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Review

# Analysis of the Status and Improvement of Microalgal Phosphorus Removal from Municipal Wastewater

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**Abstract:** Phosphorus, as one of the main pollutants in municipal sewage, has received increasing attention recently. Phosphorus recovery also increases the sustainable development of municipal wastewater. Since algae have the ability to effectively redirect nutrients, including phosphorus, from municipal sewage to algae biomass, municipal sewage treatments involving microalgae have piqued the interest of many researchers. The phosphorus removal depends on the potential of the microalgae to absorb, preserve, or degrade phosphorus in municipal wastewater. It is, therefore, of great interest to study the mechanisms underlying the absorption, storage, and degradation of phosphorus by microalgae to ensure the viability of this phosphorus removal process in wastewater. The objectives of this review were to summarize phosphorus metabolism in microalgae, examine key external and internal factors impacting phosphorous removal by microalgae from wastewater, and examine the status of phosphorous-metabolism-related research to improve our understanding of microalgae-based municipal wastewater treatments. In addition, the methods of recovery of microalgae after phosphorous removal were summarized to ensure the sustainability of municipal wastewater treatment. Finally, a potential approach using nanomaterials was proposed to enhance the overall phosphorous removal performance in municipal wastewater through the addition of nanoparticles such as magnesium and iron.

**Keywords:** microalgae; municipal wastewater; phosphorus removal; immobilization and recycle technology

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## 1. Introduction

Cities are generally densely populated and have great demands for water. For some cities, the efficient treatment of urban sewage to achieve water recycling has become a strategic solution to water shortages [1]. In China, the amount of wastewater treated has increased by more than 50% in the last decade, and the increase in the volume of wastewater has significantly endangered human and environmental welfare [2]. The fecal sludge collected by the sewage systems in some industrial cities is untreated, causing significant damage to water supplies across such cities [3]. While urban sewage treatment in developed European countries strongly reduces pollution in wastewater, the amount of sludge generated is still growing year on year. Finding methods to efficiently treat sludge is an urgent challenge [4]. In comparative studies and simulation forecasts regarding the statuses of cities in Nanjing (domestic), South Asia, and Southeast Asia, the results indicated that by 2030 the water quality in samples sites in Manila and Jakarta will have deteriorated further [5].

Phosphorus is an essential basic element in organisms that is also commonly present in water bodies and primarily occurs in a dissolved form and in association with other particles via chemical adsorption; however, regardless of whether it originates from the natural world or via human activities, phosphorus significantly impacts the environment [6]. Excessive phosphorus in a water body, for example, accelerates the growth of algae and other microorganisms, subsequently creating nutrient imbalance in the water body and accelerating eutrophication [6]. Some phosphorous-containing compounds in water can be converted into minerals and may block water supply lines and sewage treatment facilities [7,8]. In municipal drainage systems, phosphorus originates primarily from domestic sewage, with 85% of the total phosphorus entering the wastewater from domestic sewage containing human excrement and detergents [6]. Phosphorus-containing sewage compounds in urban life exist mainly in the form of orthophosphoric acid, tripolyphosphate, and pyrophosphate [9,10]. Modern technologies used for the elimination of phosphorus are primarily categorized into biological, chemical, and physical technologies [11]. The removal and recovery of phosphorus from municipal sewage not only provides conditions for the utilization of phosphorus resources, but also reduces the eutrophication and increases the sustainability of municipal sewage; however, many conventional phosphorous reduction processes used for urban sewage entail high running and maintenance costs and are not sustainable, and may even cause other pollution to the water body [12,13].

Phosphorus also has numerous constructive uses. For example, certain phosphates can be applied to water supply pipes, where they combine with heavy metals in rainwater, lowering the concentration of heavy metals in the water while still acting as a corrosion inhibitor [14,15]. To avoid direct interactions between the metal and the food in metal food containers, phosphides are often added to form an inert coating [16]. Furthermore, household detergents, toothpaste, and shampoos are often also incorporated phosphorous compounds to improve their washing efficiency, with sodium tripolyphosphate being the most popular phosphorous-containing compound in detergents [17]. The overall amount of phosphorus in the world is small and nearly 40 million tons of phosphorus is absorbed worldwide per year, making the recycling of phosphorus critically significant [18].

CiteSpace is a tool used for the analysis of scientific literature. It can help an author to explore research hotspots and research frontiers in a certain research field and to predict future development trends. The Web of Science database was used to perform a comprehensive study of the literature, using the search terms "microalgae" and "phosphorous removal". In total, 678 manuscripts published in the past five years were retrieved and a visual study of the keywords in the manuscripts was carried out using CiteSpace (version 5.7.R2), with the results shown in Figure 1. Based on the findings of the search, CiteSpace was used again to analyze clusters, with the results shown in Figure 2. Figures 1 and 2 demonstrate that the study hotspots regarding the reduction of phosphorus by microalgae focus primarily on the treatment of municipal sewage, recovery, and resource utilization. The results of this analysis provided theoretical significance for this review of relevant research frontiers regarding the treatment of phosphorus in urban sewage and the prediction of future development trends for microalgal phosphorus removal technology.

This review summarizes phosphorus elimination from municipal wastewater by microalgae and analyzes the factors influencing this phosphorous removal and the associated methods. The aim of this review is to further understand the process of phosphorus removal by microalgae in municipal wastewater, so as to optimize the process and sustainability. In this paper, the problems related to microalgae dephosphorization are introduced for researchers who are interested in municipal wastewater dephosphorization technology.

