechocystis* sp. at a rate of 20 mg g^{-1} (Coimbra et al. 2018).

1.4.2 Bioaccumulation

Bioaccumulation involves the gradual intracellular active diffusion across the cell membrane during the cellular metabolic activity and accumulation within the cells (Srimongkol et al. 2022). Regardless of the process, bioaccumulation is significantly impacted by internal and external physicochemical factors such as pH, temperature, contact time, and pollutant concentration (Abdelfattah et al. 2023). *Chlorococcum* sp. accumulated 239.09 μg g^{-1} dw Arsenic [As(III)] in 10 days when grown in 10 μM arsenic (Upadhyay et al. 2022). The combined effect of biosorption and bioaccumulation is highly significant for the elimination of metal ions from wastewater (Rempel et al. 2021). Another study assessed the toxicity, bioaccumulation, and biodegradation of atrazine in the green microalga *C. mexicana*, finding it can accumulate and biodegrade atrazine within cells, ranging from 14% to 36% at 10–100 μg L^{-1} (Kabra et al. 2014).

1.4.3 Biodegradation

Biodegradation is the process by which pollutants are broken down into simpler molecules by enzymatic activities (Rempel et al. 2021). The biodegradation of diclofenac was investigated using two microalgal strains (*Picocystis* sp. and *Graesiella* sp.), which reached 69% and 44%, respectively, at 25 mg L^{-1} of initial diclofenac concentration (Ouada et al. 2019). After 30 days of incubation, both *Oscillatoria* sp. and *Chlorella* sp. could degrade 95% and 78.71% of pyrene (50 mg L^{-1}), respectively (Aldaby and Mawad 2019). Similarly, nonylphenol was removed and biodegraded by four different freshwater microalgae. At 120 h, *Chlorella vulgaris* had the most significant nonylphenol biodegradation percentage of 68.80%, followed by 65.63% (*Ankistrodesmus acicularis*), 63.10% (*Scenedesmus quadriauda*), and 34.91% (*Chroococcus minutus*) (He et al. 2016). Microalga, *Chlamydomonas mexicana* demonstrated the ability to accumulate and biodegrade atrazine within the cell, with atrazine degradation ranging from 14% to 36% at 10–100 μg L^{-1} (Kabra et al. 2014). Therefore, these studies indicate that different types of microalgae play a role in the biodegradation of contaminants, highlighting their capacity to convert a variety of dangerous compounds into less harmful forms.

1.5 Factors Influencing Microalgal Bioremediation

Microalgal bioremediation is influenced by factors like temperature, pH, nutrient availability, and light intensity, which can all impact the growth of microalgae, which varies from species to species (Khan et al. 2018).

1.5.1 Light

Light serves as both the energy source for algal photosynthesis and a prerequisite for the development of microalgae (Wang et al. 2022). Different microalgal strains (*Pseudokirchneriella subcapitata*, *C. vulgaris*, *Microcystis aeruginosa*, and *Synechocystis salina*) were utilized to investigate the effects of light irradiance and light: dark period on microalgal growth and nutrient removal (Gonçalves et al. 2014). Complete nitrogen removal was observed in cultures irradiated with a light intensity of 180 μE m^{-2} s^{-1} (Gonçalves et al. 2014). Microalgal strains (*Desmodesmus* sp., *C. vulgaris*, *S. obliquus*, and *Ettilia pseudoalveolaris)* were cultured under different light intensities. Except for *Desmodesmus* sp., over 75% of total nitrogen and phosphorus content was removed from treated wastewater at 50 μE m^{-2} s^{-1} light intensity (Nzayisenga et al. 2020). The study examined the impact of different light qualities (red, blue, white, and mixed red and blue) on the nutrient removal, growth, and biochemical composition of *C. sorokiniana* in aquaculture effluent. Results showed that blue light degraded the most organic carbon (82.27%), while combined red and blue light removed the most total nitrogen and NH_3-N (93.25%) (He et al. 2023). These studies indicate the role light quality and intensity play in nutrient removal.

1.5.2 Temperature

Temperature plays a crucial role in regulating microalgae's metabolic processes and growth (Arora et al. 2021). An evaluation of different microalgal strains grown in secondary effluent from a treatment facility at 10 and 22 °C temperature revealed that both temperatures could help achieve 70% removal of the initial N and P (Abdelaziz et al. 2014). Another study evaluated nutrient removal efficiency in *P. subcapitata*, *C. vulgaris*, *M. aeruginosa*, and *S. salina* at 25 °C using light irradiance. The maximum nitrogen removal efficiency was 100% for all three species. Phosphorus removal efficiency varied, with *M. aeruginosa* having the lowest efficiency of 1.13% at 15 °C and *C. vulgaris* having the highest efficiency of 67.6% at 25 °C at the light intensity of 180 μE m^{-2} s^{-1} (Gonçalves et al. 2016). The study used three temperature regimes low (4 °C), high (35 °C), and alternating high-low (35 °C during the day and 4 °C at night) temperatures for 15 days to cultivate *C. vulgaris* in municipal wastewater. The highest biomass concentration (1.62 g L^{-1}) and most effective elimination of COD (83.0%), TN (96.5%), NH_3-N (97.8%), and TP

(99.2%) were observed when the alternating high-low temperature condition was used (Xu et al. 2019).

