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Review

A Review of the Harvesting Techniques of Microalgae

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Abstract: Algae are an important group of photosynthetic autotrophs and are commonly found in different types of water bodies, including paddy fields. The algal group possesses distinctive characteristics and ranges from prokaryotic cyanobacteria to eukaryotic algae. Within these, microalgae are unicellular microorganisms widely distributed in saltwater as well as freshwater environments. Microalgae species have been utilized in different fields, especially animal and human nutrition, medicine, bioremediation, and bio-fertilizers. Recently, numerous studies have reported the importance of microalgae in the production of biofuel. Further, microalgae have great carbon dioxide fixation efficiency during growth, so farmable land is not required for cultivating microalgae. Microalgae biomass production is a three-step process: cultivation, harvesting, and processing. Of these, the harvesting process is considered challenging due to its high cost, and it directly affects the processing step. In addition, several factors influence the harvesting process, including the size of microalgae cells (<30 μm), cultural conditions of microalgae, electronegative property of cell membrane, growth rate, etc. The harvesting of microalgae is an elaborate process that involves different chemical or mechanical approaches. A number of harvesting techniques have been utilized to recover algal biomass, such as membrane filtration, chemical and bio-flocculation, flotation centrifugation, sedimentation, and coagulation. In this context, this review aims to discuss various types of techniques used for harvesting microalgae. This review could be useful for selecting appropriate harvesting technology for enhancing the yield of microalgae biomass.

Keywords: algal biomass; Chlorella; microalgae; harvesting; flocculation; flotation

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1. Introduction

The algae group contains numerous photosynthetic eukaryotic species, which are usually distributed in fresh and marine water environments. The total number of living algal species ranges from 30,000 to 1 million, with distinctive features and properties from unicellular cyanobacteria to multicellular algae, and they are distributed worldwide [1]. Within these, microalgal species are fast-growing organisms with great capacity to survive and consume carbon dioxide during the growth process. Microalgae species have been used in a wide range of sectors for the benefit of mankind. Some of the important applications of microalgae are food, antibiotics and medicines, wastewater purification, biofuel, biofertilizers, and CO_2 fixation [2,3]. Furthermore, lipids produced from microalgae possess specific characteristics of neutrality and saturation levels, so they are considered to be possible substitutes for fossil fuels. Moreover, microalgae cells contain numerous bioactive substances, including lipids, proteins, carbohydrates, carotenoids, vitamins, etc. Hence, microalgae have been cultivated at an industrial level for the production of these commercially valuable metabolites [4].

The production process of microalgal biomass contains three major steps: cultivation, harvesting or collection, and processing. Of these, the microalgal harvesting step is a critical part of microalgae production. Previous studies have indicated that the cost for

the harvesting process is 20–30% of the total production cost [5]. The major challenges faced during microalgae harvesting and dewatering processes are small cell size (<30 μm), low concentration and the dilute nature of microalgal growth in culture medium (<1 g/L), exceptionally electronegative properties of the cell membrane surface, and relatively higher algal growth rate [6,7]. Consequently, the energy utilized throughout the harvesting process is higher when compared with the energy level of microalgal biomass [8].

In particular, the main aim of the harvesting method is to remove a maximum quantity of growth medium from the microalgal biomass, thereby facilitating the extraction process. In this context, several non-biological and biological harvesting techniques have been developed to collect microalgal biomass, including filtration, centrifugation, flocculation, and flotation [9]. Sometimes, a combination of two to three methods is performed to enhance harvesting efficiency. Although these harvesting methods provide some efficient results, they have some disadvantages such as being expensive, time-consuming, and harmful to the environment and requiring a high energy intake [3]. Further, these techniques account for major proportions of the total price of the collection of biomass in open systems [10].

Among the different harvesting methods, a semipermeable membrane has been employed in the filtration approach, which can hold microalgal biomass on the membrane while permitting the culture media to pass through. The filtration method can collect a high amount of cells from the culture medium. However, this method is susceptible to fouling and clogging. Hence, repeated replacement of a new filter or membrane is required [11]. The centrifugation method is also utilized to isolate microalgae cells from the growth media according to the density and particle size of each component. Although this method possesses a high cell harvesting efficiency, high time and energy consumption are major drawbacks. Further, centrifugation might cause cellular damage due to its high gravitational force [12].

Flocculation is an important harvesting method for microalgal cells. In this method, freely floating microalgae cells are accumulated and a larger particle called floc is formed by adding a chemical or bio-flocculant to eliminate the surface charge of cells [8]. However, the flocculation method has a major disadvantage owing to the high toxicity of chemical flocculants. Subsequently, additional treatment processes are required to remove these chemicals [3]. Additionally, the flotation method is used to stimulate the floating of microalgae cells on the culture media surface for harvesting easily by developing small bubbles. The flotation technique has some advantages over other methods due to its great harvesting efficiency, simple working process, and high processing throughput with low price [9].

The selection of the appropriate harvesting method is primarily based on the nature of the microalgal species used for cultivation, the microalgal cell density and size, the conditions of the end product, and the reutilization of the growth medium [3,8,13]. Based on the literature, the harvesting of microalgae is quite elaborate, and different mechanical-, chemical-, biological-, and electrical-based techniques are employed. In this context, this review aims to discuss various methods used for harvesting microalgae cells in order to understand and develop more effective microalgae harvesting techniques.

2. Harvesting Methods

The cultivation of microalgae cells has attracted increased interest, leading to the production of commercially valuable products. In general, various dewatering methods can be used according to the microalgae species. However, the high energy linked with microalgae harvesting develops a major bottleneck, which enables investigations of better, more cost-effective harvesting techniques. Different techniques used for harvesting microalgae are presented in Figure 1.