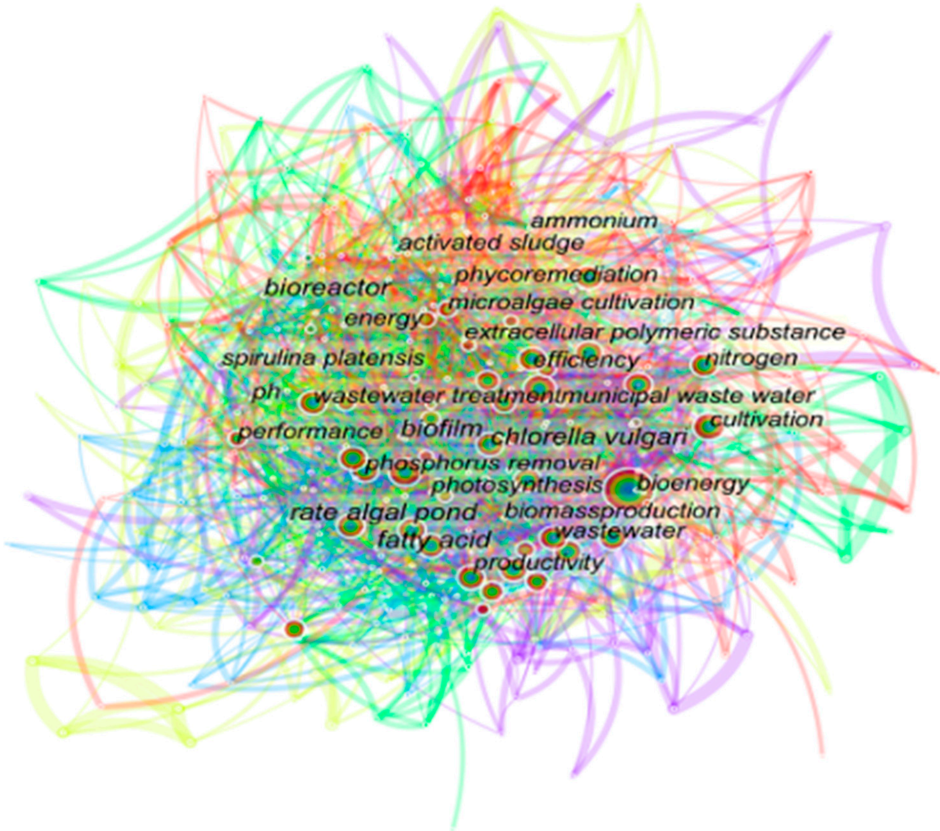


**Figure 1.** CiteSpace visual analysis diagram.

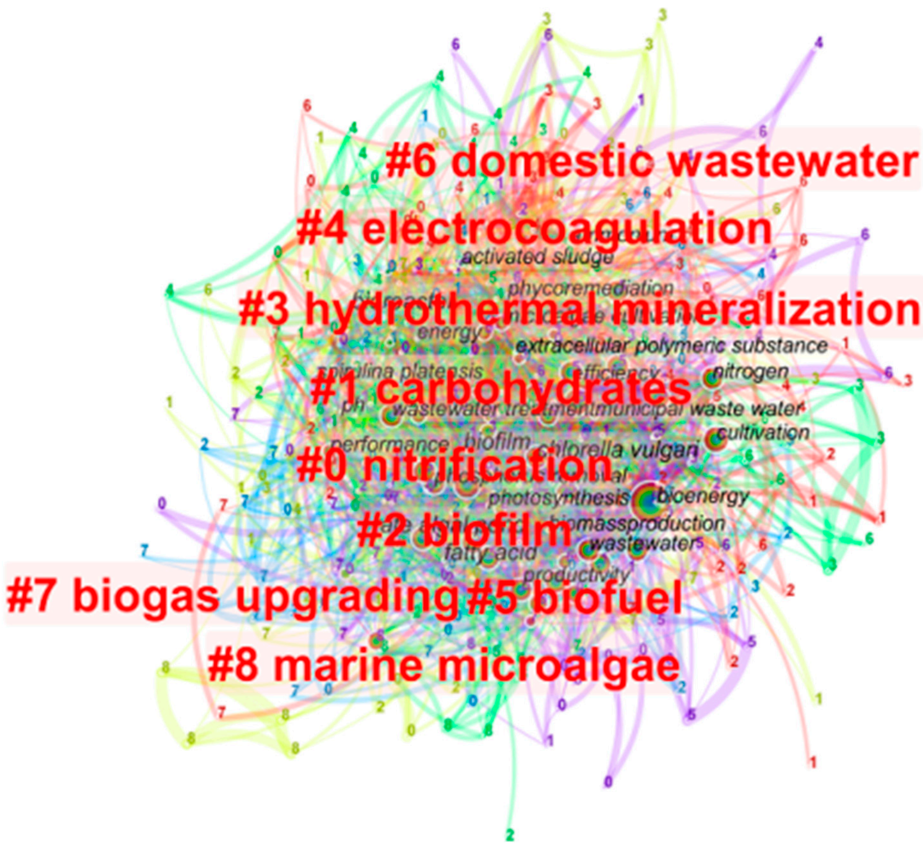


**Figure 2.** CiteSpace cluster analysis diagram.

## 2. Various Systems and Strategies for the Reduction of Phosphorus in Municipal Wastewater

Phosphorus removal from municipal sewage is mainly categorized into three categories of processes: physical, chemical, and biological. The respective technological methods and respective advantages and drawbacks of each process are shown in Table 1. Physical phosphorus removal technologies can eliminate all forms of particulate phosphorus compounds [19]. Membrane technology not only extracts phosphorus from complete suspended solids but also removes dissolved phosphorus. Membrane bioreactors (MBRs) and reverse osmosis (RO) devices with good phosphorous removal capacity have been widely used in full-scale sewage treatment plants [20]. The fundamental theory behind the elimination of chemical phosphorus is to crystallize or condense phosphorous compounds by adding chemical agents or by modifying certain reaction conditions. For example, in the coagulation and flocculation process, phosphorous-containing materials are flocculated by the addition of polymers or metal ions. This approach is effective for removing larger molecules, and its quality is determined by the charge of the salt ions [21]. Since biological phosphorus removal is used to treat municipal sewage, anaerobic or anoxic treatment is usually required first, followed by aerobic treatment and other procedures to remove phosphorus from activated sludge in municipal sewage. Microorganisms have been shown in studies to have the largest reduction effect on total phosphorus under anaerobic conditions, with removal rates exceeding 80–90% [21]. Compared to the large operational and repair costs of conventional physical and chemical phosphorous removal systems and the complexities of certain biological treatment processes, the use of microalgae has proved to be a cost-effective and long-term alternative for biological phosphorus removal, which is now commonly utilized [22]. The following is an introduction to the microalgae dephosphorization technology mechanism.

### 2.1. Microalgae Culture Methods

There are two types of microalgae culture structures: open and closed systems. The term "open systems" refers to growing systems of outdoor waters, such as lakes and reservoirs. In order to provide adequate light for the microalgae, the system's water depth is usually no greater than 0.5 m [23]. While the open system layout is simple and easy to manage, nutrients can become diluted by pollution when exposed to the open environment over a long period [24]. Closed devices are segregated from the external environment, thereby shielding the system from the harmful effects of the external environment. Photobioreactors (PBRs) are widely used as closed structures for microalgae cultivation. PBRs are usually classified into stubs and flat plates, and are generally made from glass or plastic, air, carbon dioxide ($CO_2$), and other gases may be fed into the PBRs [23,25]. On the basis of being highly controlled, the closed method can be used to evaluate the characteristics of microalgae and the effects on the purification of wastewater under different sets of conditions. It also provides a culture system for improving the conditions for the absorption of phosphorus by microalgae in urban wastewater.

Microalgae culture modes can be separated into continuous and semi-continuous batch modes [24]. A lot of the management costs are avoided in closed batches because the culture material does not always need to be replaced. The growth of the microalgae, however, will be inhibited if the nutrients in the batch system are depleted or if certain factors occur, such as cell self-shading, pH variations, and contamination, inhibiting the growth. In addition, the device must ensure a successful exchange of gas [23]. Compared to the batch model, the semi-continuous model can achieve higher biomass despite the need for periodic substitution of culture material and the continuous removal of wastewater [26].

### 2.2. Phosphorus Uptake and the Metabolism Mechanism of Microalgae

The absorption and metabolism of phosphorus by microalgae are often distinct for different types of phosphorus or under different environmental conditions. Microalgae can induce phosphatase to absorb external organophosphorus and synthesize high-affinity inorganic phosphorus transporters to assist inorganic phosphorus absorption [27]. The absorption of inorganic phosphorus also relies on the inorganic phosphorus charge and the pH of the microalgae cell membrane [28–30]. In general, the lower the molecular charge, the higher the bioavailability of inorganic phosphorus for the microalgae [28]. Most microalgae assimilate inorganic salts such as $HPO^{-}$, $HPO^{2-}$, and $PO_4^{3-}$ [31].

Polyphosphates include acid-soluble and acid-insoluble polyphosphates. Although certain microalgae do not use polyphosphates as their primary supply of phosphorus [32], in the absence of phosphorus, microalgae can assimilate and metabolize polyphosphate [31]. In addition, under the condition of excess inorganic phosphorus, microalgae can take up excess phosphorus and deposit it in the form of insoluble polyphosphate acid, where it can be used for cell metabolism when inorganic phosphorus is lacking [33]. Excessive phosphorus and high light intensity in municipal wastewater tend to facilitate the removal of phosphorus by microalgae [31,34]; however, some studies have shown that excessive phosphorus can impede the growth of some microalgae due to excessive accumulation of polyphosphate in the cells [35].

Photosynthesis is the basis of the metabolism of microalgae. Over the entire photosynthesis process, phosphorus is required in the reaction that produces the energy substance ATP. The equation for this reaction is as follows:

$$2\,H_2O + 2\,NADP^+ + 3\,ADP + 3\,P_i + 8\text{-}10\text{ Photons (light)} \rightarrow 2\,NADPH + 2\,H^+ + 3\,ATP + O_2$$

To summarize, the electrons in the water are transferred to $NADP^+$ after the absorption of light energy by the microalgae. $H^+$ formed by water allows ADP and inorganic phosphorus to form ATP on the thylakoid membrane in the cell [31]. Phosphorus is also important for the synthesis of DNA, RNA, and cell membranes [36].

**Table 1.** Different physical, biological, and chemical phosphorus removal technologies used in wastewater treatment processes [20,37–39].

| Methods | Technologies | Advantages | Disadvantages |
|---|---|---|---|
| physical methods | physical absorption | widely used for phosphorus removal | not yet perfect for phosphorus adsorption |
| | sand filtration | removes all P compounds | only for the primary stage |
| | the membrane purification | simple and efficient | high operation and maintenance costs |
| | ion exchange | can treat hazardous waste and higher concentrations of phosphorus | lack of selectivity for specific ions and complex process |
| chemical methods | by precipitation of metal salts and lime | high phosphorus removal efficiency and economical | may cause secondary contamination |
| | crystal | reusable, little environmental harm | need to add chemicals and low stability |
| | Coagulation and flocculation | can be used for reaction by adding metal ions such as polymers or aluminum | need high charge for salt ions |
| biological methods | artificial aeration | mainly used for dephosphorization of lakes | no significant effect in shallow lakes |
| | enhanced biological phosphorus removal | no chemicals need to be added | low stability and biological population competition |
| | photosynthetic microorganisms immobilized on cellulose, ceramic, or gel carriers | can effectively immobilize and remove more than one type of microorganism or contaminant | not easily removed for most phototrophs |
| | phosphoric acid binds proteins | can work in low phosphorus environments | the use of this protein is limited |

## 3. Factors Impacting the Elimination of Phosphorus from Municipal Wastewater by Microalgae

There are several internal and external factors influencing the removal of phosphorus by microalgae, such as the temperature, strength and period of illumination, and pH [31,40]. These common factors have been extensively studied and summarized, although a review regarding their impacts on the treatment of urban sewage by microalgae is lacking. In this chapter, the impacts of microalgae on the removal of phosphorus in municipal wastewater will be examined in terms of the hydraulic retention time, ratio of nitrogen to phosphorus, carbon dioxide concentration, species of microalgae, and different types of wastewater.

### *3.1. Hydraulic Retention Time*

The hydraulic retention time (HRT) for microalgae in bioreactors influences their growth and phosphorous removal performance. An appropriate HRT not only improves the efficiency of the microalgae wastewater treatment but also reduces the operating and maintenance costs of the system. In the batch regime pilot scale photobioreactor system, activated microalgae sludge is used for the treatment of municipal sewage. Findings have shown that when the hydraulic retention time is 2–6 days, the phosphorous removal efficiency of the microalgae is improved by around 30% to 90% [41]; however, owing to the deterioration of the switch virtual interface (SVI) and other factors in the later stage, the phosphorous removal performance of the microalgae does not improve with the increase in hydraulic retention time [41]. Similarly, the optimum hydraulic retention time in the high-level algae pond also occurs on the sixth day when the bacteria and algae system is used to treat sewage [42].

The short HRT means that ammonia nitrogen ions in steam pools used for wastewater treatment cannot be fully nitrified [43–45]. By separating HRT and sludge retention times (SRT), a next-generation anaerobic–aerobic algal bioreactor was developed to solve the problem of inadequate HRT [44–46]. Toledo-Cervantes et al. [45] used a new form of photobioreactor involving hypoxia–aerobic algae to investigate the removal of phosphorus in water under varying hydraulic retention periods. Their findings revealed that when the HRT of the bioreactor decreased from 4 to 2 days, the removal rate of P-$PO_4^{3-}$ decreased from about 22% to approximately 11%. A schematic diagram of the photobioreactor involving anoxic–aerobic algae–bacteria is shown in Figure 3. Similarly, when a mixed-culture microalgae membrane bioreactor was used to treat secondary wastewater from municipal wastewater, the optimal retention time was decreased to 1 day [39]. In general, the optimum hydraulic retention time for the removal of phosphorus by microalgae is approximately 6 days. By enhancing the treatment technology, such as the selection of a suitable bioreactor, the hydraulic retention time can be reduced, along with the running and repair costs of the treatment.

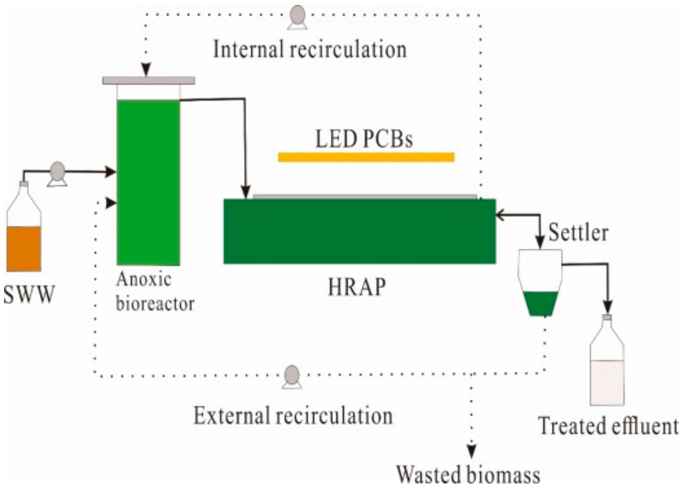


**Figure 3.** Schematic diagram of anoxic–aerobic algae–bacteria photobioreactor [45]. SWW: secondary wastewater; HRAP: open photobioreactor.