1.5.3 Nutrient Availability

The effect of wastewater discharge on the environment is lessened when water is pre-treated with microalgae to remove nutrients. *S. obliquus* was assessed for its growth and nutrient removal efficiency in a solution containing diammonium phosphate, urea, and glucose as P, N, and C sources (Shashirekha et al. 2016). The results showed a nearly 80% reduction in COD and BOD levels, and a maximum biomass yield of 1.23 g L^{-1} was achieved in 5 days (Shashirekha et al. 2016). Complete removal of nitrates, carbon and phosphorus by *S. obliquus* was observed at C:N:P ratio of 0.2:0.14:0.08 in sugar mill effluent. The study assessed the nutrient removal efficiency of *Chlorella* and *Scenedesmus* under varying initial N and P concentrations (Beuckels et al. 2015). The concentration of nitrogen in the wastewater impacted the nutrient removal capacity of microalgae, where *Chlorella* and *Scenedesmus* showed a phosphorus removal of 2 mg L^{-1} under 20 mg L^{-1} nitrogen. In contrast, the phosphorus removal capacity increased to 6 mg L^{-1} at more than 40 mg L^{-1} nitrogen concentrations.

1.5.4 pH

pH is a crucial factor in microalgal cultures, influencing metabolism and regulating the availability and solubility of nutrients and CO_2. The ideal pH range for the generation of lipids and biomass in microalgal species is reported to be strain-specific (Qiu et al. 2017). The nutrient absorption by *Chlorella* sp. at pH values ranging from 5.0 to 11.0 showed that neutral or slightly alkaline conditions produced higher absorption efficiencies for total nitrogen and phosphorus, while acidic conditions led to lower absorption (Zhang et al. 2014). At an initial pH of 7.0, 87.77% of total nitrogen and 92.05% of phosphorus were removed by *Chlorella* sp. from 50% BG-11 medium (Zhang et al. 2014). Sutherland et al. (2015) concluded that co-varying CO_2 and pH impacted the removal of dissolved inorganic nitrogen. At the lowest pH, the proportion of nitrogen removed increases at the greatest dissolved inorganic carbon concentration (Sutherland et al. 2015). Even at pH levels below 3, the mixed culture predominated by *Parachlorella* efficiently absorbed NH_4^+-N and grew well in anaerobic digestion effluent (Yu et al. 2022).

1.6 Commercial Applications

Green solutions, like phycoremediation, are cost-effective, sustainable, and energy-efficient substitutes that yield critical secondary metabolites, biofuel, and nutrient recovery from contaminated freshwater sources (Kumar et al. 2023). Microalgae recovered after wastewater processing, due to their high content of proteins, carbohydrates, and lipids, can be exploited to create value-added products (Udaiyappan et al. 2017). The industrial applications of the generated biomass are discussed below.

1.6.1 Biofuel Production

Microalgae are considered the most realistic green biodiesel production alternative due to their ability to grow on wastewater and their adaptability to changing atmospheric conditions (Bhalamurugan et al. 2018). Many studies have reported the possibility of biofuel production along with microalgal remediation (Table 1.3). Treating sewage effluent using *C. vulgaris* reduced 93% nitrates, 95% COD, and 92% BOD and simultaneously generated 0.67 g L^{-1} biomass and 0.26 g $L–^{1}$ lipid yields (Pooja et al. 2022). Optimizing *Nostoc ellipsosporum* growth conditions, including 20% Fog's medium and 80% wastewater, increased biomass yield from 1.42 to 2.9 g L^{-1}, removed 87.59% nitrogen and 88.31% phosphate, and produced 24.62% bio-oil (Devi and Parthiban 2020). Biogas was generated from residual microalga consortium (*Nostoc*, *Phormidium*, and *Geitlerinema*) biomass in

Table 1.3 Microalgae-based pollutant removal from different wastewater with potential applications of the generated biomass

Microalgae	Wastewater treatment	Application	Biomass production	Reference
Neochloris sp.	Polluted river water pharmaceutical effluent	Feed and healthy food applications	520 mg L^{-1}	Singh et al. (2020)
Chlorella sp.			498 mg L^{-1}	
Chlorococcum sp.			450 mg L^{-1}	
Chlorococcum humicola	Textile mill effluent	Bioethanol and biobutanol	$0.87*10^7$ cells mL^{-1} d^{-1}	Borah et al. (2020)
Chlorella sp.	Black gunpowder production effluents	Biofuel production, aquaculture, and the food industry.	380.60 mg L^{-1}	Condori et al. (2024)
Scenedesmus sp.			454.57 mg L^{-1}	
Neochloris sp.	Aquaculture wastewater	Fish feed ingredient	0.317 ± 0.0473 g L^{-1}	Kashem et al. (2023)
Calothrix sp.	Sewage wastewater	Biofertilizers and pigments	916.67 ± 5.77 mg L^{-1}	Renuka et al. (2013)
Chlamydomonas debaryana	Dairy wastewater	Biodiesel production	3.66 ± 0.21 g L^{-1}	Arora et al. (2016)
Chlorella fusca	Urban wastewater	Biofuel production	65.5× 10^6 cell mL^{-1}	Arrojo et al. (2022)

secondary effluent after phycobiliprotein extraction, yielding methane from 159 to 199 mL CH_4 g^{-1} volatile solid (Arashiro et al. 2020). These studies indicate that microalgae cultivation in wastewater has the potential for commercial application in biofuel production.