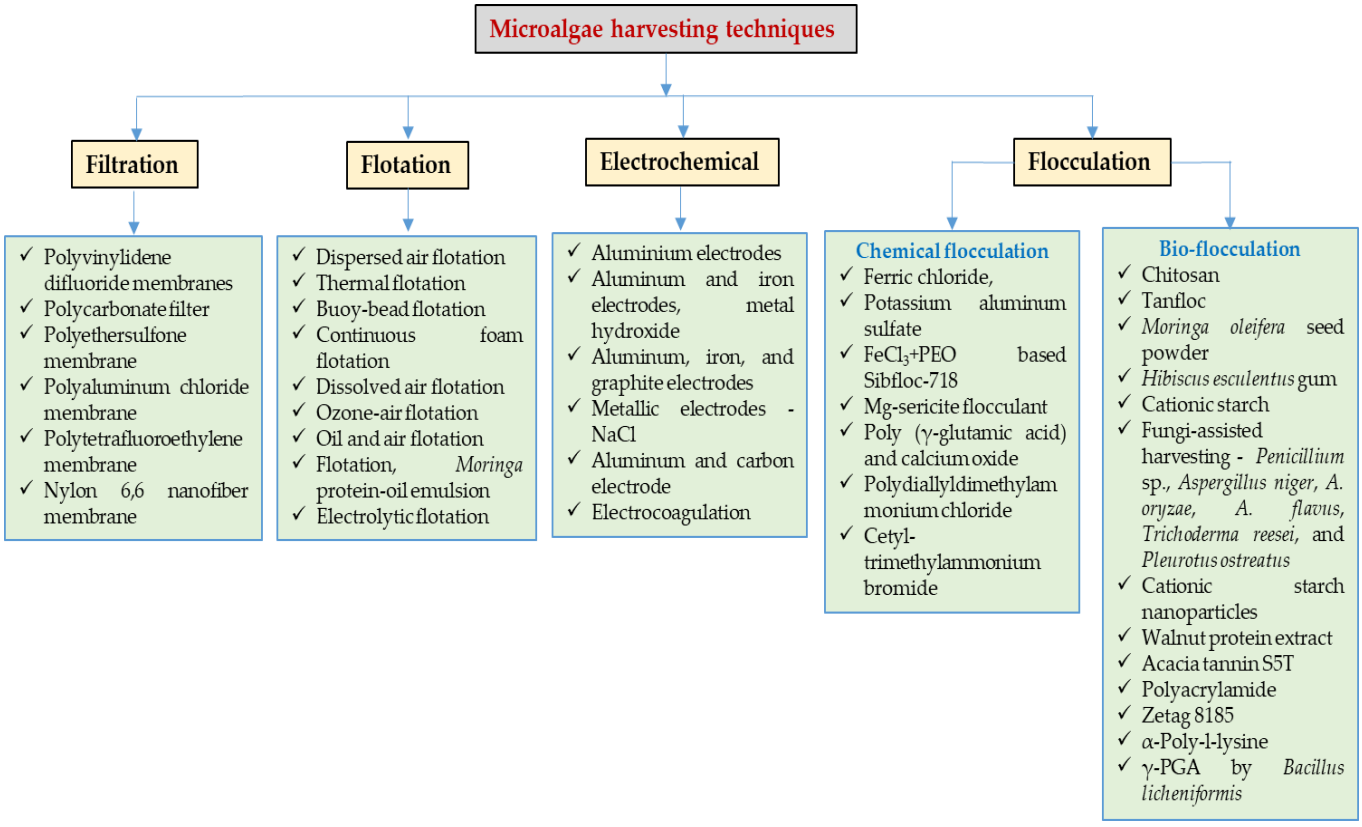

Figure 1. Different kinds of approaches utilized in microalgae harvesting.

2.1. Filtration

The filtration technique is an important physical method performed to isolate solids from liquids, in which only fluid can pass. Different types of filtration approaches, including microfiltration, ultrafiltration, vacuum filtration, pressure filtration, etc., are utilized to harvest microalgae biomass (Table 1 and Figure 2).

In the filtration method, membranes play a key role in harvesting microalgae because membrane fouling with expensive harvesting processes associated with low fluxes is a major issue. To diminish fouling formation, a surface-coating material like hydrophilic polyvinyl alcohol polymer was used in one study. The polyvinyl alcohol coating improved the hydrophilic nature of the membrane surface and performance by increasing the maximum flux (36%), with a 100% recovery rate [14]. Transparent exopolymer particles produced from microalgae have also been used to reduce membrane fouling in different filtration approaches. Discart et al. [16] explained the importance of transparent exopolymers in the fouling of microfiltration membranes for harvesting various broth solutions of *C. vulgaris*. Bilad et al. [21] studied the significance of submerged microfiltration for harvesting *C. vulgaris* and *Phaeodactylum tricornutum*. For harvesting *C. vulgaris* from algal froth, coagulation (polyaluminum chloride—Al_2O_3) followed by using a hydrophilic polytetrafluoroethylene membrane was an effective process [18].

Table 1. Microalgal biomass recovery by filtration technique.

Microalgae	Place	Filtration	Recovery (%)	References
Chlorella sp.	South Korea	Crossflow membrane filtration—hydrophilic polyvinyl alcohol polymer	100%	[14]
	South Korea	Ultrafiltration, fouling-resistant PVDF membranes	94% and 100%	[15]
	Germany	Transparent exopolymeric particles—polycarbonate filter	97%	[16]
Chlorella vulgaris	Istanbul, Turkey	Crossflow membrane filtration, UH050 membrane—hydrophilic polyethersulfone	100%	[5]
		Crossflow filtration, ultra-low-pressure filtration system	76%	[17]
	Southern Taiwan	Coagulation—polyaluminum chloride and polytetrafluoroethylene membrane	31% lipid, 28% protein, and 8% carbohydrate	[18]
	Malaysia	Nylon 6,6 nanofiber membrane, polyvinylidene fluoride phase-inverted membrane	Enhanced its competitiveness	[19]
	Perak, Malaysia	Nylon 6,6 nanofiber membrane	-	[20]
	Belgium	Combining the submerged membrane bioreactor microfiltration with centrifugation	-	[21]
Chlorella pyrenoidosa (Syn: *Chlorella vulgaris*)	China	Diatomite dynamic membrane	-	[22]
Nannochloropsis gaditana (Syn: *Microchloropsis gaditana*)	Tarragona, Spain	Dynamic filtration, polyethersulfone membrane	-	[23]
Dictyosphaerium sp.	Belgium	Combination of patterned membrane filtration and flocculation at standardized chitosan dosage, crossflow filtration, polyethylene glycol	Highest stable membrane permeance	[24]
Phaeodactylum tricornutum	Belgium	Combining the submerged membrane bioreactor microfiltration with centrifugation	-	[21]
	Tarragona, Spain	Dynamic filtration, polyethersulfone membrane	-	[23]
Aurantiochytrium sp.	South Korea	Dynamic filtration module, an FMX B-class	100%	[25]
Microalgae	UK	Microfiltration, porous superabsorbent polymer beads	90%	[26]
Desmodesmus sp.	Belgium	Polysulfone and polyethylene glycol	100%	[27]