### 3.2. The Ratio of Nitrogen to Phosphorus

Nitrogen and phosphorus are important urban wastewater elimination indices, as well as important microalgae nutrient sources. An appropriate N/P ratio provides a good growth environment for microalgae and increases the phosphorous removal performance of the microalgae. The optimal N/P ratio range for microalgae development in freshwater ranges is 6.8 to 10 [47]. In the photobioreactor method, when microalgae are used to treat urban wastewater, the optimum N/P ratio range for the elimination of total phosphorus is 5–30 [48].

Molazadeh et al. [49] performed post-screening, biological treatment, and disinfection treatment of wastewater drained from sedimentation tanks of urban wastewater treatment plants. They controlled the N/P ratio and $CO_2$ concentration through the injection of potassium dihydrogen phosphate and used a high $CO_2$ concentration. The analyses showed that *Chlorella vulgaris* demonstrated strong potential to remove phosphorus under all concentrations of $CO_2$ and N/P, exhibiting a removal range of 70.0–96.0%. Under conditions with 16% $CO_2$ and a ratio of 10:1 N:P, algae biomass was the highest, with an increase in lipid productivity, which makes a powerful contribution to the eventual recovery of microalgae for biofuel [49]. To summarize, the reduction of total phosphorus from urban wastewater by microalgae is not only related to the required N/P ratio but is also directly proportional to the biomass of the microalgae [48]; however, as microalgae process urban waste, the feedbacks between the N/P ratio, the concentration of $CO_2$ in the water body, and the regulation of the optimum concentration remain unclear [49].

### 3.3. Carbon Dioxide Concentration

Carbon is the most fundamental component of living things. When microalgae are used to treat urban waste, the source of carbon comes not only from sewage but also from $CO_2$ in the air. Shanshan Ma [50] added 10% $CO_2$ mixed gas to unsterilized sewage to support *Tetradesmus obliquus* PF3 for the treatment of sewage and nutrient recovery. Compared to the addition of air, sewage with 10% $CO_2$ added shows greater TP (99 $\pm$ 0%) removal performance under unsterilized conditions. This is due to the increased supply of carbon and the high concentration of $CO_2$ changing the pH to an optimal growth range (6.8–7.8) [50]. Chaudhary et al. [51] used Chlorella ATCC13482 for the treatment of urban wastewater in bubble column photobioreactors at a volume of 7 liters. The findings revealed that the rate of microalgae orthophosphate elimination with 5% $CO_2$ air was as high as 92.8%.

Increasing $CO_2$ concentrations not only enhances phosphorous reduction by the microalgae but also increases the biomass of the microalgae. The higher the biomass of the microalgae, the higher the phosphorous removal performance [48]. Studies have shown that in the case of *Nannochloropsis* sp., where the concentration of $CO_2$ was 15%, the biomass of the microalgae and intracellular lipids was dramatically increased [52]. In general, high $CO_2$ concentrations not only boost the phosphorous removal performance of microalgae but also improve the lipid content of microalgae cells and increase the recovery value of the microalgae.

### 3.4. Species of Microalgae

The optimal growth conditions for each microalgae species are different, such that suitable algae species are chosen for different initial concentrations of urban waste in order to achieve the maximum benefit of phosphorous elimination; however, in experimental or practical applications, a single type of algae is rarely used to treat municipal wastewater.

By using mixed microalgae for the treatment of municipal wastewater, dominant algae species can be chosen on the basis of the sewage characteristics. For example, Toledo-Cervantes et al. [45] increased the rate of phosphate elimination from a water body from about 10% to around 50% by reducing the C/N ratio from 9 to 7. At the same time, *Chlorella vulgaris*, the dominant species of algae, was eventually replaced by *Phormidium* sp. [45]. This type of research approach can reliably and efficiently find appropriate microalgae for certain sewage treatment plants by screening the dominant algae species and adding mixed microalgae for realistic conditions.

More studies have shown that relative to single algae species in wastewater treatment, there is a cooperative or competitive partnership between mixed algae species, resulting in biodiversity and making the treatment system more stable and efficient. Paches et al. [53] performed batch and mixed cultures for four types of microalgae using anaerobic membrane bioreactors. Their findings showed that the mixed microalgae culture could increase the rate of phosphorous removal and the productivity of water by letting the species compete with each other [53]. Devi et al. [54] also revealed through their research that mixed microalgae showed a high degree of phosphorous elimination in wastewater and concluded that using mixed cultures was one of the better methods to handle municipal wastewater and other low-toxicity wastewater.

### 3.5. Different Municipal Wastewater Treatment Technologies

In a city sewage treatment facility, the municipal wastewater can be separated into three levels based on the extent of treatment, ranging from primary wastewater treatment, where the wastewater has not yet been deeply treated, up to tertiary wastewater treatment, in which the wastewater is at the final cleaning process. Although primary wastewater exhibits several negative factors, such as high optical density (OD) and bacterial contamination, the concentration of nutrient species in primary wastewater is much higher than in other wastewater treatment levels, making it more favorable for microalgae development [50,55]. Secondary wastewater is partially treated and most of the nitrogen at this stage is available as nitrate due to nitrification. This is a negative factor for microalgae, as microalgae preferentially absorb nitrogen in the form of ammonia [50]. Bellucci et al. [56] used microalgae to treat secondary wastewater and evaluated the combined function of microalgae as a disinfectant and nutrient remover. Their findings revealed that the microalgae contributed to an E. coli count equal to that of standard ultraviolet therapy in the batch disinfection test, and that the count was smaller than that of light experiments without microalgae. The E. coli population decreased by an order of magnitude in subsequent continuous studies. The rate of elimination of total phosphorus in the secondary wastewater was 100%. In addition to the level of municipal wastewater treatment, the forms of municipal wastewater can also be categorized according to the special new wastewater created by the treatment process. Various processes and techniques are used to treat different forms of wastewater with microalgae, as shown in Table 2.

Anaerobic digestion is a mechanism that converts polluted waste to energy materials; however, this method volatilizes harmful gasses such as high-viscosity, high-moderation, and highly volatile fatty acids. In addition, these reactive compounds are also poisonous to plants and microalgae. Products from untreated anaerobic digests cannot, therefore, be released immediately into nature [57]. The immediate discharge of untreated anaerobic digestive fluid into bodies of water can cause eutrophication [57]. The integrated technology of using microalgae to treat digested products is a technology that can offer economic gains, while also being environmentally sustainable [58]. In regards to the uptake of nitrogen and phosphate from wastewater, algae have demonstrated higher removal efficiency than other microorganisms [59]. Ermis et al. [60] used an experimental batch sequencing device to investigate the use of mixed microalgae in the treatment of anaerobic liquor digestion. The digestive juice was diluted to 2%, 5%, 7%, and 10%, so that the original concentrations of ammonia nitrogen and phosphate in the digestive juice were regulated at 18.6–87.1 mg $L^{-1}$ and 1.85–6.88 mg $L^{-1}$, respectively. It was found that the absorption of nitrogen by mixed microalgae was 10 times greater than that of phosphorus. Based on a biokinetic coefficient of the phosphorus measurements, the reaction rate coefficient was 0.21 mg $PO_4$-P $mg^{-1}$ chl a $day^{-1}$ and the saturation constant was 2.94 mg $L^{-1}$, with a yield coefficient of 5.03 mg chl a $mg^{-1}$ $PO_4$-P.

The main goal of treating eutrophic water bodies is to remove organic and inorganic compounds from the wastewater. Nitrogen and phosphorus, however, are not readily eliminated [61]. The utilization of photosynthetic–autotrophic digestion by microalgae means that $CO_2$ or inorganic carbon in the water or air can be used as a carbon source and source of energy. Autotrophic microalgae release extracellular organic matter (EOM) that converts inorganic carbon to organic carbon, increasing the concentration of organic carbon in the water [62]. While microalgae do not specifically remove organic matter from eutrophic water sources, they can be mixed with bacteria and other heterotrophic microorganisms to treat bodies of water with high amounts of organic matter; this specific topic will be detailed in the next chapter.

Sludge ozone technology can not only degrade several refractory organic compounds so that the production of the sludge can be decreased to 50–100%, but no harmful by-products are generated during the application of this technology [63,64]; however, owing to the high concentrations of nitrogen, ammonia, COD, and heavy metals in the excess ozone sludge, an additional burden is placed on the sewage treatment plant, which decreases the effectiveness of the sewage treatment [64–68]. Lei et al. verified the possibility of growing algae in sludge-concentrated wastewater ozone. While generating biomass, the microalgae can also extract nutrients from the water [64]. Their findings revealed that the bacteria–algae system had greater elimination effects in terms of total phosphorus removal than the pure microalgae system, with the systems showing 93% and 53.9% elimination effectiveness, respectively.

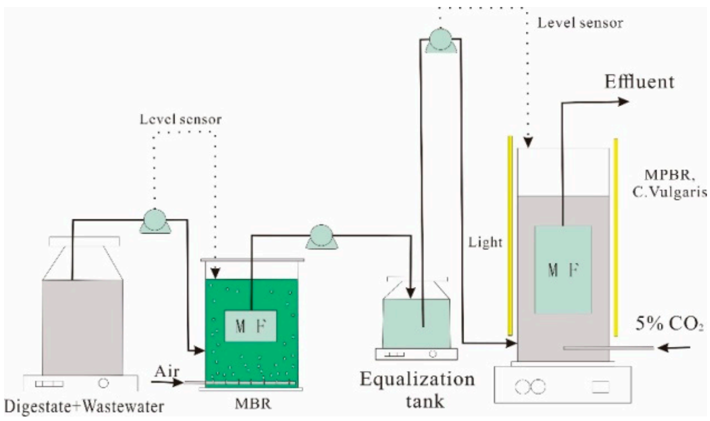


**Figure 4.** Schematic diagram of the MBR-MPBR experimental setup [72].

**Table 2.** Phosphorus removal effects of microalgae on municipal wastewater samples treated using different technologies.

