Asia-Pacific Journal of Science and Technology: Volume: 29. Issue: 02. Article ID.: APST-29-02-01. Review Article

APST

Asia-Pacific Journal of Science and Technology

https://www.tci-thaijo.org/index.php/APST/index

Published by the Research and Graduate Studies,
Khon Kaen University, Thailand

The comprehensive review of essential role of microalgae in organic pollutants mechanisms of phycoremediation

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Received 4 January 2023
Revised 8 June 2023
Accepted 3 October 2023

Abstract

Waste management is a significant problem affecting both developed and developing countries. In addition, the presence of organic pollutants in waste poses a substantial threat to environmental quality and public health. To address this issue, bioremediation using microalgae (phycoremediation) such as *Chlorella vulgaris*, *Botryococcus braunii*, *Desmodesmus* sp., and *Chlamydomonas* sp., is considered an environmentally friendly and uncomplicated solution. The biomass produced by microalgae can also be used as a source of biodiesel, lipid, biofertilizer, and biohydrogen. Therefore, this research aimed to explore the underlying mechanisms of phycoremediation in addressing organic pollutants, which include biosorption, consumption, and biodegradation. The results showed that phycoremediation was more effective than non-biological methods, but it has not been fully optimized. This showed that further research should focus on the optimization of phycoremediation and its integration with biorefinery to maintain environmental quality from organic pollutants and produce biomass as feedstock in biorefinery activities.

Keywords: Biorefinery, Organic pollutants, Phycoremediation, Pollution, Waste

1. Introduction

In North America and Europe, 67 billion m^3 of wastewater are generated annually, with an average of 231 m^3/capita^{-1} in the US. The accumulation of this waste has led to pollution of various water sources in the region [1]. Generally, waste is divided into organic and inorganic, based on the constituents of its compounds. Organic waste primarily consists of compounds such as dichlorodiphenyl trichloroethane (DDT), polychlorinated biphenyls (PCBs) and polybrominated biphenyls (PBBs), polycyclic aromatic hydrocarbons (PAHs), Organophosphorus insecticides (OPs), dieldrin, tributyltin (TBT), and methyl tert-butyl ether (MTBE).

Organic compounds with strong hydrophilic properties can be degraded by bacteria, fungi, and microalgae, while those that are persistent degrade slower [2]. Several processes are applied in treating organic pollutants, including chemical oxidation technology, adsorption, solvent extraction, and incineration. The incineration method is used to destroy organic components into carbon dioxide and water [2]. However, this method has adverse effects on the environment and health due to the phase change to ash, which can be released into the air. To address this issue, phycoremediation using microalgae has been found as a potentially, safe, environmentally, and relatively easy strategy [3-5].

In phycoremediation process, microalgae biomass produced can be used for biodiesel production [5]. The absorption of organic pollutants by microalgae from the environment is facilitated by specific polymers within their cells, through three general mechanisms, namely biodegradation, consumption, and biosorption. Moreover, the nutrients found in organic pollutants can be used by microalgae, including *Chlorella* and *Dunaliella*, as their energy sources to enhance growth-producing biomass, known as biorefinery concept. Due to their relatively high carbohydrate and lipid contents, biorefinery concept focuses on the conversion of raw materials into industrial intermediate or final products such as biodiesel, feed, biohydrogen, biogas, bioethanol, and electric power.

Several advantages of using microalgae in phycoremediation include their rapid growth rates, high adaptability to various pollutants, capability to absorb nutrients, the potential for integration with other microorganisms, CO_2 absorption from the air, and biomass for biorefinery activities [6]. The effectiveness of phycoremediation is influenced by the strain, showing the importance of selecting and using superior strains with a great ability to absorb organic pollutants and produce high biomass. Additionally, optimizing the process can also incorporate synergistic bacteria and fungi [7].

2. Phycoremediation of organic pollutants by microalgae

Table 1 shows several microalgae species capable of being phycoremediation agents of synthetic dyes, while some isolates reported to remediate other organic pollutants are presented in Table 2.

Table 1 Some potential microalgae in synthetic dyes phycoremediation.