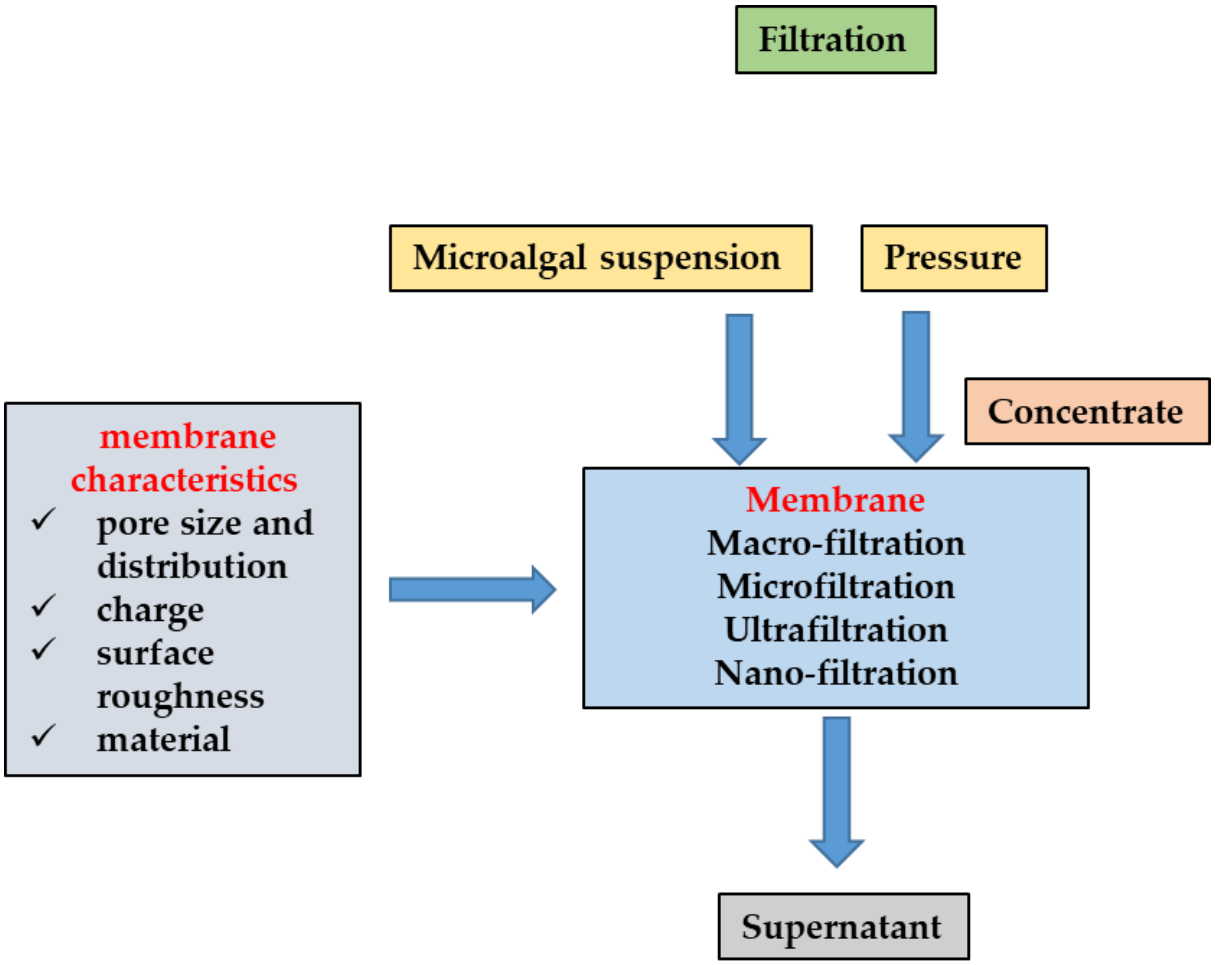

Figure 2. Filtration techniques employed in microalgae harvesting.

Zhang et al. [22] studied the influence of a diatomite dynamic membrane on the dewatering capacity of *C. pyrenoidosa* and found that the diatomite dynamic membrane interrupted polysaccharides, protein- and humic-like substances, and some low-molecular-weight organic compounds. Nurra et al. [23] reported that vibrating membrane filtration was the best technique to harvest the cells of *N. gaditana* and *Phaeodactylum tricornutum* when compared with conventional crossflow filtration techniques. Kim et al. [25] compared different chemical, physical, and mechanical approaches for harvesting *Aurantiochytrium* sp. The authors reported that about 100% harvesting efficiency with the minimum water level in *Aurantiochytrium* sp. biomass was attained through membrane filtration coupled with an anti-fouling turbulence generator. Another study showed that an algae retention of 100% was achieved using a polyvinylidene fluoride membrane combined with PEGylated polyethyleneimine particles and pluronic F-127 additive [15]. In the crossflow membrane filtration process, a microfiltration membrane (polyvinylidene fluoride—0.2 µm) and three ultrafiltration membranes (polyethersulfone, hydrophilic polyethersulfone, and regenerated cellulose) were used to harvest microalgal cells. Of these, the hydrophilic polyethersulfone (UH050) membranes exhibited higher performance in terms of affecting membrane fouling, transmembrane pressure, and crossflow velocity [5].

One study indicated that a nylon 6,6 nanofiber membrane exhibited higher filtration efficiency for *C. vulgaris* due to its higher pore size opening, surface pore density, and fouling resistance [19,20]. Zhao et al. [27] demonstrated that membranes with higher polysulfone and polyethylene glycol produced more noticeable patterns. Further, less membrane fouling and higher membrane fluxes were achieved with larger patterns. Recently, ultra-low-pressure membrane filtration combined with aeration was used for harvesting

C. vulgaris. In this process, the permeabilities of *C. vulgaris* broth significantly decreased [17]. A patterned polysulfone membrane prepared with polyethylene glycol (28%) exhibited the highest permeance of clean water and membrane in a microalgal suspension [24]). Chen et al. [26] fabricated porous superabsorbent polymer beads for the filtration of microalgal cultures. These beads possess high water absorption capacity in a microalgal suspension. In a microalgal concentrate, the beads can be easily separated and reused after treatment.