| Sewage Source | Microalgal Species | Initial Conditions | Experimental Conditions | Results | Cites | Notes |
|---|---|---|---|---|---|---|
| General municipal sewage | Mutant *Chlorella* | After 121 °C autoclave treatment<br>$COD_{Cr}$: 190–230 mg/L<br>TP: 4.5–5.6 mg/L<br>TN: 40–60 mg/L<br>$NH_3$-N: 20–35 mg/L<br>pH: 6.6–7.6 | a symbiotic system of PAOs and bacillariophyta | absorb 3.05 mg/L phosphorus, keep TP below 0.46 mg/L | [69] | |
| Synthetic domestic wastewater | *Chlorella vulgaris* and *Phormidium* sp. | COD: 632 ± 45 mg/L<br>TOC: 196 ± 9 mg/L<br>IC: 195 ± 12 mg/L<br>TN: 43 ± 3 mg/L<br>N-$NH_4^+$: 24 ± 3 mg/L<br>P-$PO_4^{3-}$: 13.1 ± 0.8 mg/L | Anoxic–aerobic algal–bacterial photobioreactor structure | the maximum removal rate of P-$PO_4^{3-}$ was 47 ± 5% | [45] | low C/N ratio, Chlorella is the main algae, otherwise Phormidium SP will be dominant |
| | *Chlorella vulgaris* | COD: 300 mg/L<br>TN: 30 mg/L<br>TP: 10 mg/L | the new MAIFAS SBR | more than 51% phosphorus was removed without mechanical aeration | [70] | |
| Aerobic wastewater | Mixed microalgae collected in lakes | pH: 7.7 ± 0.2<br>TN: 99.5 mg/L<br>TP: 5.5 mg/L<br>COD: 475 mg/L<br>TOC: 245.6 mg/L<br>pH: 7.2 | Photoperiod:12 h/d, immobilized microalgae, operated at 5 different HRTS for 2–10 days | the removal rate of phosphorus was 93% | [71] | collected in an aeration tank of a distributed domestic sewage treatment plant based on ASP |
| Unsterilized sewage | *Tetradesmus obliquus* | N-$NH_4^+$: 28 mg/L | the mixed gas containing 10% $CO_2$ was added to the unsterilized sewage | The removal rate of TP was 99.0% | [50] | |
| Anaerobic digester | *Chlorella* sp. and *Scenedesmus* sp. | COD: 12600 ± 300 mg/L<br>TKN: 1692 ± 256 mg/L<br>$NH_3$-N: 900 ± 62 mg/L<br>$NO_3$-N: 0.13 ± 0.02 mg/L<br>TP: 105 ± 7.5 mg/L<br>$PO_4$-P: 64 ± 6 mg/L<br>TSS: 15880 ± 932 mg/L<br>pH: 9.00–9.15 | in an adaptive room with continuous illumination: 150 mol photon $M^{-2}$ $S^{-1}$, 25 ± 2 °C.<br>cultured at a dilution ratio of 2%, 5%, 7% and 10% | reaction rate coefficient: 0.21 mg $PO_4$-P$mg^{-1}$ CHl a $day^{-1}$, saturation constant:2.94 mg $L^{-1}$, yield coefficient: 5.03 mg CHL A $mg^{-1}$ $PO_4$-P | [60] | |

**Table 2.** *Cont.*

| Sewage Source | Microalgal Species | Initial Conditions | Experimental Conditions | Results | Cites | Notes |
|---|---|---|---|---|---|---|
| | *Chlorella* | activated sludge:<br>COD: 500 mg/L;<br>$NH_4^{+1}$-N: 40 mg/L;<br>$NO_3^-$-N: 2 mg/L;<br>$PO_4^{3-}$-P: 8 mg/L | treated in a membrane photobioreactor (MPBR) in a continuous mode | the removal rate of orthophosphate exceeded 99% | [72] | schematic diagram is shown in Figure 4 |
| | | anaerobic digester:<br>COD: 5–10 g/L;<br>$NH_4^{+1}$-N: 0.7–1.2 g/L;<br>$NO_3^-$-N: 90–300 mg/L;<br>$PO_4^{3-}$-P: 60–190 mg/L | | | | |
| Ozonation sludge wastewater | *Scendesmus* sp. is the dominant species | MLSS: 1500 mg/L<br>algae: sludge = 1:3 ($w/w$). | run for 10 days,<br>under 2500 lx on the inner wall of the reactor,<br>photoperiod: 12 h/d (from 5:00–17:00),<br>magnetic stirring rod (80 RPM) | the removal rate of TP was 53.9 ± 1.4%, higher than microalgae alone | [64] | sludge is obtained from secondary sedimentation tanks |
| Secondary wastewater from sewage treatment plants | Natural algal bloom (*Chlorella* mainly) | TP: 0.43 mg/L<br>TN: 7 mg/L<br>Mg: 0.45 mM<br>Ca: 1.12 mM | continuous bubbling tower photobioreactor (BCPBR), flocculation–precipitation method | the removal rate of total dissolved phosphorus was greater than 99% under continuous operation | [73] | |
| Secondary wastewater from sewage treatment plants | *Chlorella* | COD: 111 mg/L<br>pH: 7.9 ± 0.9<br>$NH_3$-N: 22 ± 2.6 mg/L<br>$NO_3$-N: 0.30 ± 0.42 mg/L<br>$PO_4^{-3}$-P: 3.2 ± 1.3 mg/L<br>Turbidity: 184 ± 23 FAU<br>*E. coli*: $4.7 \times 10^6 \pm 3 \times 10^6$ CFU 100 m/L | laboratory-scale photobioreactor,<br>10% of the effluent mixed with secondary effluent from a large municipal wastewater treatment plant,<br>tertiary disinfection by ultraviolet treatment | The removal rate of TP was 100% | [56] | |

**Table 2.** *Cont.*

| Sewage Source | Microalgal Species | Initial Conditions | Experimental Conditions | Results | Cites | Notes |
|---|---|---|---|---|---|---|
| Sewage discharged from sedimentation tanks of municipal wastewater treatment plants | Common *Chlorella* | $NH_4^{+1}$-N: 64.84 mg/L<br>$NO_3^{-1}$-N: 4.21 mg/L<br>$PO_4^{-3}$-P: 3.78 mg/L<br>COD: 82.00 mg $O_2$/L<br>pH: 8.52<br>Alkalinity: 91.80 mg $CaCO_3$/L | Through the different concentrations of $CO_2$ and different N/P ratios | Absorbance of 95.00% phosphorus for the medium supplemented under 16% $CO_2$ and N:P ratio of 10 | [49] | the wastewater was screened, biotreated, and disinfected. |
| Synthetic wastewater from municipal wastewater and laterite nickel mine | *Chlorella* | The two types of sewage were mixed in different proportions | temperature: 25 °C, light intensity: 4000 lux, Photoperiod:14 h/d, sterilized before experiment, added after sampling high-pressure deionized water of the same volume 6 times | The removal rate of TP was 39.3% | [74] | |
| Primary sedimentation tank wastewater | *Chlorella* | $NH_4^{+1}$-N: 25 ± 1.24 mg/L<br>TKN: 42.0.47 mg/L<br>$NO_3$-N: 2.5 ± 0.39 mg/L<br>sCOD: 156 ± 2.6 mg $O_2$/L<br>pH: 6.7 ± 0.05<br>DO: 3.5 ± 0.08 mg/L<br>sBOD: 65 ± 3.4 mg/L<br>TOC: 45.3 ± 1.12 mg/L<br>TIC: 1.24 ± 0.07 mg/L<br>TN: 46 ± 1.25 mg/L | carried out in a 7 L bubbling photobioreactor, temperature: 25 ± 2 °C, Photoperiod:14 h/d, pumped into the air with different concentrations of $CO_2$ | the removal rate of orthophosphate was 92.8% under 5% $CO_2$ (*v/v*) for 7 days | [51] | |
| Settlement of sewage | Mixed algae | / | Wastewater Treatment and Resource Recovery (STaRR) system | the phosphorus recovery content was 71.6% | [75] | |

## 4. Research Status Analysis of Phosphorus Removal from Municipal Wastewater by Microalgae

### *4.1. Symbiotic Systems of Bacteria and Algae*

The treatment of wastewater by pure microalgae is usually limited to laboratory conditions, while sterile water is difficult to find in sewage treatment plants. Microalgae often work with endophytic bacteria to purify the wastewater [76]. There are high concentrations of activated sludge in some urban sewage treatment plants. Urban sewage provides a culture substrate for microalgae, which can reduce the high costs associated with microalgal artificial culture medium. Cultures of microalgae can also be mixed with heterotrophic microorganisms in activated sludge to meet the sustainability requirements for urban sewage purification [77]. Bacteria and algae can form a good symbiotic relationship [78], enhancing the effects of the microalgae in the purification of urban sewage. For the treatment of municipal wastewater and industrial wastewater, there is a trend of combining selected algal and bacterial species [79,80]. For example, a culture made up of *C. vulgaris* and *P. putida* can remove organic matter and other nutrients and shows good performance in synthetic municipal wastewater [81,82]. Lananan et al. [83] co-cultured Chlorella and effective microorganisms (EM-1), and their findings revealed that this mix could extract 99.15% of the total phosphorus from domestic sewage. Qing et al. [69] screened Klebsiella from activated sludge and treated municipal wastewater with *C. pyrenoidosis*. Their findings revealed that the phosphorous microbe not only boosted the phosphorous absorption performance of the microalgae (up to 3.05 mg/L), helping to regulate the total phosphorous concentration in the water to 0.46 mg/L, but also increased the lipid yield and the average productivity of the microalgae (90.1% and 13.6%, respectively).

### *4.2. Adding Metal Compounds*

Magnesium ions ($Mg^{2+}$) are some of the most essential components for microalgae photosynthesis. P in wastewater can be removed by trimagnesium diphosphate ($Mg_3(PO_4)_2$) and $MgNH_4PO_4$ precipitation with other ions such as $NH_4^+$-N and $Mg^{2+}$. The assimilation of $PO_4^{3-}$-P could be hindered to some degree under $Mg^{2+}$ deficiency [54,84–87]. Studies have shown that the development of *C. vulgaris* is inhibited in media without $Mg^{2+}$, whereas microalgae grown in media with $Mg^{2+}$ are four times more productive than the blank group [88,89]. The concentration of $Mg^{2+}$ has a significant influence on the metabolism of microalgae in urban wastewater treatment [59]. Nickel laterite ore wastewater (NLOWW) provided by the hydrometallurgical recovery of the nickel contains high concentrations of $Mg^{2+}$ in the range of 20–40 g $L^{-1}$ [74]. Conventional NLOWW treatment for recovery of $Mg^{2+}$ consists of a series of integrated chemical–physical processes requiring investment in equipment and chemicals that are energy intensive and produce solid waste requiring further treatment [90]. Chen et al. [74] mixed urban and lateral nickel ore wastewater to cultivate *C. sorokiniana*. Their findings showed that the growth of microalgae cells in a culture without nickel laterite ore wastewater was slower and had a low biomass yield, whereas the microalgae biomass production rate in mixed wastewater containing nickel laterite ore increased by 1.89 times, the photosynthetic activity (Fv/Fm value) increased by 3.77 times, and the phosphorus removal rate increased by 39.3%; however, for 100% nickel laterite ore wastewater, excess $Mg^{2+}$ can contain high amounts of reactive oxygen species, which inhibit the growth of microalgae.