Microalgae isolates	Organic pollutants	Initial concentrations	Efficiency removal (%)	Time	References
Arthrospira (*Spirulina*) *platensis*	Naphthol green-B	0.5-3 g/L	30.39-99.92	60^{*}	[8]
	Indigo blue dye	100 mg/L	89.90	3^{***}	[9]
Chlamydomonas variabilis	Methylene blue	82.4 mg/L	98	30^{*}	[10]
Chlorella vulgaris	Methylene blue	100 mg/L	83	3^{***}	[11]
	Aniline blue	100 mg/L	46.10	11^{***}	[12]
	Azo green	20 mg/L	21.98	14^{***}	[13]
	Optilan red	50 mg/L	>50	96^{***}	[14]
Sphaerocystis schroeteri	Azo dyes blue	20 mg/L	10.98	14^{***}	[13]

Table 2 Some potential microalgae in other organic pollutants phycoremediation.

Microalgae isolates	Organic pollutants	Initial concentrations	Efficiency removal (%)	Time	References
Chlamydomonas mexicana	Atrazine	100 µg/L	64	14^{***}	[15]
	Diazinon	20 mg/L	64	12^{***}	[16]
Chlorella reinhardtii	17 β-estradiol	5 mg/L	86	7^{***}	[17]
	17α-ethinylestradiol	5 mg/L	71	7^{***}	[17]
	Tris(2-butoxyethyl) phosphate	-	15	7^{***}	[17]
	Bisphenol A	-	22	7^{***}	[17]
Chlorella sorokiniana	Salicylic acid	25 mg/L	> 67	4^{***}	[18]
	Paracetamol	25 mg/L	73	4^{***}	[18]
Chlorella vulgaris	Diazinon	20 mg/L	94	12^{***}	[16]
	17β-estradiol	5 mg/L	100	40^{***}	[19]
Haematococcus pluvialis	Estrone	5 mg/L	97	40^{***}	[19]
	17α-ethynylestradiol	5 mg/L	85	40^{***}	[19]
	17β-estradiol	5 mg/L	100	40^{***}	[19]
Nannochloropsis sp.	Paracetamol waste	50.5-44.4 µg/mL	12.07	24^{*}	[20]
	Ibuprofen waste	50.4 - 44.3 µg/mL	12.70	24^{**}	[20]
	Olanzapin	49.0 - 33.1 µg/mL	32.40	12^{**}	[20]
Platymonas subcodicormis	Nonylphenol	0.5-2.5 mg/L	82.38	5^{***}	[21]
Selenastrum capricornutum	17β-estradiol	5 mg/L	100	40^{***}	[19]
	Estrone	5 mg/L	80	40^{***}	[19]

Note: * min, ** h, and *** day

2.1 Synthetic dyes

2.1.1 Methylene blue

Methylene blue (MB), with the chemical formula $C_{16}H_{18}ClN_3S$, is an irritative substance commonly used in dyeing fabric, the printing of tannin, and for medicine or antiseptic purposes [10]. Although methylene blue is widely used as a dye in the clothing, textile, and paper industries, its presence in aquatic ecosystem can be harmful. Chin et al., 2020 reported that *Chlorella vulgaris* absorbed organic dyes, methylene blue, achieving an absorption rate of 83% within three days with an initial concentration of 100 mg/L^{-1}. These absorption mechanisms occurred through the interaction between oppositely charged objects (the positively charged methylene blue and the negatively charged microalgae cell surface). The negative charge is attributed to functional groups on the cell surface, such as -OH and -COOH [11].

Chlamydomonas variabilis also serves as a natural bio-adsorbent for methylene blue dye, achieving an absorption percentage of 98% against 82.4 mg/L concentration of methylene blue. This absorption process is facilitated by the presence of nanopores and nanoparticles on the cell surface. Furthermore, High-Resolution Transmission Electron Microscopy analysis showed a protective sheath on the cell surface of *C. variabilis,* enhancing the tolerance of the cells when exposed to the toxic effect of the dye [10].

2.1.2 Naphthol green-B

Naphthol green-B (NGB) is a derivative of naphthoic acid and a water-soluble salt, with similar applications to methylene blue. The main advantage of using Naphthol green-B is its cost-effectiveness and efficient electron transfer [8]. Moreover, *spirulina* (*Arthropsira*) *platensis* can be used as a bio-adsorbent for synthetic dyes. This microalgae absorbed the dyes by binding to the cell surface, achieving an absorption rate, ranging from 30.39 to 99.92% with a dye concentration of 0.5-3 g/L within 60 min. The absorption occurred due to the difference in charge between the cell surface and the dye. At low pH (pH = 3), the amino-functional group (NH_3^+) on the cell surface becomes protonated, facilitating the interaction between dyes containing negatively charged groups (SO_3^-) through elec mg trostatic interactions [8].