1.6.2 Lipids and Pigment Production

The algal biomass produced during wastewater treatment can be used as a source of pigments as well as animal feed supplements (Marella et al. 2020). The study on *Phaeodactylum tricornutum* showed that treating it with swine wastewater: water ratio of 1:1 maximized algal biomass at 1.54 g L^{-1} and fucoxanthin accumulation to 28.41 mg L^{-1}, highlighting its efficiency in nutrient removal and fucoxanthin production from swine effluent (Jiang et al. 2022). *Nostoc*, *Phormidium*, and *Geitlerinema* cultured in wastewater were effectively used to extract the natural pigments phycocyanin and phycoerythrin up to 20.1 mg^{-1} g^{-1} and 8.1 mg^{-1} g^{-1} dry weight, respectively (Arashiro et al. 2020). *C. vulgaris* showed carotenoid concentrations of 6.34 mg L^{-1} and 201.69 mg L^{-1} after 5 and 10 days of incubation in 10% and 5% tofu wastewater medium, while *A. platensis* had 1.16 mg L^{-1} and 72.20 mg L^{-1} carotenoid content (Ajijah et al. 2020). Mixed algal cultures of *Euglena gracilis* and *Selenastrum*, grown in sludge-amended pike perch aquaculture wastewater, produced the maximum quantities of algal biomass (1.5 g L^{-1}), tocopherol (877.2 μg L^{-1}), and lipid (84.9 mg L^{-1}) (Tossavainen et al. 2019). Research shows microalgae species can effectively treat wastewater by removing nutrients and producing beneficial compounds like lipids, carotenoids, fucoxanthin, phycocyanin, and phycoerythrin.

1.6.3 Carbon Sequestration

Microalgae can be a significant carbon sink while utilizing nutrients from wastewater (Viswanaathan et al. 2022). They can sequester 1.3 kg of carbon dioxide per kilogram of biomass, and in open pond cultures, they can absorb and convert 513 tons of CO_2 into 280 tons of dry biomass per hectare per year (Sarwer et al. 2022). *Chlorella* sp. achieved maximum biomass growth (1.52 g L^{-1}) and CO_2 fixation rate (187.65 mg L^{-1} d^{-1}) by the fifth day, with over 70% nutrient removal, indicating successful wastewater treatment and CO_2 capture (Yadav et al. 2019). *Chlorella* sp. cultivated in tannery effluent showed the maximal concentration of protein, carbohydrate, and lipid to be 160 μg mL^{-1}, 250 μg mL^{-1}, and 0.95 g L^{-1}, respectively; with 60.50% CO_2 removal (Rajalakshmi et al. 2021). These studies indicate that microalgae may be an effective alternative for the sequestration of carbon dioxide and wastewater treatment.

1.6.4 Biofertiliser Production

Wastewater-grown microalgal biomass from two consortia (*Anabaena, Phormidium, Westiellopsis, Spirogyra, Fischerella*) and unicellular (*Scenedesmus, Chlorella, Chroococcus, and Chlorococcum*) significantly improved wheat crop productivity and growth compared to the prescribed dosage of NPK fertilizers (Renuka et al. 2016). Five different microalgae strains (*Chlorella* sp., *Monoraphidium* sp., *Neochloris* sp., *Dictyosphaerium* sp., and *Scenedesmus* sp.) grown in municipal wastewater were found to be more effective in providing nitrogen content to wheat plants than NPK fertilizer (Das et al. 2019). The consortium of *Chlorella* sp. and *Scenedesmus* sp. grown in 75% wastewater with inorganic fertilizer improved shoot and root length (44% and 89%, respectively) of *Solanum lycopersicum* with 174% enhancement yield (Silambarasan et al. 2021). These studies suggest that microalgae obtained after wastewater treatment have significant potential as sustainable biofertilizers.

1.7 Conclusions

Microalgal bioremediation has recently attracted attention worldwide due to less negative environmental impacts. However, there is a need to reduce the cultivation cost and enhance the reliability of the biological approach. Optimizing various ecological factors and selecting wastewater-adapted microalgal strains would help overcome the underlying limitations. Furthermore, genetically modifying the microalgal strains can increase nutrient removal efficiency, make them resistant to toxic contaminants and enhance growth. Technology development for treating large volumes of wastewater would uplift the economic barrier to make this technology sustainable.

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