As an essential micronutrient for the growth of algae, iron ions also play an important role in the physiological synthesis and enzymatic reactions of algae. Iron can coordinate active oxygen in algal cells and take part in electron transport, enzyme reactions, photosynthesis and respiration, and the synthesis of proteins and nucleic acids, and can promote the metabolism and absorption of nutrients [91,92]. As mentioned earlier, phosphorus is an important nutrient for synthesizing cell proteins and nucleic acids. Qiu et al. [93] compared the effects of various forms of iron on the growth of *Anabaena flos-aquae*, and the results revealed that ferric ammonium citrate, EDTA-Fe, iron ions, and ferric oxalate are the forms of iron that can stimulate the development of microalgae. When the iron concentration was regulated in the range of 0.1 mg $L^{-1}$ to 0.8 mg $L^{-1}$, the impact of the iron type on microalgae growth was still greater than that of the iron concentration.

*4.3. Biofilm Technology*

In the 21st century, several wastewater treatment plants in the United States found that combining mobile bed bioreactor and fixed-membrane-activated sludge technologies not only enhanced the wastewater nitrification technology but also reduced the footprint of the facilities [70,94]. The integrated fixed-membrane-activated sludge process is an innovative biological wastewater treatment process that incorporates biofilm carriers into conventional activated sludge to eliminate nitrifiers, resulting in an improved retention time for the heterotrophic bacteria [70]. The nitrifying bacteria can be applied to the biofilm without being affected by the washing of the nitrifying agent, while the biological nitrogen can be eliminated by the nitrifying reaction [70]. Compared to the moving bed bioreactors (MBBR) system, the Integrated Fixed-Film Activated Sludge (IFAS) system can decouple the SRT of nitrifiers and polyphosphate bioaccumulators (PAO) by maximizing the elimination performance of biological nitrogen and phosphorus [70].

Jared Church et al. [70] incorporated microalgae into an optimized fixed-membrane-activated sludge configuration for photooxygenation and examined the symbiotic reactions of microalgae and bacteria to suspended matter and IFAS biofilms. In sequential batch mode, the microalgae were combined with the IFAS method to remove 51% of the phosphorus without mechanical aeration. This study also showed that the addition of microalgae to the IFAS system modified the metabolic function of multiple bacterial populations. This study was not only desirable for the reduction of phosphorus in water sources, but it also offers new research ideas for the improvement of various water bodies, the use of microalgae–IFAS technologies to modify the behavior of bacterial species, and the evolution of water quality.

Abeysiriwardana-Arachchige et al. [75] suggested a research approach for the treatment and recovery of wastewater based on algae (STaRR). Their findings revealed that the STARR device had a recovery output of 71.6% of nutrient phosphorus and that the removal of phosphorus per unit of energy consumption was calculated to be 0.1 g/kJ. This indicates that the STaRR system could be a green alternative for water treatment and nutrient recovery.

Anaerobic membrane bioreactor technology has the benefits of absorbing less energy and producing less sludge relative to more conventional aerobic systems, while still producing biomethane.; however, inorganic contaminants such as nitrogen and phosphorus cannot be removed from anaerobic reactors. Other nitrogen-containing compounds convert ammonia, which increases the concentration of ammonia in the water, one of the major microalgae nutrients [53,95,96]. Microalgae, thus, play an important role in the production of this system. Microalgae demonstrate good growth conditions in 5–30% nitrified digestion solutions combined with municipal wastewater. For example, the addition of a 10% nitrification solution for digestion in a two-stage bacterial-microalgal phase can eliminate 77% of the phosphate. Under the same conditions, the continuous use of a microalgae-based photobioreactor (MPBR) membrane will extract more than 99% of the phosphate [72].

*4.4. Recovery Technology*

From an economic and sustainable development point of view, the recovery of microalgae is a significant link in the treatment of wastewater by microalgae [97,98]. Currently, more mainstream approaches for the recovery of microalgae include centrifugation, filtration, sedimentation by gravity, and flocculation [98,99]. In addition, flocculant recovery technology and immobilized recovery technology are used.

4.4.1. Flocculant Recovery Technology

Microalgae can also be flocculated and retrieved by inorganic coagulants, such as aluminum sulfate or ferric chloride, polymeric flocculants, mixtures of the two components, or by using automated chemical flocculation methods such as pH modification [100–102]. Mennaa et al. [73] researched the continuous activity of the BCPBR (bubble column photobioreactor) using a flocculation–precipitation system to examine the effects of phosphorous removal and recovery of natural microalgae plants in urban wastewater. Their findings showed that continuous-mode experiments extract up to 99% of the total dissolved phosphorus without controlling the volume of $CO_2$ or regulating pH. PAC, $Fe_2(SO_4)_3$, and $Al_2(SO_4)_3$ were found to have positive effects on the recovery of microalgae; however, the addition of a flocculant not only increases the cost, but also may cause other pollution to the water body, reducing the sustainability of urban sewage.

4.4.2. Immobilized Recovery Technology

Compared with flocculant recovery technology, immobilized recovery technology can avoid secondary water pollution and increase the possibility of the sustainable development of urban sewage. The elimination of sewage from suspended algae systems can result in a low concentration of algae in the reactors, resulting in a reduction in the treatment rate [33,103,104]. The use of alginate beads to immobilize microalgae cells helps to retain a high concentration of microalgae in the reactor, which can easily remove nutrients from the water body, while the hydraulic retention period is less than 12 h [80,105]. Immobilized microalgae beads can settle rapidly to promote screening and regeneration, and these beads can be used directly as fertilizer or biomethane production after digestion [106–108]. In addition, beads produced by immobilized microalgae can also shield the culture from harmful contaminants in wastewater [109]. Kube [110] demonstrated that different concentrations of nitrogen and phosphorus can influence the absorption of phosphorus by Chlorella and that the immobilization of microalgae does not hinder the rate or ratio of nitrogen and phosphorous absorption. In addition, cell immobilization and co-cultivation of *Bacillus vulgaris* and *P. brasiliensis* can boost the removal rates of ammonia and phosphorus [111].

Katam et al. [71] used mixed microalgae to investigate the removal efficiency of carbon and nutrients in the treatment of real wastewater in an activated sludge reactor and set up two separate treatment systems for the simultaneous treatment of domestic wastewater. Their findings revealed that the total phosphorus removal performance of the immobilized microalgae system was as high as 93%, which was higher than the suspended activated sludge system. In addition, the microalgae developed higher lipid and carbon contents than the suspended activated sludge solution in the immobilized microalgae system.

*4.5. Other Improved Technologies*

Photobioreactors used to grow microalgae also have a major effect on the treatment of urban wastewater by microalgae. For example, relative to other reactors, BPBR has the advantages of high heat and mass transfer speeds, compact construction, and low operational and maintenance costs. Since the reactor has a higher surface-to-volume ratio, good mixability, lower shear stress, high scalability potential, simple sterilization, low emissions, and decreased photoinhibition, it can better monitor the growth parameters (such as temperature) of photooxidation [112].

The microalgal elimination of phosphorus can also be improved via genome building. Guerra-Renteria et al. [113] developed a genome-scale biochemical reaction network for the co-cultivation of *Chlorella* spp. and *Pseudomonas aeruginosa* bacteria using a metabolic pathway analysis (MPA). This analysis considers the metabolic ability of co-cultivation and determines the best conditions for the removal of nutrients. The theoretical phosphorous removal yield under photoheterotrophic conditions was determined as follows: 0.042 mmol of $PO_4^{3-}$ per g DW of *C. vulgaris*, 19.43 mmol of phosphorus (Pi) per g DW of *C. vulgaris*, and 4.90 mmol of phosphorus (Pi) per g DW of *P. aeruginosa.* These theoretical yields are important because they can help in the design of biological systems and in the understanding of the theoretical requirements of oxygen and carbon dioxide in order to achieve maximum nutrient absorption. In this system, other by-products containing nitrogen or phosphorus may not even be formed, and all nutrient absorption is directed toward the growth of microalgae and bacteria [113].

## 5. Conclusions and Perspectives

While microalgae-based technologies provide a sustainable alternative for the removal of phosphorus from urban wastewater, the substitution of conventional water treatment technologies remains a major challenge. In this paper, we present the microalgae culture methods and the microalgae dephosphorization process. In addition, considerations influencing the elimination of microalgae phosphorus in urban water include traditional factors, but also the species characteristics of the microalgae and urban sewage types. This study, however, cannot completely summarize all of the factors influencing the dephosphorization of microalgae. It is necessary to further study the mechanisms and factors impacting microalgae dephosphorization from a microscopic perspective.

The study of phosphorous removal by microalgae showed that microalgae were often combined with other municipal wastewater treatment systems. Symbiotic relationships between bacteria and microalgae are common in municipal wastewater treatment plants. Biofilm has commonly been used in the treatment of sewage in the 21st century, and its combination with microalgae has encouraged the elimination of phosphorus from wastewater. While the microalgae biomass may be improved and phosphorous removal efficiency may be increased by increasing the concentrations of magnesium and iron ions in water, the mechanism for the removal of phosphorus by the inclusion of certain metals is less studied. In addition, certain metal nanomaterials have good adsorption and other characteristics, although the study of metal nanomaterials on microalgae is still lacking. This study also shows that immobilized microalgae technology can not only solve the problems of microalgae recycling and urban sewage sustainability, but can also improve the efficiency of phosphorus removal. This immobilization technology also offers a research concept to solve secondary contamination caused by the addition of metal ions to support the growth of microalgae.