2.1.3 Aniline blue

Aniline blue ($C_6H_5NH_2$) is an organic compound, characterized by different solubility levels in water compared to Naphthol-green B. These characteristics include lipid consistency, moderate level solubility in water, and volatility with fatty odor [22]. In the case of *Chlorella vulgaris* growth in the medium containing aniline blue, the exponential phase occurred from the 3rd day and lasted until the 9th day. The absorption percentages of microalgae growth medium, with the addition of 5, 50, 75, and 100 mg/L of aniline blue, were 58, 53, 53, and 46.1% on the 11th day [12].

2.1.4 Azo dyes

Azo dyes are the most widely used type in the present and future textile, printing, and paper manufacturing industries. These dyes have various colors that depend on their bonds and characterized by the functional group -N−N, connecting both symmetrical and asymmetrical identical (McLaren in,[23]). Microalgae such as *C. vulgaris* and *Sphaerocystis schroeteri,* have the capacity to absorb azo dyes in blue and green hues, with the highest absorption and decolorization rates being observed on the 14th day of incubation. The maximum decolorization percentage by *C. vulgaris* against azo green was found to be 21.98% with a dye concentration of 20 mg/L. Meanwhile, *Sphaerocystis schroeteri* absorbed 10.98% and 17.58% of azo dyes blue and green with an initial concentration of 20 mg/L. The growth rate of *C. vulgaris* in the medium containing the dye was higher compared to *Sphaerocystis schroeteri* due to greater cell division. This increased cell division is a metabolic process taking place within the cell, with the dye serving as a nutrient source for microalgae cells [13].

2.1.5 Disperse red 3B

Disperse red 3B is an anthraquinone dye characterized by bright color and light, excellent sun fastness, and slightly poor sublimation readiness, making it applicable for the printing of polyester and nylon fabrics [24]. When considering its remediation, the consortium of *Chlorella sorokiniana* and the fungi *Aspergillus* sp. showed the ability to absorb and disperse red 3B. During the 4th day of incubation, the consortium achieved an impressive 98.09% decolorization of the dye. The process of dye decolorization depends on two essential enzymes, including lignin and manganese peroxidase, both intracellular and extracellular. The activity of extracellular enzymes was higher compared to intracellular in the degradation. Moreover, the degradation of disperse red 3B is the breaking down of the anthraquinone dye molecule ($C_{20}H_{12}NO_4$) into simpler compounds with lower toxicity. These enzymes oxidize the ring-opening reaction of the dye molecules, resulting in the formation of products such as diisobutyl phthalate, guaiacol, and ammonia. Subsequently, diisobutyl phthalate passes through an oxidative decarboxylation reaction to produce acetone, o-xylene, and 4-hydroxy-2-butane-one. Each of these products is further oxidized to produce CO_2, followed by the oxidation of guaiacol to obtain CO_2 [25].

2.1.6 Indigo blue dye

Indigo dye ($C_6H_{10}N_2O_2$) is a blue organic pollutant extracted from the leaves of the Indigofera genus or processed by fermentation. Although indigo blue dye is a suitable color for natural fibers, water contaminated with it may contain acid and alkaline agents [26]. This dye can effectively be absorbed by *Spirulina platensis* within 3 days of incubation, achieving an adsorption rate of 89.9% with an initial concentration of 100 mg/L^{-1}. The optimal conditions for this absorption were found at pH 4 and 50°C, due to high temperature, which increased the energy [9]. The rate of dye decolorization in microalgae is affected by several factors such as cell concentration, pH, exposure time, and the properties of wastewater. Immobilized *Chlorella vulgaris* using sodium alginate also can be used as a bio-adsorbent for indigo blue dye. The most effective decolorization rate of 49.03% was observed at pH 5, with a contact time of 24 h, at an algae concentration of 50,000 cells mL^{-1} and 100 number bead densities. However, the repeated use of immobilized cells resulted in a lower rate of decolorization. Several functional groups on the cell surface such as amino, alkyl, alkenyl, carbonyl, and phenyl groups, play a significant role in the adsorption of the dye. Amino groups in the indigo blue dye and microalgae cell surfaces contribute to reductive amination reactions, alkylation, or conversion to amides. The reaction between these functional groups initiates the absorption and degradation of dye compounds, which leads to decolorization [27].