In the membrane filtration process, different types of permeable membranes have been used to filter microalgal biomass. Previously, several authors investigated the effect of mainly microfiltration and ultrafiltration membranes on microalgae harvesting. The membrane filtration methods can be improved with different pore sizes. Fouling is a major issue in the membrane filtration technique due to the clogging of pores. Hence, the pore size is the major criterion employed to categorize microfiltration (100–10,000 nm), ultrafiltration (2–100 nm), and nano-filtration (0.5–2 nm) membranes. In general, microfiltration membranes have a wide range of pore sizes, and ultrafiltration membranes have a narrower pore size, whereas nano-filtration membranes possess the smallest pore size [11]. Further, membranes with smaller pore sizes decrease the rate of filtration (permeation flux) (Liber et al., 2020). Polyvinyl chloride, polyvinylidene fluoride, polyethersulfone, polyacrylonitrile, and polytetrafluoroethylene are the extensively employed membranes [3].

Commercial membranes such as polyvinylidene fluoride (0.2 μm), polyvinylidene fluoride (150 kDa), and polyethersulfone (150 kDa) have been tested. At a uniform transmembrane pressure (100 kPa) and flow rate (8 L/min), polyvinylidene fluoride with a pore size of 0.2 μm registered a harvesting efficiency of 97.3% in 240 min operation time. Although polyethersulfone (150 kDa) showed a higher harvesting efficiency (99.8%) in 180 min operation time, the water content in the harvested microalgal biomass was 83.9%. However, the water content in the harvested microalgal biomass was 0% while using polyvinylidene fluoride membranes [25]. In another study, three commercial ultrafiltration membranes (30, 50, and 150 kDa) and one microfiltration membrane with a pore size of 0.2 μm were used. In all the tested membranes, nearly 100% microalgal biomass recovery was attained [5]. A nylon 6,6 nanofiber membrane with 25.82% surface porosity, 0.12 μm pore size, and clean water permeance of 1018 L/m^{2}h bar rapidly decreased its pristine value from 1018 to 528 L/m^{2}h bar within 15 min, and it further declined to 300 L/m^2 h bar toward the end of filtration [19]. Recently, ultra-low-pressure membrane filtration registered low energy consumption for harvesting *C. vulgaris* (4.4×10^{-3} kWh/m^3) [17]. Based on the previous findings, membranes with a pore size ranging from 40 to 100 kDa were found to be effective for long-term use. Moreover, ultrafiltration membranes exhibit better flux with fouling resistance when compared with microfiltration membranes.

2.2. Flotation

In recent times, flotation has become an important separation technique to remove microalgae from suspension. In the flotation approach, air or gas is converted into bubbles via a solid/liquid suspension. Consequently, solid particles in the medium are attached to gaseous molecules and are accumulated on the surface. Based on the size of the bubble, the flotation process is categorized into different types such as dissolved and dispersed air, electrolytic, and ozonation dispersed flotation approaches (Table 2).

Table 2. Microalgal biomass recovery by flotation technique.

Microalgae	Place	Flotation	Recovery (%)	References
Chlorella vulgaris	Taiwan	Dispersed air flotation	93%	[28]
	India	Dissolved air flotation	90%	[29]
	China	Surfactant, hexadecyltrimethyl-ammonium bromide and tea saponin.	89.23%	[30]
	China	Buoy-bead flotation, surface-layered polymeric microspheres	98.43%	[31]
	China	Thermal flotation	91.96%	[32]
	China	N,N′-bis(cetyl dimethyl)-1,4-butane diammonium dibromide	99.2%	[33]
	China	Buoy-bead flotation	89.9%	[34]
	Abu Dhabi	Colloidal gas aphrons technology, surfactants—cationic hexadecyl trimethyl ammonium bromide, anionic sodium dodecylbenzene sulfonate, sodium dodecyl sulfate, and combinations of these surfactants	95%	[35]
	China	Buoy-bead flotation, sodium alginate microspheres	93.78%	[36]
	UK	Continuous foam flotation, cationic trimethyl-ammonium bromide	96%	[37]
Chromochloris zofingiensis	US	Dissolved air flotation, dissolved organic matter, increasing Al^{3+} concentration	95.2%	[38]
Chlorella sp.	Mexico	Al^{3+} and cetyltrimethylammonium bromide	98.73%	[39]
Chlorella sorokiniana	Brotas, Brazil	Dissolved air flotation, pH modulation	96.5–97.9%	[40]
Scenedesmus obliquus (Syn: *Tetradesmus obliquus*)	Taiwan	Dispersed air flotation	93% microalgae	[28]
	India	Dissolved air flotation	90%	[29]
	China	Thermal flotation	91.96%	[32]
Ochromonas danica	USA	Oil and air flotation	98%	[41]
Dunaliella salina	France	Flocculation/flotation	80%	[42]
Arthrospira platensis	Abu Dhabi	surfactants—cationic hexadecyltrimethylammonium bromide, anionic sodium dodecylbenzene sulfonate, sodium dodecyl sulfate, and combinations of these surfactants	95%	[35]
Nannochloropsis sp.	Malaysia	Flotation, *Moringa* protein–oil emulsion	86%	[43]
	Malaysia	Dissolved air flotation, tannin-based biopolymer flocculant, AFlok-BP1	-	[44]
Nannochloropsis oculata	Abu Dhabi	surfactants—cationic hexadecyl trimethyl ammonium bromide, anionic sodium dodecylbenzene sulfonate, sodium dodecyl sulfate, and combinations of these surfactants	95%	[35]

In a dissolved air flotation method, Zhang et al. [38] suggested that the harvesting efficiency of *Chromochloris zofingiensis* reached more than 90% when increasing the dosage of $Al^{(3+)}$. Another study indicated that dispersed air flotation using saponin and chitosan was an effective strategy to harvest *C. vulgaris* and *Scenedesmus obliquus* [28]. For harvesting *C. vulgaris* using a flotation method, Shen et al. [30] found that surfactant hexadecyltrimethylammonium bromide registered higher harvesting efficiency when compared to tea saponin. In this method, hexadecyltrimethylammonium bromide neutralized the algal potential, whereas tea saponin changed the microalgal surface nature from hydrophilic to hydrophobic. The results revealed that hexadecyltrimethylammonium bromide and tea saponin surfactants could enhance the affinity between *C. vulgaris* and bubbles, allowing the microalgae to be harvested easily. In a continuous foam flotation process, Al-Humairi et al. [37] demonstrated that the percentage of harvesting efficiency for *C. vulgaris* improved when increasing the initial concentration of cationic trimethyl-ammonium bromide. In the case of *C. sorokiniana*, the pH modulation with the dissolved air flotation method is considered an efficient method for biomass harvesting from wastewater [40]. One study indicated that the dissolved air flotation technique with tannin-based biopolymer flocculant (AFlok-BP1) exhibited higher harvesting efficiency for marine *Nannochloropsis* sp. [44].