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## References

1. Alkhudhiri, A.; Bin Darwish, N.; Hilal, N. Analytical and forecasting study for wastewater treatment and water resources in Saudi Arabia. *J. Water Process. Eng.* **2019**, *32*, 100915. [CrossRef]
2. Deng, Y.; Wheatley, A. Wastewater Treatment in Chinese Rural Areas. *Asian J. Water, Environ. Pollut.* **2016**, *13*, 1–11. [CrossRef]
3. Ingallinella, A.M.; Sanguinetti, G.; Koottatep, T.; Montangero, A.; Strauss, M. The challenge of faecal sludge management in urban areas—Strategies, regulations and treatment options. *Water Sci. Technol.* **2002**, *46*, 285–294. [CrossRef]
4. Wurz, A.; Kuchta, K.; Onay, T.T. Review on municipal sewage sludge management in Turkey and Europe. *Int. J. Glob. Warm.* **2011**, *3*, 116. [CrossRef]
5. Kumar, P. Numerical quantification of current status quo and future prediction of water quality in eight Asian megacities: Challenges and opportunities for sustainable water management. *Environ. Monit. Assess.* **2019**, *191*, 319. [CrossRef]
6. Azam, H.M.; Alam, S.T.; Hasan, M.; Yameogo, D.D.S.; Kannan, A.D.; Rahman, A.; Kwon, M.J. Phosphorous in the environment: Characteristics with distribution and effects, removal mechanisms, treatment technologies, and factors affecting recovery as minerals in natural and engineered systems. *Environ. Sci. Pollut. Res.* **2019**, *26*, 20183–20207. [CrossRef] [PubMed]
7. Heinzmann, B.; Betriebe, B.W. Phosphorus recovery in wastewater treatment plants. In Proceedings of the Second International Conference, Moscow, Russia, 29 November–3 December 1999.

8. Fattah, K. Finding Nutrient-Related Problems in Wastewater Treatment Plants. In Proceedings of the 2nd International Conference on Environmental, Bio-Medical and Biotechnology, Dubai, United Arab Emirates, 4–5 August 2012.
9. Jenkins, D.; Ferguson, J.F.; Menar, A.B. Chemical processes for phosphate removal. *Water Res.* **1971**, *5*, 369–389. [CrossRef]
10. Stensel, H.D. *Phosphorous and Nitrogen Removal from Municipal Wastewater: Principles and Practice*; Routledge: Boca Raton, FL, USA, 1991.
11. Zhang, M.; Lawlor, P.G.; Hu, Z.; Zhan, X. Nutrient removal from separated pig manure digestate liquid using hybrid biofilters. *Environ. Technol.* **2013**, *34*, 645–651. [CrossRef] [PubMed]
12. Grégorio, C.; Eric, L. Advantages and disadvantages of techniques used for wastewater treatment. *Environ. Chem. Lett.* **2018**, *17*, 145–155.
13. Bunce, J.T.; Ndam, E.; Ofiteru, I.D.; Moore, A.; Graham, D.W. A Review of Phosphorus Removal Technologies and Their Applicability to Small-Scale Domestic Wastewater Treatment Systems. *Front. Environ. Sci.* **2018**, *6*, 8. [CrossRef]
14. Cardew, P. Measuring the benefit of orthophosphate treatment on lead in drinking water. *Water Health* **2009**, *7*, 123–131. [CrossRef]
15. Volk, C.; Dundore, E.; Schiermann, J.; Lechevallier, M. Practical evaluation of iron corrosion control in a drinking water distribution system. *Water Res.* **2000**, *34*, 1967–1974. [CrossRef]
16. House, J.E.; House, K.A. *Descriptive Inorganic Chemistry*; Academic Press: Cambridge, MA, USA, 2001.
17. Wind, T. *The Role of Detergents in the Phosphate-Balance of European Surface Waters*; European Water Management Online: Hennef, Germany, 2007; pp. 1–19.
18. Shu, L.; Schneider, P.; Jegatheesan, V. An economic evaluation of phosphorus recovery as struvite from digester supernatant. *Bioresour. Technol.* **2006**, *97*, 2211–2216. [CrossRef]
19. Ehama, M.; Hashihama, F.; Kinouchi, S.; Kanda, J.; Saito, H. Sensitive determination of total particulate phosphorus and particulate inorganic phosphorus in seawater using liquid waveguide spectrophotometry. *Talanta* **2016**, *153*, 66–70. [CrossRef]
20. Panasiuk, O. Phosphorus Removal and Recovery from Wastewater Using Magnetite. Master's Thesis, Industrial Ecology, Royal Institute of Technology, Stockholm, Sweden, 2010.
21. Morse, G.; Brett, S.; Guy, J.; Lester, J. Review: Phosphorus removal and recovery technologies. *Sci. Total. Environ.* **1998**, *212*, 69–81. [CrossRef]
22. Mohsenpour, S.F.; Hennige, S.; Willoughby, N.; Adeloye, A.; Gutierrez, T. Integrating micro-algae into wastewater treatment: A review. *Sci. Total. Environ.* **2021**, *752*, 142168. [CrossRef] [PubMed]
23. Lutzu, G.A.; Ciurli, A.; Chiellini, C.; di Caprio, F.; Concas, A.; Dunford, N.T. Latest developments in wastewater treatment and biopolymer production by microalgae. *J. Environ. Chem. Eng.* **2021**, *9*, 104926. [CrossRef]
24. Borowitzka, M.A.; Moheimani, N.R. *Open Pond Culture Systems*; Springer Science and Business Media: Cham, Switzerland, 2013; pp. 133–152.
25. Acién, F.F.G.; Fernández, S.J.M.; Molina, G.E. Photobioreactors for the production of microalgae. *Rev. Environ. Sci. Biotechnol.* **2013**, *12*, 131–151. [CrossRef]
26. Lee, Y.-K.; Shen, H. Basic Culturing Techniques. *Handb. Microalgal Cult.* **2007**, 40–56. [CrossRef]
27. Donald, K.M.; Scanlan, D.J.; Carr, N.G.; Mann, N.H.; Joint, I. Comparative phosphorus nutrition of the marine cyanobacterium *Synechococcus* WH7803 and the marine diatom *Thalassiosira weissflogii*. *J. Plankton Res.* **1997**, *19*, 1793–1813. [CrossRef]
28. Solovchenko, A.; Khozin-Goldberg, I.; Selyakh, I.; Semenova, L.; Ismagulova, T.; Lukyanov, A.; Mamedov, I.; Vinogradova, E.; Karpova, O.; Konyukhov, I.; et al. Phosphorus starvation and luxury uptake in green microalgae revisited. *Algal Res.* **2019**, *43*, 101651. [CrossRef]
29. Cembella, A.D.; Antia, N.J.; Harrison, P.J. The Utilization of Inorganic and Organic Phosphorous Compounds as Nutrients by Eukaryotic Microalgae: A Multidisciplinary Perspective: Part I. *CRC Crit. Rev. Microbiol.* **1982**, *10*, 317–391. [CrossRef] [PubMed]
30. Cembella, A.D.; Antia, N.J.; Harrison, P.J.; Rhee, G.Y. The Utilization of Inorganic and Organic Phosphorous Compounds as Nutrients by Eukaryotic Microalgae: A Multidisciplinary Perspective: Part II. *CRC Crit. Rev. Microbiol.* **1984**, *11*, 13–81. [CrossRef] [PubMed]
31. Su, Y. Revisiting carbon, nitrogen, and phosphorus metabolisms in microalgae for wastewater treatment. *Sci. Total. Environ.* **2021**, *762*, 144590. [CrossRef]
32. Diaz, J.M.; Björkman, K.M.; Haley, S.T.; Ingall, E.; Karl, D.; Longo, A.F.; Dyhrman, S.T. Polyphosphate dynamics at Station ALOHA, North Pacific subtropical gyre. *Limnol. Oceanogr.* **2015**, *61*, 227–239. [CrossRef]
33. Whitton, R.; Ometto, F.; Pidou, M.; Jarvis, P.; Villa, R.; Jefferson, B. Microalgae for municipal wastewater nutrient remediation: Mechanisms, reactors and outlook for tertiary treatment. *Environ. Technol. Rev.* **2015**, *4*, 133–148. [CrossRef]
34. Sforza, E.; Calvaruso, C.; la Rocca, N.; Bertucco, A. Luxury uptake of phosphorus in *Nannochloropsis salina*: Effect of P concentration and light on P uptake in batch and continuous cultures. *Biochem. Eng. J.* **2018**, *134*, 69–79. [CrossRef]
35. Li, Q.; Fu, L.; Wang, Y.; Zhou, D.; Rittmann, B.E. Excessive phosphorus caused inhibition and cell damage during heterotrophic growth of *Chlorella regularis*. *Bioresour. Technol.* **2018**, *268*, 266–270. [CrossRef]
36. Dyhrman, S.T. Nutrients and their acquisition: Phosphorus physiology in microalgae. In *The Physiology of Microalgae*; Borowitzka, M., Beardall, J., Raven, J., Eds.; Springer: Dordrecht, The Netherlands, 2016; pp. 155–183. [CrossRef]
37. Montgomery, J.M. *Water Treatment Principles and Design*; Wiley: New York, NY, USA, 1985.
38. Rittmann, B.E.; Mayer, B.; Westerhoff, P.; Edwards, M. Capturing the lost phosphorus. *Chemosphere* **2011**, *84*, 846–853. [CrossRef]