2.1.7 Optilan red

Chlorella vulgaris showed the ability to absorb red dye, with an EC50 value of 23.16 mg/L. Microalgae absorbed more than 50% of the dye with an initial concentration of 50 mg/L within 96 h. However, the red dye is toxic to microalgae cells, impacting proteins, pigments, and other essential compounds, thereby reducing the growth of the strain. This phenomenon shows the importance of implementing effective phycoremediation practices [14].

2.2 Textile waste

The textile industries conduct two main activities, weaving and fabric dyeing. Most of these industries use synthetic dyes that have stable and various color options, and their waste contains phosphate, nitrate, and other micronutrients. Consequently, certain microalgae can effectively use textile wastewater to support their growth, offering opportunities for integration with environmental bioremediation processes and the production of microalgae biomass as a lipid source in biodiesel production [28]. *N. muscorum* and *A. variabilis* have been used to reduce biochemical oxygen demand (BOD) and chemical oxygen demand (COD) levels in textile industrial effluents. After 15 days of treatment, BOD decreased from 110 to 50 (*N. muscorum*) and 48 mg/L for *A. variabilis,* while COD reduced from 560 to 380 and 360 mg/L. The COD reduction was 54.55% and 56.36% for *N. muscorum* and *A. variabilis*, while the COD efficiency was 32.14% and 35.71% (*N. muscorum* and *A. variabilis*) [29].

Das et al. [30] explored the potential of using microalgae consortium of *Chlorella* sp. and *Phormidium* sp. for phycoremediation to reduce tanning wastewater (TW) pollution. Microalgae were grown in 100% TW waste for 20 days, resulting in a reduction of various parameters, including BOD = 93.40%, COD = 92.60%, total nitrogen (TN) = 91.16%, and total phosphorus (TP) = 88%. Other types of *Chlorella,* such as *Chlorella pyrenoidosa* have showed their effectiveness in managing pollution in textile wastewater, containing BOD (0.01-1.8 g O_2 L^{-1}), COD (1.1-4.6 g O_2 L^{-1}), total dissolved solids (TDS) (50 mg/L) with a pH range of 5-9. Pathak et al. [31] reported that textile wastewater contained organic, inorganic, and dye components needed for microalgae growth. *Chlorella pyrenoidosa* reduced BOD, TN, and TP by 81%, 62%, and 87%, respectively. Furthermore, green microalgae *Golenkinia radiata* grew in sterilized and non-sterilized textile wastewater. This microalga was tolerant toward the physical properties of textile waste, showing its potential as a candidate in phycoremediation process [32].

2.3 Nonylphenol (NP) waste

NP is a nonionic surfactant often packed in the form of detergent, emulsifier, and dispersing agents. Due to its low solubility and high hydrophobicity, NP tends to form sediments, resulting in an accumulation [33]. NP could be absorbed by using microalgae *Phaecystis globosa*, *Nannochloropsis oculata*, *Dunaliella salina*, and *Platymonas subcodicormis*. Furthermore, it is removed through absorption, which is the process of NP being taken up into cells, while adsorption is the binding pollutants to the cell surface. In the four strains that were observed, both processes decreased along with the increasing concentration of NP [21].

Biodegradation of phenol compounds by microalgae occurs under aerobic conditions through a series of reactions to produce pyruvate and CO_2. The capability of *P. subcodicormis* to absorb NP from the medium is attributed to the recovery process of damaged chlorophyll when exposed to NP, showing the adaptation process. Moreover, the effectiveness of phycoremediation process depends on the successful adaptation of microalgae cells to the polluted environment, as the tolerance of the cells to pollutants determines the absorption rate. This shows that a higher growth rate of microalgae cells results in greater absorption. Meanwhile, pollutants content also caused stress to microalgae cells, leading to an increase in certain metabolites [21].