The buoy-bead flotation process is frequently used to enhance the harvesting efficiency of microalgae to minimize the level of chemicals. Based on this method, surface-layered polymeric microspheres were developed to harvest *C. vulgaris*. The data established that the highest harvesting efficiency (98.43%) was attained at a 0.7 g/L concentration of surface-layered polymeric microspheres. Further, surface-layered polymeric microspheres can be effectively reused for harvesting [31]. In another study, a harvesting efficiency of 89.9% was attained using the buoy-bead flotation method. Further, the authors reported that pH, microsphere diameter, and agitation speed highly influenced the harvesting efficiency for microalgae [34]. Another study indicated that sodium alginate microspheres were utilized to harvest *C. vulgaris.* The results showed that sodium alginate microspheres combined with a minimum amount of aluminum sulfate registered higher harvesting efficiency than air flotation and buoy-bead flotation methods [36].

The thermal flotation method was used for harvesting *C. vulgaris* and *S. obliquus*. The results showed that the harvesting efficiency for *S. obliquus* (88.16%) was higher than that for *C. vulgaris* (47.16%) because the thermal pre-flocculation process degraded the lipids, carbohydrates, and proteins on the cell surfaces of microalgae [32]. Hosseini et al. [41] developed an additive-free method for harvesting *Ochromonas danica* and achieved a 98% recovery rate. *Dunaliella salina* is an important microalgal species that is industrially utilized for its ability to yield higher quantities of carotenoid pigments. For harvesting this species, the flocculation/flotation method with the addition of NaOH was used [42].

In recent times, colloidal gas aphrons technology has been used in flotation. The stable colloidal gas aphrons method was used to remove *Arthrospira platensis*, *N. oculata*, and *C. vulgaris* [35]. A bio-flotation method using *Moringa* protein extract and oil emulsion was utilized to avoid chemical residues during the harvesting process of microalgae. This method provided 86.5% of harvesting efficiency for *Nannochloropsis* sp. [43]. Huang et al. [33] used N,N′-bis(cetyldimethyl)-1,4-butane diammonium dibromide (Gemini surfactant) for harvesting *C. vulgaris*. The novel Gemini surfactant registered excellent harvesting performance when compared to monomeric cetyltrimethylammonium bromide. To harvest *Chlorella* sp., Al^{3+} combined with cetyltrimethylammonium bromide greatly improved the algal floc size, and showed a higher level of hydrophobicity, thereby facilitating the flotation [39].

2.3. Flocculation

Flocculation is a widely used approach to harvest microalgal biomass. In this process, scattered units in the medium are accumulated together and these particles are settled down using various kinds of chemicals and bio-flocculants (Tables 3–5).

Table 3. Microalgal biomass recovery by chemical flocculation technique.

Microalgae	Place	Flocculation	Recovery (%)	References
Chlorella sp.	IARI, India, and Uppsala University, Sweden	Ferric chloride, potassium aluminum sulfate, chitosan solution	82%	[45]
	Republic of Korea	Acidified flocculation, coagulant—$Fe_2(SO_4)_3$ and H_2SO_4	98%	[46]
	China	$FeCl_3$ and polyacrylamide	90.5%	[47]
	Republic of Korea	Ca^{2+} and CO_3^{2-}, amorphous nano-flakes, rhombohedral calcites, and spherical vaterites	90–99%	[48]
	Texas, USA	Centrifugation or flocculation with $FeCl_3$	90%	[49]
	Russia	Mixture of coagulant—$FeCl_3$ and flocculant—PEO-based Sibfloc-718	90%	[50]
	Republic of Korea	Mg-sericite flocculant	99%	[51]
	China	Mixture of flocculants, poly-γ-glutamic acid, and calcium oxide	95%	[52]
Chlorella vulgaris	India	Alum and ferric chloride	90%	[29]
	Israel	Polydiallyldimethylammonium chloride	90%	[53]
	Trebon, Czech Republic	Cooking oil (rapeseed oil) in an aqueous solution of cetyl-trimethylammonium bromide (2.7 mg/L)	90%	[54]
	Austin, USA	Fe^{3+} ($FeCl_3$), chitosan, and Ca^{2+} ($CaCl_2$)	43.2%, 49.5% and 39.6%	[55]
	Tehran, Iran	Alum and pH adjustment	90%	[56]
	Wuhan, China	Sulfate ($Al_2(SO_4)_3$ and $Fe_2(SO_4)_3$) and chloride flocculants ($AlCl_3$ and $FeCl_3$)	93.5–98.8%	[57]
	Australia	Polyacrylamide addition, alkaline addition, and centrifugation	-	[58]
	Oban, UK	Aluminum sulfate, ferric sulfate, and ferric chloride	98.8%	[59]
Chlorella sorokiniana	Spain	$AlCl_3$	95.23%	[60]
Porphyridium purpureum	Australia	Polyacrylamide addition, alkaline addition, and centrifugation	-	[58]
Phaeodactylum tricornutum	Belgium	Brucite and calcite	90%	[61]
	Australia	Polyacrylamide addition, alkaline addition, and centrifugation	-	[58]
Synechocystis sp.	IARI, India and Uppsala University, Sweden,	Ferric chloride, potassium aluminum sulfate, chitosan solution	82%	[45]
Scenedesmus sp.	Doha, Qatar	Coagulation flocculation (ferric chloride (72–96 mg/L)	90%	[62]
Scenedesmus obliquus (Syn: (*Tetradesmus obliquus*)	India	Alum and ferric chloride	90%	[29]
Dunaliella salina	India	Potash alum or $FeCl_3 \cdot 6H_2O$	99%	[63]
Arthrospira maxima (Syn: *Limnospira maxima*)	Porto, Portugal	NaOH or $CaCl_2$	90%	[64]
Scenedesmus acuminatus (Syn: (*Tetradesmus lagerheimii*)	China	Alum coagulation with extracellular polymeric substances	-	[65]

Table 4. Microalgal biomass recovery by electro-flocculation technique.