39. Wang, C.; Jiang, H.L. Chemicals used for in situ immobilization to reduce the internal phosphorus loading from lake sediments for eutrophication control. *Crit. Rev. Environ. Sci. Technol.* **2016**, *46*, 947–997. [CrossRef]
40. Larsdotter, K. WasteWater treatment with microalgae—A literature review. *Vatten* **2006**, *62*, 31–38.
41. Solmaz, A.; Işık, M. Optimization of membrane photobioreactor; the effect of hydraulic retention time on biomass production and nutrient removal by mixed microalgae culture. *Biomass Bioenergy* **2020**, *142*, 105809. [CrossRef]
42. Arcila, J.S.; Buitrón, G. Microalgae-bacteria aggregates: Effect of the hydraulic retention time on the municipal wastewater treatment, biomass settleability and methane potential. *J. Chem. Technol. Biotechnol.* **2016**, *91*, 2862–2870. [CrossRef]
43. Metcalf, E. *Wastewater Engineering and Reuse, 4th ed*; Mc. GrawHill: New York, NY, USA, 2003.
44. García, D.; Alcántara, C.; Blanco, S.; Pérez, R.; Bolado, S.; Muñoz, R. Enhanced carbon, nitrogen and phosphorus removal from domestic wastewater in a novel anoxic-aerobic photobioreactor coupled with biogas upgrading. *Chem. Eng. J.* **2017**, *313*, 424–434. [CrossRef]
45. Toledo-Cervantes, A.; Posadas, E.; Bertol, I.; Turiel, S.; Alcoceba, A.; Muñoz, R. Assessing the influence of the hydraulic retention time and carbon/nitrogen ratio on urban wastewater treatment in a new anoxic-aerobic algal-bacterial photobioreactor configuration. *Algal Res.* **2019**, *44*, 101672. [CrossRef]
46. Alcántara, C.; Domínguez, J.M.; García, D.; Blanco, S.; Pérez, R.; Garcia-Encina, P.A.; Muñoz, R. Evaluation of wastewater treatment in a novel anoxic–aerobic algal–bacterial photobioreactor with biomass recycling through carbon and nitrogen mass balances. *Bioresour. Technol.* **2015**, *191*, 173–186. [CrossRef] [PubMed]
47. Wang, L.; Min, M.; Li, Y.; Chen, P.; Chen, Y.; Liu, Y.; Ruan, R. Cultivation of green *Algae Chlorella* sp. in different wastewaters from municipal wastewater treatment plant. *Appl. Biochem. Biotechnol.* **2010**, *162*, 1174–1186. [CrossRef]
48. Choi, H.J.; Lee, S.M. Effect of the N/P ratio on biomass productivity and nutrient removal from municipal wastewater. *Bioprocess Biosyst. Eng.* **2015**, *38*, 761–766. [CrossRef]
49. Molazadeh, M.; Danesh, S.; Ahmadzadeh, H.; Pourianfar, H.R. Influence of $CO_2$ concentration and N:P ratio on Chlorella vulgaris-assisted nutrient bioremediation, $CO_2$ biofixation and biomass production in a lagoon treatment plant. *J. Taiwan Inst. Chem. Eng.* **2019**, *96*, 114–120. [CrossRef]
50. Ma, S.; Yu, Y.; Cui, H.; Yadav, R.S.; Li, J.; Feng, Y. Unsterilized sewage treatment and carbohydrate accumulation in Tetradesmus obliquus PF3 with $CO_2$ supplementation. *Algal Res.* **2020**, *45*, 101741. [CrossRef]
51. Chaudhary, R.; Tong, Y.W.; Dikshit, A.K. Kinetic study of nutrients removal from municipal wastewater by Chlorella vulgaris in photobioreactor supplied with $CO_2$-enriched air. *Environ. Technol.* **2018**, *41*, 617–626. [CrossRef]
52. Jiang, L.; Luo, S.; Fan, X.; Yang, Z.; Guo, R. Biomass and lipid production of marine microalgae using municipal wastewater and high concentration of $CO_2$. *Appl. Energy* **2011**, *88*, 3336–3341. [CrossRef]
53. Pachés, M.; Martínez-Guijarro, R.; González-Camejo, J.; Seco, A.; Barat, R. Selecting the most suitable microalgae species to treat the effluent from an anaerobic membrane bioreactor. *Environ. Technol.* **2018**, *41*, 267–276. [CrossRef]
54. Devi, M.P.; Subhash, G.V.; Mohan, S.V. Heterotrophic cultivation of mixed microalgae for lipid accumulation and wastewater treatment during sequential growth and starvation phases: Effect of nutrient supplementation. *Renew. Energy* **2012**, *43*, 276–283. [CrossRef]
55. Bohutskyi, P.; Kligerman, D.C.; Byers, N.; Nasr, L.K.; Cua, C.; Chow, S.; Su, C.; Tang, Y.; Betenbaugh, M.J.; Bouwer, E.J. Effects of inoculum size, light intensity, and dose of anaerobic digestion centrate on growth and productivity of Chlorella and Scenedesmus microalgae and their poly-culture in primary and secondary wastewater. *Algal Res.* **2016**, *19*, 278–290. [CrossRef]
56. Bellucci, M.; Marazzi, F.; Naddeo, L.S.; Piergiacomo, F.; Beneduce, L.; Ficara, E.; Mezzanotte, V. Disinfection and nutrient removal in laboratory-scale photobioreactors for wastewater tertiary treatment. *J. Chem. Technol. Biotechnol.* **2019**. [CrossRef]
57. Abdullahi, Y.; Akunna, J.; White, N.; Hallett, P.; Wheatley, R. Investigating the effects of anaerobic and aerobic post-treatment on quality and stability of organic fraction of municipal solid waste as soil amendment. *Bioresour. Technol.* **2008**, *99*, 8631–8636. [CrossRef] [PubMed]
58. Bjornsson, W.J.; Nicol, R.W.; Dickinson, K.E.; McGinn, P.J. Anaerobic digestates are useful nutrient sources for microalgae cultivation: Functional coupling of energy and biomass production. *J. Appl. Phycol.* **2013**, *25*, 1523–1528. [CrossRef]
59. Salama, E.S.; Kurade, M.B.; Abou-Shanab, R.; El-Dalatony, M.M.; Yang, I.S.; Min, B.; Jeon, B.H. Recent progress in microalgal biomass production coupled with wastewater treatment for biofuel generation. *Renew. Sust. Energy Rev.* **2017**, *79*, 1189–1211. [CrossRef]
60. Ermis, H.; Altınbaş, M. Determination of biokinetic coefficients for nutrient removal from anaerobic liquid digestate by mixed microalgae. *J. Appl. Phycol.* **2018**, *31*, 1773–1781. [CrossRef]
61. Mujtaba, G.; Lee, K. Treatment of real wastewater using co-culture of immobilized *Chlorella vulgaris* and suspended activated sludge. *Water Res.* **2017**, *120*, 174–184. [CrossRef]
62. Vandamme, D.; Foubert, I.; Fraeye, I.; Muylaert, K. Influence of organic matter generated by *Chlorella vulgaris* on five different modes of flocculation. *Bioresour. Technol.* **2012**, *124*, 508–511. [CrossRef] [PubMed]
63. Yan, S.-T.; Chu, L.-B.; Xing, X.-H.; Yu, A.-F.; Sun, X.-L.; Jurcik, B. Analysis of the mechanism of sludge ozonation by a combination of biological and chemical approaches. *Water Res.* **2009**, *43*, 195–203. [CrossRef] [PubMed]
64. Lei, Y.J.; Tian, Y.; Zhang, J.; Sun, L.; Kong, X.W.; Zuo, W.; Kong, L.C. Microalgae cultivation and nutrients removal from sewage sludge after ozonizing in algal-bacteria system. *Ecotoxicol. Environ. Saf.* **2018**, *165*, 107–114. [CrossRef]