2.4 Estrogen waste

Estrogens are essential steroid hormones, existing in both synthetic and natural forms, produced by the endocrine system, which are crucial for brain function, reproduction, and bone growth. However, when estrogens contaminate aquatic systems, there is a possibility of negative effects on humans, fish, and animals. For example, their contamination with drinking water increases the risk of cancer and induces cardiovascular diseases [34]. Microalgae were reported to have the capacity to degrade estrogen compounds consisting of estrone, 17β-estradiol, and 17α-ethynylestradiol. *Haematococcus pluvialis*, *Selenastrum capricornutum*, *Scenedesmus quadricauda*, and *Chlorella vulgaris* also degraded estrogen pollutants causing disturbances in the human reproductive system by 97, 80, and 97%, respectively. These strains were cultivated using blue-green medium (BG11) supplemented with estrogen at a concentration of 5 mg/L for 40 days Although 17β-estradiol can be absorbed by the four microalgae completely, 17α-ethynylestradiol was absorbed by *Haematococcus pluvialis* and *Scenedesmus quadricauda* at a rate of 85%. The absorption of estrogens through a biotransformation process occurs by breaking the estrogen ring through several pathways to form simpler compounds [19].

Selenastrum capricornutum has been reported to effectively absorb 17-estradiol (E_2) and 17α-ethinylestradiol (EE_2) from wastewater. The efficiency of E_2 and EE_2 absorption by these strains ranged from 88 to 100% and 60 to 95%, respectively, with an initial concentration of 5 mg/L for 7 days. The strain also absorbed 15% tris (2-butoxy ethyl) phosphate (TBEP) and 22% bisphenol A (BPA). Furthermore, E_2, EE_2, BPA, and TBEP were absorbed for 7 days by *C. reinhardtii* with a removal efficiency of 86, 71, 22, and 15%, respectively [17].

2.5 Phosphorus, ammonium, phosphate-containing wastewater

Phosphorus is used as a structural compound of cell membranes and DNA for microalgae cells. In previous research by Sharma and Khan [35], *Chlorella minutissima* showed a higher efficiency compared to *Scendesmus* spp, *Muscorum* spp., and *Nostoc* spp. to remove nutrition in Indian Agricultural Research Institute (IARI) and common effluent treatment plant (CETP) wastewaters within 12 days. The removal efficiency for TDS, nitrogen, phosphorus, BOD, and COD was 97%, 90%, 70%, 95%, and 90%, respectively. *C. minutissima* had also succeeded in reducing TDS 96%, BOD 27%, and COD 31%, with high biomass and lipids, making it an effective phycoremediation agent [36]. The research conducted by Apandi et al. [37] on wet market wastewater showed that organic content such as TP, TN, and total organic carbon (TOC) decreased by 75%, with the cell density of *Scenedesmus* sp. cultured in waste were 10^6-10^7 cells mL^{-1} for ± 33 days.

2.6 Diazinon waste

Diazinon waste (O,O-diethl O-(2-isopropyl-6-methylpyrimidin-4-yl) thiophosphate) is widely used in the agriculture field. Some microalgae, including *C. ellipsoidea, E. elastica,* and *Chlamydomonas,* experience disruptions in their photosynthetic processes due to diazinon exposure [38]. However, at a higher concentration, the growth of microalgae was slowed down by 72% due to the accumulation of diazinon, which can damage the cell compartment [16,38]. The metabolism of diazinon in *C. vulgaris* is facilitated by the phosphatase enzyme, which catalyzes diazinon to form two other compounds with lower toxicity, 2-isopropyl-6-methyl-4-pyrimidinol and diethyl thiophosphate. Additionally, diazinon can serve as a carbon and phosphorus source for *C. vulgaris* cells [16].

2.7 Atrazine waste

Similar to diazinon waste, atrazine is a commonly used pesticide for corn crops, sorghum, and sugarcane, which is widely recognized for its gradual decomposition by water, sunlight, and microorganisms [39]. *Chlamydomonas mexicana* has a relatively high tolerance for atrazine exposure, achieving a reduction rate of 64% in the medium with an initial concentration of 100 g/L within 14 days. However, this pesticide reduced the concentration of chlorophyll-a in the microalgae cell by inhibiting the synthesis of the chlorophyll precursor, protoporphyrin IX. This removal was attributed to the accumulation of atrazine in the cells, which would be higher along with the increased cell growth in certain phases. Exposure to 100 g/L atrazine decreased the saturated fatty acid and increased the unsaturated fatty acid. Meanwhile, by increasing the concentration (25-100 g/L), the carbohydrate content increased by 47-52%. This showed that atrazine stress enhanced carbohydrate synthesis in the microalgae cells [15].