Microalgae	Place	Electro-Flocculation	Recovery (%)	References
Chlorella sp.	IARI, India, and Uppsala University, Sweden	Different DC voltages (6, 9, and 12 V)	98%	[45]
Chlorella vulgaris	China	Aluminium electrolysis	98%	[66]
	China	Flocculant-free electrolytic flotation	90%	[67]
Synechocystis sp.	IARI, New Delhi, India, and Uppsala University, Sweden	Different DC voltages (6, 9, and 12 V)	98%	[45]
Nannochloropsis oculata	UK	Salt bridge electro-flocculation (300 mA in 45 min.)	90.4%	[68]
Dunaliella salina	Iran	Aluminum electrodes	97.44%	[69]
	China	Electro-flocculation	95.13% to 98.09%	[70]
	China	Precipitation of aluminum hydroxide hydrates	97%	[71]

Table 5. Microalgal biomass recovery by bio-flocculation technique.

Microalgae	Place	Bioflocculation	Recovery (%)	References
Chlorella sp.	IARI, India, and Uppsala University, Sweden	Chitosan	98%	[45]
	Brazil	Tanfloc, seed powder of *Moringa oleifera*, gum from *Hibiscus esculentus*, and cationic starch	80.3 to 92%	[72]
	USA	Fungi-assisted harvesting, *Penicillium* sp.	99.26%	[73]
	Malaysia	*Aspergillus niger*	90%	[74]
	China	Edible fungi-assisted harvesting—*Pleurotus ostreatus*	64.86%	[75]
	China	Microbial flocculant poly (γ-glutamic acid)	90%	[76]
	USA	Fungal pelletization—*Aspergillus niger*	90%	[77]
	USA	Yeast modified with 2-chloro-N,N-diethylethylamine hydrochloride	-	[78]
Chlorella vulgaris	India	*Strychnos potatorum*	99.68%	[79]
	India	Chitosan	90%	[29]
	Tehran, Iran	Cationic starch nanoparticles	90%	[80]
	Finland	Chitosan	90%	[81]
	Wuhan, China	Chitosan (10 mg/L), neutral pH	89%	[82]
	Wuhan, China	Walnut protein extract	40%	[82]
	Wuhan, China	Chitosan (6 mg/L) and walnut protein extract	98%	[82]

Table 5. *Cont.*

Microalgae	Place	Bioflocculation	Recovery (%)	References
	China	*Aspergillus oryzae*	99.23%	[83]
	Vakin, Umeå, Sweden	Cationic starch, chitosan, and acacia tannin S5T	93%	[84]
Chlorella vulgaris	Wuhan, China	Chitosan, Tanfloc, cationic starch, and *Moringa oleifera*	>90%	[85]
	China	Chitosan and polyacrylamide	98.10% and 94.57%	[86]
	Oban, UK	Zetag 8185, chitosan, Tanfloc SG	97.9%	[59]
Chlorella pyrenoidosa (Syn: *Auxenochlorella pyrenoidosa*)	China	Chitosan	96.83%	[87]
Microalgae	Brazil	Tannin-based coagulant	84%	[88]
Microalgae	Brazil	Tannin-based coagulant	90%	[89]
Chlorella protothecoides (Syn: *Auxenochlorella protothecoides*)	China	Microbial flocculant poly (γ-glutamic acid)	90%	[76]
	UK	Cationic starch—coagulation flocculation	80%	[90]
	Texas, USA	Chitosan	99%	[91]
Chlorella sorokiniana	Mexico	*Aspergillus flavus*-assisted pelletization	96%	[92]
	Canada	Flocculant hairy cationic nanocrystalline cellulose	82%	[93]
Chlorella ellipsoidea (Syn: *Chloroidium ellipsoideum*)	Republic of Korea	Bio-polymeric flocculant α-poly-l-lysine	98%	[94]
Nannochloropsis oculata	Belgium	Cationic cellulose nanocrystals	95%	[95]
Desmodesmus brasiliensis		γ-PGA obtained from *Bacillus licheniformis*	98%	[96]
Synechocystis sp.	Brazil	Tanfloc, seed powder of *Moringa oleifera*, gum from *Hibiscus esculentus*, and cationic starch	80.3 to 92%	[72]
Scenedesmus sp.	Thailand	*Aspergillus niger*, *Trichoderma reesei*, and *Aspergillus oryzae*—pellet formation	94%	[97]
Scenedesmus obliquus (Syn: (*Tetradesmus obliquus*)	Vakin, Umeå, Sweden	Cationic starch, chitosan, and acacia tannin S5T	93%	[84]
	Wuhan, China	Chitosan, Tanfloc, cationic starch, and *Moringa oleifera*	>90%	[85]

2.3.1. Chemical Flocculation

Chemical flocculation is regarded as an effective technique for microalgae harvesting. In one study, the flocculant Mg-sericite was also utilized to harvest *C. vulgaris.* It was reported that the pH of the growth medium highly affects the harvesting efficiency [51]. Ma et al. [52] used combined flocculants, poly (γ-glutamic acid), and calcium oxide to harvest *C. vulgaris*. The results demonstrated that the concentration of flocculants was significantly decreased while using combined flocculants. Gerchman et al. [53] compared the flocculation properties of polydiallyldimethylammonium chloride (cationic polymer) with chitosan and Superfloc® for the sedimentation of *C. vulgaris* and found that polydiallyldimethylammonium chloride was the most efficient one, with 90% flocculation efficiency at the concentration of 5 mg/L (60 min at pH10). Further, this flocculant was very effective in improving the harvest of *N. salina* through filtration. In another study, brucite or $Mg(OH)_2$ was used to harvest the diatom *Phaeodactylum tricornutum* [61]. In a recycled medium, it was found that flocculation with $FeCl_3$ showed a significant effect on the *C. vulgaris* biomass without affecting its lipid profile [49]. Similarly, *Scenedesmus* sp. was harvested from BG-11 media and wastewater using ferric chloride. The harvesting efficiency was improved when reducing the pH of the culture below 6.5 [62]. One study found that microwave-assisted flocculation effectively reduced the concentration of flocculants in the culture medium [55].