65. Chu, L.; Yan, S.; Xing, X.-H.; Sun, X.; Jurcik, B. Progress and perspectives of sludge ozonation as a powerful pretreatment method for minimization of excess sludge production. *Water Res.* **2009**, *43*, 1811–1822. [CrossRef] [PubMed]
66. Yan, S.-T.; Zheng, H.; Li, A.; Zhang, X.; Xing, X.-H.; Chu, L.-B.; Ding, G.; Sun, X.-L.; Jurcik, B. Systematic analysis of biochemical performance and the microbial community of an activated sludge process using ozone-treated sludge for sludge reduction. *Bioresour. Technol.* **2009**, *100*, 5002–5009. [CrossRef] [PubMed]
67. Wang, J.-H.; Zhang, T.-Y.; Dao, G.-H.; Xu, X.-Q.; Wang, X.-X.; Hu, H.-Y. Microalgae-based advanced municipal wastewater treatment for reuse in water bodies. *Appl. Microbiol. Biotechnol.* **2017**, *101*, 2659–2675. [CrossRef]
68. Semblante, G.U.; Hai, F.I.; Dionysiou, D.; Fukushi, K.; Price, W.E.; Nghiem, L. Holistic sludge management through ozonation: A critical review. *J. Environ. Manag.* **2017**, *185*, 79–95. [CrossRef]
69. Wang, Q.; Jin, W.; Zhou, X.; Guo, S.; Gao, S.-H.; Chen, C.; Tu, R.; Han, S.-F.; Jiang, J.; Feng, X. Growth enhancement of biodiesel-promising microalga *Chlorella pyrenoidosa* in municipal wastewater by polyphosphate-accumulating organisms. *J. Clean. Prod.* **2019**, *240*, 118148. [CrossRef]
70. Church, J.; Ryu, H.; Sadmani, A.H.A.; Randall, A.A.; Domingo, J.S.; Lee, W.H. Multiscale investigation of a symbiotic microalgal-integrated fixed film activated sludge (MAIFAS) process for nutrient removal and photo-oxygenation. *Bioresour. Technol.* **2018**, *268*, 128–138. [CrossRef]
71. Katam, K.; Bhattacharyya, D. Simultaneous treatment of domestic wastewater and bio-lipid synthesis using immobilized and suspended cultures of microalgae and activated sludge. *J. Ind. Eng. Chem.* **2019**, *69*, 295–303. [CrossRef]
72. Praveen, P.; Guo, Y.; Kang, H.; Lefebvre, C.; Loh, K.-C. Enhancing microalgae cultivation in anaerobic digestate through nitrification. *Chem. Eng. J.* **2018**, *354*, 905–912. [CrossRef]
73. Mennaa, F.Z.; Arbib, Z.; Perales, J.A. Urban wastewater photobiotreatment with microalgae in a continuously operated photobioreactor: Growth, nutrient removal kinetics and biomass coagulation–flocculation. *Environ. Technol.* **2019**, *40*, 342–355. [CrossRef] [PubMed]
74. Chen, Z.; Qiu, S.; Amadu, A.A.; Shen, Y.; Wang, L.; Wu, Z.; Ge, S. Simultaneous improvements on nutrient and Mg recoveries of microalgal bioremediation for municipal wastewater and nickel laterite ore wastewater. *Bioresour. Technol.* **2020**, *297*, 122517. [CrossRef] [PubMed]
75. Arachchige, I.S.A.A.; Munasinghe-Arachchige, S.; Delanka-Pedige, H.M.K.; Nirmalakhandan, N. Removal and recovery of nutrients from municipal sewage: Algal vs. conventional approaches. *Water Res.* **2020**, *175*, 115709. [CrossRef] [PubMed]
76. Kouzuma, A.; Watanabe, K. Exploring the potential of algae/bacteria interactions. *Curr. Opin. Biotechnol.* **2015**, *33*, 125–129. [CrossRef] [PubMed]
77. Ferreira, A.; Ribeiro, B.; Marques, P.A.; Ferreira, A.F.; Dias, A.P.; Pinheiro, H.M.; Reis, A.; Gouveia, L. *Scenedesmus obliquus* mediated brewery wastewater remediation and CO2 bio-fixation for green energy purposes. *J. Clean. Prod.* **2017**, *165*, 1316–1327. [CrossRef]
78. He, P.; Mao, B.; Lü, F.; Shao, L.; Lee, D.; Chang, J. The combined effect of bacteria and *Chlorella vulgaris* on the treatment of municipal wastewaters. *Bioresour. Technol.* **2013**, *146*, 562–568. [CrossRef]
79. Alsan, S.; Kapdan, I.K. Batch kinetics of nitrogen and phosphorus removal from synthetic wastewater by algae. *Ecol. Eng.* **2006**, *28*, 64–70.
80. Pires, J.C.M.; Alvim-Ferraz, M.D.C.; Martins, F.; Simões, M. Wastewater treatment to enhance the economic viability of microalgae culture. *Environ. Sci. Pollut. Res.* **2013**, *20*, 5096–5105. [CrossRef] [PubMed]
81. Mujtaba, G.; Rizwan, M.; Lee, K. Simultaneous removal of inorganic nutrients and organic carbon by symbiotic co-culture of Chlorella vulgaris and Pseudomonas putida. *Biotechnol. Bioprocess Eng.* **2015**, *20*, 1114–1122. [CrossRef]
82. Gómez-Guzmán, A.; Jiménez-Magaña, S.; Guerra-Rentería, A.S.; Gómez-Hermosillo, C.; Parra-Rodríguez, F.J.; Velázquez, S.; Aguilar-Uscanga, B.R.; Solis-Pacheco, J.; González-Reynoso, O. Evaluation of nutrients removal (NO3-N, NH3-N and PO4-P) with *Chlorella vulgaris*, *Pseudomonas putida*, *Bacillus cereus* and a consortium of these microorganisms in the treatment of wastewater effluents. *Water Sci. Technol.* **2017**, *76*, 49–56. [CrossRef]
83. Lananan, F.; Hamid, S.H.A.; Din, W.N.S.; Ali, N.; Khatoon, H.; Jusoh, A.; Endut, A. Symbiotic bioremediation of aquaculture wastewater in reducing ammonia and phosphorus utilizing Effective Microorganism (EM-1) and microalgae (*Chlorella* sp.). *Int. Biodeterior. Biodegradation* **2014**, *95*, 127–134. [CrossRef]
84. Andersson, I.; Backlund, A. Structure and function of Rubisco. *Plant Physiol. Biochem.* **2008**, *46*, 275–291. [CrossRef] [PubMed]
85. Sasaki, Y.; Nagano, Y. Plant Acetyl-CoA Carboxylase: Structure, Biosynthesis, Regulation, and Gene Manipulation for Plant Breeding. *Biosci. Biotechnol. Biochem.* **2004**, *68*, 1175–1184. [CrossRef]
86. Pasternak, K.; Kocot, J.; Horecka, A. Biochemistry of magnesium. *J. Elementol.* **2010**, *15*, 601–616. [CrossRef]
87. Sydney, E.; Sturm, W.; de Carvalho, J.; Soccol, V.T.; Larroche, C.; Pandey, A.; Soccol, C.R. Potential carbon dioxide fixation by industrially important microalgae. *Bioresour. Technol.* **2010**, *101*, 5892–5896. [CrossRef] [PubMed]
88. Ayed, H.B.A.-B.; Taidi, B.; Ayadi, H.; Pareau, D.; Stambouli, M. Effect of magnesium ion concentration in autotrophic cultures of *Chlorella vulgaris*. *Algal Res.* **2015**, *9*, 291–296. [CrossRef]
89. Ren, H.; Liu, B.; Kong, F.; Zhao, L.; Xie, G.; Ren, N. Enhanced lipid accumulation of green microalga *Scenedesmus* sp. by metal ions and EDTA addition. *Bioresour. Technol.* **2014**, *169*, 763–767. [CrossRef]
90. Mu, W.N.; Shi, S.Z.; Zhai, Y.C. Magnesium Recovery from Desiliconization Slag of Nickel Laterite Ores by Carbonization. *Adv. Mater. Res.* **2013**, *813*, 255–258. [CrossRef]

91. Alexova, R.; Fujii, M.; Birch, D.; Cheng, J.; Waite, T.D.; Ferrari, B.C.; Neilan, B.A. Iron uptake and toxin synthesis in the bloom-forming Microcystis aeruginosa under iron limitation. *Environ. Microbiol.* **2011**, *13*, 1064–1077. [CrossRef]
92. Boyd, P.; Ellwood, M. The biogeochemical cycle of iron in the ocean. *Nat. Geosci.* **2010**, *3*, 675–682. [CrossRef]
93. Qiu, Y.; Wang, Z.; Liu, F.; Liu, J.; Zhou, T. Effect of different kinds of complex iron on the growth of *Anabaena flosaquae*. *Environ. Technol.* **2018**, *40*, 2889–2896. [CrossRef]
94. Onnis-Hayden, A.; Majed, N.; Schramm, A.; Gu, A.Z. Process optimization by decoupled control of key microbial populations: Distribution of activity and abundance of polyphosphate-accumulating organisms and nitrifying populations in a full-scale IFAS-EBPR plant. *Water Res.* **2011**, *45*, 3845–3854. [CrossRef] [PubMed]
95. Ho, J.; Sung, S. Methanogenic activities in anaerobic membrane bioreactors (AnMBR) treating synthetic municipal wastewater. *Bioresource Technol.* **2010**, *101*, 2191–2196. [CrossRef] [PubMed]
96. Podevin, M.; de Francisci, D.; Holdt, S.L.; Angelidaki, I. Effect of nitrogen source and acclimatization on specific growth rates of microalgae determined by a high-throughput in vivo microplate autofluorescence method. *Environ. Boil. Fishes* **2015**, *27*, 1415–1423. [CrossRef]
97. Sukenik, A.; Bilanovic, D.; Shelef, G. Flocculation of microalgae in brackish and sea waters. *Biomass Bioenergy* **1988**, *15*, 187–199. [CrossRef]
98. Grima, E.M.; Belarbi, E.-H.; Fernández, F.G.A.; Medina, A.R.; Chisti, Y. Recovery of microalgal biomass and metabolites: Process options and economics. *Biotechnol. Adv.* **2003**, *20*, 491–515. [CrossRef]
99. Lee, S.; Kim, S.B.; Kim, J.E.; Kwon, G.S.; Yoon, B.D.; Oh, H.M. Effects of harvesting method and growth stage on the flocculation of the green alga *Botryococcus braunii*. *Lett. Appl. Microbiol.* **1998**, *27*, 14–18. [CrossRef]
100. Siew Moi, P. Handbook of microalgal culture. Biotechnology and applied phycology. *J. Appl. Phycology.* **2004**, *16*, 159–160.
101. Shelef, G.; Sukenik, A.; Green, M. Microalgae Harvesting and Processing: A Literature Review. Available online: https://www.osti.gov/biblio/6204677 (accessed on 1 August 2021).
102. Chen, C.-Y.; Yeh, K.-L.; Aisyah, R.; Lee, D.-J.; Chang, J.-S. Cultivation, photobioreactor design and harvesting of microalgae for biodiesel production: A critical review. *Bioresour. Technol.* **2011**, *102*, 71–81. [CrossRef]
103. Park, J.; Craggs, R.; Shilton, A. Wastewater treatment high rate algal ponds for biofuel production. *Bioresour. Technol.* **2011**, *102*, 35–42. [CrossRef] [PubMed]
104. Filippino, K.C.; Mulholland, M.R.; Bott, C.B. Phycoremediation strategies for rapid tertiary nutrient removal in a waste stream. *Algal Res.* **2015**, *11*, 125–133. [CrossRef]
105. Whitton, R.; Santinelli, M.; Pidou, M.; Ometto, F.; Henderson, R.; Roddick, F.; Jarvis, P.; Villa, R.; Jefferson, B. Tertiary nutrient removal from wastewater by immobilised microalgae: Impact of wastewater nutrient characteristics and hydraulic retention time (HRT). *H2Open J.* **2018**, *1*, 12–25. [CrossRef]
106. De-Bashan, L.E.; Bashan, Y. Immobilized microalgae for removing pollutants: Review of practical aspects. *Bioresour. Technol.* **2010**, *101*, 1611–1627. [CrossRef]
107. Lam, M.K.; Lee, K.T. Immobilization as a feasible method to simplify the separation of microalgae from water for biodiesel production. *Chem. Eng. J.* **2012**, *191*, 263–268. [CrossRef]
108. Yadavalli, R.; Heggers, G.R.V.N. Two stage treatment of dairy effluent using immobilized *Chlorella pyrenoidosa*. *J. Environ. Health Sci. Eng.* **2013**, *11*, 36. [CrossRef]
109. Covarrubias, S.A.; De-Bashan, L.E.; Moreno, M.; Bashan, Y. Alginate beads provide a beneficial physical barrier against native microorganisms in wastewater treated with immobilized bacteria and microalgae. *Appl. Microbiol. Biotechnol.* **2012**, *93*, 2669–2680. [CrossRef]
110. Kube, M.; Spedding, B.; Gao, L.; Fan, L.; Roddick, F. Nutrient removal by alginate-immobilized *Chlorella vulgaris*: Response to different wastewater matrices. *J. Chem. Technol. Biotechnol.* **2020**, *95*, 1790–1799. [CrossRef]
111. Choix, F.J.; De-Bashan, L.E.; Bashan, Y. Enhanced accumulation of starch and total carbohydrates in alginate-immobilized *Chlorella* spp. induced by *Azospirillum brasilense*: I. Autotrophic conditions. *Enzym. Microb. Technol.* **2012**, *51*, 294–299. [CrossRef]
112. Kantarci, N.; Borak, F.; Ulgen, K.O. Bubble column reactors. *Process Biochem.* **2005**, *40*, 2263–2283. [CrossRef]
113. Guerra-Renteria, A.S.; García-Ramírez, M.A.; Gómez-Hermosillo, C.; Gómez-Guzmán, A.; González-García, Y.; González-Reynoso, O. Metabolic Pathway Analysis of Nitrogen and Phosphorus Uptake by the Consortium between *C. vulgaris* and *P. aeruginosa*. *Int. J. Mol. Sci.* **2019**, *20*, 1978. [CrossRef] [PubMed]