2.8 Pharmaceutical waste

Pharmaceutical waste is pollutants resulting from both medical activity and manufacturing processes. This waste consists of two kinds, namely liquid and solid, posing a significant threat to the aquatic systems. One of the concerning contaminants issulphamethoxazole, with surface water contamination reaching 1900 ng/l [40]. Other microalgae such as *Nannochloropsis* sp. have been successfully investigated by Encarnação et al. [20] due to their efficacy in reducing harmful pharmaceuticals such as paracetamol, ibuprofen, and olanzapine which are resistant to photodegradation, and can damage aquatic ecosystems. *Nannochloropsis* sp. cell worked effectively on the polyvinyl alcohol (PVA) matrix and degraded the pharmaceutical ingredients within 24 h, achieving a reduction rate of 12.07, 12.7, and 32.4%, respectively. *Nannochloropsis* sp. had high durability and halotolerant characteristics, making it suitable for remediating environments with elevated levels of pharmaceutics content.

3. Mechanisms of phycoremediation

Environmental remediation with microalgae is facilitated by several methods, including biosorption, consumption, and biodegradation.

3.1 Biosorption

Biosorption is a physical process in which pollutants can adsorbed, complexed, or precipitated on the surface of algal cell walls [41,42]. As a potential biosorbent, algae possess several specialties, including the presence of multiple functional groups on the surface of cell walls, facilitating the ability to bind with various kinds of pollutants. The effectiveness of biosorption is affected by the existence of several binding molecules such as amine, carboxyl, hydroxyl, sulphuryl, phosphoryl, carbohydrate, imidazole, and others [43]. Additionally, monovalent ions such as H^+, Na^+/K^+, and similar ones show higher efficiency in adsorbing toxic substances compared to divalent ions [44].

All biosorption mechanisms are interconnected and can occur through metabolic or non-metabolic microbial activity, despite their different classifications. Non-metabolism-dependent mechanisms are mainly facilitated by rapid and reversible physical-chemical interactions between metals and the functional groups on the cell surface [46]. Meanwhile, intracellular uptake phenomena, known as bioaccumulation, are influenced by cellular metabolism at slower rates [47]. The precipitation of metals is affected by microbial metabolism, occurring when microorganisms produce compounds that facilitate the process [48]. This process can also occur during a simple chemical interaction between metals and the cell surface [49].

In the metabolism-dependent process, biosorption is categorized into two groups. The first is passive biosorption, which refers to the process where substances, nutrients, or pollutants in wastewater interact with the cell wall of dead biomass through the functional groups [50]. This process is independent of metabolism and occurs rapidly within a timeframe of 5 to 10 min from the initial interactions [51-53]. Meanwhile, active biosorption, known as bioaccumulation, occurs within wet biomass or living cells, consisting of a two-step process. The first step is adsorption, resembling passive biosorption, which is a fast metabolic process independent of the binding substances. The second step is slow in metabolism and depends on the binding affinity, facilitated by the transport of bonded substances across the cell membrane [51,52].

3.2 Consumption

Consumption is a bioaccumulation mechanism possessed by microorganisms, including microalgae, in the remediation of pollutants from polluted environments. Certain pollutants, containing various nutrients, are subjected to metabolization by microalgae cells through specific enzymatic reactions according to each pollutants [54]. Wastewater containing organic pollutants in the form of nutrients is easily consumed by microalgae cells which will be used as nutrients for their biomass growth. The presence of these nutrients supports the growth of microalgae heterotrophically [55].

Euglena sp. is a strain capable of using excess nutrients in a polluted environment as a source of nutrition for its growth. Organic waste rich in nutrients, such as tofu liquid containing abundant sources of phosphate and nitrogen, has been reported to increase growth, biomass, and accumulation of lipids, proteins, and carbohydrates in pigments [6]. Pollutants with heavy metals can also be used by microalgae to increase their metabolite products when present in specific concentrations. This is because certain metals are microelements needed for microalgae to increase the growth of their biomass and metabolic products [5]. For example, *Pseudochlorococcum typicum* showed an increase in protein and chlorophyll-a concentrations when small concentrations of Hg, Cd, and Pb were added to the medium [5,56].