A metal coagulant ($Fe_2(SO_4)_3$) and sulfuric acid were also employed for harvesting *Chlorella* sp. KR-1. In addition, the authors suggested that the acid-treated *Chlorella* sp. KR-1 biomass could be directly utilized for extracting lipids without adding any catalyst [46]. Koley et al. [29] investigated different harvesting techniques to harvest *S. obliquus*, and the results showed that the maximum flocculation efficiencies were achieved for $FeCl_3$ (80.2%), alum (95%), and chitosan (91%). Also, similar trends were noticed for harvesting *C. vulgaris*. The flocculant $FeCl_3 \cdot 6H_2O$ (0.50 mM) was found to be an effective process to harvest *D. salina* at pH 9 [63]. *Arthrospira maxima* biomass was effectively harvested through flocculation using 0.2–2.0 g/L of $CaCl_2$ [64]. Lin et al. [47] developed a homemade helical tube for harvesting microalgal biomass of *Chlorella*. In this process, a recovery rate of nearly 90% was attained using the diluted algal solution of 0.12 g/L with flocculation and sedimentation times of 18 s and 15 min, respectively. The formation of $CaCO_3$ in the culture medium is also an effective strategy to harvest microalgal biomass [48].

Machado et al. [59] investigated the effect of different inorganic and organic flocculants on harvesting efficiency for *C. vulgaris*. Of these, Zetag 8185 showed maximum efficiency at 50 mg/L (98.8%) and 100 mg/L (97.9%). Another study reported the use of the alum flocculation method to harvest *C. vulgaris* biomass. Another study reported that the highest flocculation efficiency (>90%) was attained at the concentration of 0.5 g/L flocculant with pH 8.2 of the growth medium [56]. A recent study indicated that sulfate flocculants (aluminum sulfate and ferric sulfate) and chloride flocculants (aluminum chloride and ferric chloride) were utilized for *C. vulgaris* harvesting. The data demonstrated that the flocculation efficiency of these flocculants was between 93.5 and 98.8% at the concentration of 60 mg of sulfate salts and 100 mg of chloride salts per liter of algal culture. Further, the used flocculants did not alter the composition of the biomass [57]. In one flocculation process, alkaline pH played a key role in the morphology of harvested cells of *C. vulgaris*, *Porphyridium purpureum*, and *Phaeodactylum tricornutum*. Further, the addition of polyacrylamide led to a higher harvesting efficiency with higher retention of industrially important chemical components [58].

2.3.2. Electro-Flocculation

In one study, different flocculation approaches such as auto-flocculation, chemical, and electrolytic flocculation were employed to harvest *Chlorella* sp. MJ 11/11 and *Synechocystis* PCC 6803. Among them, electro-flocculation registered the highest flocculation efficiency of 98% [45]. In another study, electro-flocculation using aluminum electrolysis was employed to harvest *C. vulgaris*. Microalgae were harvested rapidly when applying higher current density [66]. An effective electro-flocculation approach integrated with

local sand was investigated for harvesting *D. Salina*. In this method, the average electrical energy consumption was decreased to 51.03% when compared with conventional electro-flocculation approaches [70]. In the electro-flocculation process, an aluminum–air battery was successfully employed for harvesting *D. salina* [71]. Zenouzi et al. [69] also utilized the electro-flocculation strategy to harvest microalgae *Dunaliella* biomass. Luo et al. [67] investigated the electrolytic flotation process without any flocculants to harvest microalgae. In this process, stainless steel (cathode) and carbon (anode) were chosen according to the harvesting efficiency. One study reported that alkali-induced flocculation integrated with an electrolysis (salt bridge electro-flocculation) approach was effectively employed to harvest microalgae biomass. Moreover, the salt bridge strongly inhibited microalgal cells from being damaged by the oxidation of anodes and there was no external contaminant of the algal biomass [68]. Fayad et al. [98] studied the improvement of harvesting technology for *C. vulgaris* using electro coagulation flocculation with aluminum and iron electrodes. The aluminum electrodes showed a higher harvesting efficiency, and this process had no effect on the level of *C. vulgaris* lipids and pigments.

2.3.3. Bio-Flocculation

Among the different flocculation strategies, bio-flocculation is an attractive technology to harvest microalgae to avoid adverse effects caused by chemical residues. In this context, chitosan (cationic polyelectrolyte) is extensively used for harvesting many algal species due to its non-toxic and biodegradable properties. However, the cost of chitosan is too high, which initiated research on finding other bio-flocculants. One study reported that the flocculation efficiency of chitosan (6 mg/L) was increased up to 98% in the presence of walnut protein extract [82]. In a comparative study, >90% of microalgal biomass yield was attained at the optimal concentration of 0.25 g/L chitosan [81]. Chitosan was also used to harvest *C. sorokiniana*, and its flocculation efficiency reached >99% at a pH below 7 [91]. Cationic starch, Greenfloc 120, was also used for harvesting *C. protothecoides*, and the maximum flocculation efficiency was attained at pH 7.7 and 10 [90]. Lopez-Exposito et al. [93] studied the flocculation efficiency of cationic nanocrystalline cellulose for harvesting *C. sorokiniana* suspensions and found that cationic nanocrystalline cellulose successfully flocculated *C. sorokiniana* cultures at concentrations below or above the isoelectric point. α-Poly-l-lysine (a cationic biopolymer) was employed to harvest microalgae, and this bio-flocculant effectively inhibited biological contamination due to its inherent antimicrobial activity [94]. Wang et al. [86] optimized conditions for harvesting *C. vulgaris* using chitosan and polyacrylamide. Chitosan (98.10% at 10 mg/L) registered a higher flocculation efficiency when compared with polyacrylamide (94.57% at 25 mg/L). In addition, there were no changes in the chemical composition of biomass.