3.3. Biodegradation

Microalgae are producers of various enzymes crucial for the biodegradation of organic pollutants and the production of biosurfactants useful in hydrocarbon waste bioremediation. Moreover, biodegradation is recognized as essential method for removing pollutants from effluents using bacteria [57]. The process of organic waste using microalgae, is a natural technique friendly to the environment compared to chemical, physical, and others [58]. Generally, microalgae have the ability to break down organic materials in water into tiny molecules, which can be used as nutrients for their growth [59]. Several research have reported the ability of algae to transform pollutants into intermediates or enhance the degradation capabilities of nearby microbial communities. Microalgae show the capability to decompose plastic waste such as polyethylene, polystyrene, polypropylene, and polyvinyl chloride [60]. In the marine environment, degradation of low-density polyethylene layers can also occur by decreasing weight and the appearance of cracks, holes, and surface erosion of low-density polyethylene due to enzymatic activities [61]. Furthermore, there is an increased biodegradation rate of caffeine, with a 100% efficiency rate [62], enhancing crop productivity by decomposing pesticides. Scenedesmus sp. is well-adapted for wastewater treatment, effectively removing thiamethoxam and conventional pollutants. Biodegradation is also applied to diclofenac pharmaceutical product contaminants that are contained in feces. This pollutants can be removed by *Picocystis* and *Graesiella,* with DCF removal efficiencies reaching 73% and 52% of 25 mg/L as the initial concentration [63].

Microalgae play a crucial role in absorbing CO_2 from the environment to produce organic materials. These materials serve as nutrients for bacteria, supporting their cell metabolism and oxygen production. Bacteria produce CO_2 through the cellular respiration process, which becomes a valuable photosynthetic material for microalgae. Furthermore, bacteria synthesize various compounds that support the growth process of cells, such as vitamins. This symbiosis will produce antioxidants that protect bacterial and microalgae cells, thereby enhancing the metabolic process by absorbing nitrogen and minerals from the environment [64-66]. Co-culture between *C. vulgaris* and *Enterobacter* sp. increased the percentage of heavy metal absorption, including chromium (Cr) by 62%, cadmium (Cd) by 37%, copper (Cu) by 60%, and lead (Pb) by 63% from the concentration of each metal in the medium of 20% [66].

Microorganisms, such as microalgae, are potential biotransformation agents due to their rapid growth, high surface-volume ratios, and metabolic rates. Biotransformation is the process of converting high-toxicity pollutants into less toxic states, facilitated by enzymes obtained from mitochondria and lipophilic membranes in the endoplasmic reticulum. One of essential enzymes in this process is alcohol dehydrogenase, which converts ethanol to acetaldehyde. However, the biotransformation process can also occur in the absence of enzymes, when the material is physically stressed, such as exposure to extreme pH, leading to alterations in the chemical structure [67].

Azo is a commercial synthetic dye containing aromatic ring compouds and is commonly used as a textile dye [68]. The biotransform of these compounds into aromatic amines is facilitated by microalgae through the catalyzation of the azo reductase enzyme. The aromatic amine is further processed to form simpler components and CO_2. Moreover, the biotransformation of benzene compounds by microalgae occurs through the conversion of benzene into phenol catalyzed by the dioxygenase enzyme. Phenol compounds are converted to catechol by the enzyme phenol monooxygenase. However, direct benzene biotransformation is facilitated by the enzyme dioxygenase/dehydrogenase to form catechol. The catechol compound is further converted to form cis, cis-muconic acid catalyzed by the catechol 1,2-dioxygenase enzyme, and 2-hydroxymuconic semialdehyde catalyzed by the catechol 2,3-dioxygenase enzyme. Subsequently, 2-hydroxymuconic semialdehyde is converted to form 4-oxalocrotonate through the enzyme 2-hydroxymuconic semialdehyde-NAD^+-dependant dehydrogenase. This process is followed by the conversion of 4-oxalocrotonate through a series of reactions that produce pyruvate and CO_2 [69].

Nitrite and ammonium at very high concentrations can be toxic to microalgae cells potentially damaging the photosynthetic system in microalgae cells. However, microalgae cells have a specific ability to assimilate nitrite and ammonium in cytosol, effectively reducing toxicity. During absorption, the reductase process takes place to produce nitrate, which is then transported into the chloroplast. The accumulation of nitrite in the chloroplast can damage photosystem II and disrupt the reaction. This disruption can be overcome by converting nitrite directly into ammonium through nitrite reductase. The ammonium is also used by cells as a non-toxic nitrogen source through glutamine synthetase-glutamine oxoglutarate aminotransferase (GS-GOGAT). Meanwhile, suboptimal ammonium assimilation can be toxic to the photosynthetic system of the cell [70]. In *Chlorella*, the assimilation of ammonium by GS-GOGAT is high, which allows the effective conversion of ammonium into organic N-source, reducing the risk associated in the chloroplast [71,72].

GS-GOGAT is a primary and secondary ammonium assimilation pathway. GS plays a significant role in ammonia and glutamate ligation to form glutamine which requires adenosine triphosphate (ATP). Meanwhile, GOGAT is functional in the redox reaction responsible for the transfer of amide from glutamine to ketoglutarate. Consequently, GS-GOGAT provides a pathway for the assimilation of inorganic N into various types of organic N components in the cells [73,74].

Co-cultures of microalgae and bacteria have shown the ability to absorb low-sulfur diesel hydrocarbons, showing their potential for hydrocarbon waste absorption in the environment. The symbiosis between microalgae and bacteria can stimulate the degradation and absorption of diesel oil waste in the aquatic environment. At the beginning of the bioremediation process, bacteria and cyanobacteria significantly influence the biodegradation of these hydrocarbon compounds. Meanwhile, in the final stage, there will be an increase in microalgae population due to the ability of microalgae cells to absorb hydrocarbon compounds that are less bio-available from the medium. When the density increases, the bacteria population decreases, showing that a higher mass of microalgae cells will cause a reduction in the number of hydrocarbons in the medium [75].

4. Conclusion

In conclusion, this research showed that phycoremediation served as a safe and environmentally friendly method for restoring the environment from organic pollutants. Furthermore, its biomass can be used as a source of biodiesel [5], lipid, biofertilizer, and biohydrogen production. Several investigations have reported the use of pollutants such as ammonium, phosphorus, and phosphate in sewage by microalgae. Wang et al. [76] reported that microalgae *Chlorella* sp. reduced the content of NH_4^+ by 83% and PO_4^{3-} by 90% in municipal waste. *Chlorella vulgaris* and *Spirulina platensis* were also found to remove nitrate and phosphate in simulated aqueous solutions to enhance the removal of pollutants.

Parachlorella kessleri, *Desmodesmus* sp., and *Chlamydomonas* sp. have shown potential for bioethanol production, particularly when isolated from pig farms for swine wastewater remediation. These isolates have carbohydrate contents of 40, 37, and 49%, with a COD reduction rate of 47, 38, and 47%. Bioethanol production is facilitated by enzymes such as glucoamylase, -amylase, -glucosidase, endoglucanase, and cellobiohydrolase. On *Chlamydomonas* sp., the maximum ethanol concentration was 61 g/L [77], with biomass of *Desmodesmus subspicatus* at 1277.44 mg/L when treated with CO_2. The lipid content in the biomass reached 18%, with lipid content in the form of palmitic acid (C16:0) and oleic acid (C18:1) [78].

Several criteria for microalgae strains to be considered suitable candidates for biofuel-producing lipid sources include rapid growth rate, high lipids content, adaptability to various environmental conditions, resistance to contamination, ease of harvest, and compatibility with modern lipid extraction methods, and potential for use in various biorefinery activities [79]. Therefore, phycoremediation optimization is needed to maintain environmental quality from organic pollutants and produce biomass as feedstock in biorefinery activities. To facilitate the effective growth during phycoremediation, environmental factors must be considered, such as water, salinity, light, nutrition, and pH. Light and carbon dioxide play an important role in supporting the photosynthetic process of microalgae, while salinity affects their resilience in sustaining life and cell metabolism. Although certain microalgae are tolerant to high salinity, freshwater strains are not resistant. The levels of pH also influence the enzymatic activity, as optimal value enhances their growth. Water is the main medium for the growth of microalgae and requires control over its physical, chemical, and biological factors. The growth of microalgae in unfavorable conditions, for example, when cultivated in media such as pollutants, can be assisted by adding certain nutrients, including nitrogen and phosphorus, both exogenously and endogenously. When these environmental factors are managed effectively, microalgae grow optimally, ensuring the maximum potential of phycoremediation process for organic pollutants.

5. References

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