Some studies have reported the use of microbial metabolites as bio-flocculants. Prochazkova et al. [78] confirmed the flocculation efficiency of spent brewer's yeast to harvest *C. vulgaris*. After hydrolysis, 2-chloro-N,N-diethylethylamine hydrochloride was used to chemically modify yeast, then the flocculation efficiency to harvest *C. vulgaris* was determined. Poly-γ-glutamic acid was employed for harvesting *C. vulgaris* and *C. protothecoides*. Further, the results demonstrated that there was no damage to the harvested microalgal cells and thereby no lipid loss during the flocculation process [76]. One study demonstrated that poly-γ-glutamic acid obtained from *Bacillus licheniformis* was employed to harvest *Desmodesmus brasiliensis* [96]. Luo et al. [75] studied the harvesting efficiency of *Pleurotus ostreatus* for harvesting *Chlorella* sp. Another study found that the filamentous fungus *Trichoderma reesei* QM 9414 exhibited an excellent pellet-forming potential for harvesting *Scenedesmus* sp. [97].

Co-cultivation of *Aspergillus niger* and *C. vulgaris* with the addition of glucose (2 g/L) exhibited a harvesting efficiency of >90%. The results suggested that the carbon source was essential to improve fungal growth and produce cell pellets [77]. Toscano et al. [92] reported that the co-cultivation *Aspergillus flavus* and *C. sorokiniana* was found to be the most efficient for forming pellets in nutrient-supplemented BG-11. *Aspergillus niger* was also utilized

as a bio-flocculant to harvest microalgae. The bio-flocculant exhibited the capability to adapt to a broad range of pH (3.0–9.0) [74]. Chu et al. [83] used *Aspergillus oryzae* pellets to harvest *C. vulgaris*, and the maximum harvesting efficiency (99.23%) was achieved at 30 °C, 130 rpm, and a 1:1 fungi:algae ratio. The authors suggested that metabolites produced in the medium might be responsible for its bio-flocculation property. Fungal spore- and pellet-assisted methods for harvesting *Chlorella* sp. were employed by Chen et al. [73]. In this process, co-cultivation of *Chlorella* sp. with *Penicillium* sp. spores or pellets registered the highest flocculation efficiency (99%).

Strychnos potatorum seed powder was also used to harvest *C. vulgaris*, and the highest efficiency (99.68%) was attained under the standardized conditions of 100 mg/L bioflocculant concentration, 35 °C, and 150 rpm, with an incubation time of 30 min [79]. Cassini et al. [72] compared alternative coagulants with the chemical coagulant aluminum sulfate for harvesting microalgae cells. Among the different coagulants, cationic starch registered a higher microalgae biomass yield in a wide range of pH. In an acidic pH range, the seed powders of *Moringa oleifera* and *Hibiscus esculentus* gum improved biomass recovery by up to 50%. Extracellular polymeric substances obtained from *S. acuminatus* were used to harvest the same algae through the flocculation approach. The results exhibited that the addition of extracellular polymeric flocculant at 3.2 mg/g markedly decreased the usage of alum coagulant (Al^{3+}) from 77.6 to 4.5 mg/g [65]. The flocculation efficiency of cationic cellulose nanocrystals for harvesting *N. oculata* was investigated [95]. Niemi and Gentili [84] studied the effect of natural organic flocculants, and the results suggested that tannin S5T registered the same flocculation efficiency in the tested microalgae. In another study, four natural flocculants, chitosan, Tanfloc, cationic starch, and *Moringa oleifera*, were employed to harvest *C. vulgaris* and *S. obliquus*. Among them, Tanfloc presented the highest harvesting efficiency (98%) for *C. vulgaris* at 30 mg/L and for *S. obliquus* at 20 mg/L [85].

2.4. Electrochemical Harvesting

Electrochemical techniques are one of the recent strategies for harvesting microalgal biomass, and are mainly based on the view of electrocoagulation, electro-flocculation, etc. Several kinds of electrodes have been used in the electrochemical-based harvesting of microalgae (Table 6).

Table 6. Microalgal biomass recovery by electrochemical techniques.

Microalgae	Place	Electrochemical	Recovery (%)	References
Chlorella vulgaris	France	Aluminum and iron electrodes, metal hydroxide	36.6%	[98]
	Iran	Aluminium electrodes—carbon cloth (anode) and stainless-steel (cathode)	98.00%	[99]
	India	Electroflotation	99%	[29]
	Czech Republic	Electrocoagulation, electrolysis with iron electrodes	85%	[100]
Nannochloropsis oculata	Iran	Aluminum, iron, and graphite electrodes	89.68%	[101]
Scenedesmus obliquus (Syn: (*Tetradesmus obliquus*)	South Africa	Metallic electrodes—NaCl	83%	[102]
	India	Electroflotation	99%	[29]
Arthrospira platensis	Brazil	Electrocoagulation flotation, aluminum and carbon electrode	98–99%	[103]
Tribonema sp.	China	Electroflotation	96.3%	[104]
Tetraselmis sp.	Doha, Qatar	Electrocoagulation (asymmetrical aluminum electrodes)	90.9%	[105]
	Qatar	Electrocoagulation, interdigitated electrodes	96.18%	[106]

A recent study reported that graphite electrode treatment (67.44%) registered the lowest harvesting efficiency of *N. oculata*. Conversely, a higher harvesting efficiency of *N. oculata* was achieved when using aluminum and iron electrodes. In the electrocoagulation technique, the biochemical composition of *N. oculata* was significantly altered by different electrodes [101]. Misra et al. [102] used non-sacrificial electrodes for harvesting *S. obliquus* in order to avoid the depletion of metallic electrodes. For harvesting microalgae, carbon cloth (anode) and the stainless-steel (cathode) were used to replace aluminum electrodes, and these electrodes showed the highest flotation efficiency (98%) and registered the lowest pollution owing to their less corrosive properties [99].

Hawari et al. [105] developed a new electrocoagulation electrode for harvesting *Tetraselmis* sp. by inducing dielectrophoretic force. In electrocoagulation, a new cylindrical interdigitated electrode array was also used to harvest *Tetraselmis* sp. Moreover, the microalgal harvesting efficiency increased up to 96.18% while shortening the distance of the electrode to 0.5 cm [106]. A continuous electrocoagulation approach was developed to harvest *C. vulgaris*. In this process, harvesting efficiency above 85% with a minimum level of Fe contamination was attained [100]. Recently, Qi et al. [104] compared the harvesting efficiency of microalgae through electroflotation without coagulation in terms of different stages of hydrophobicity. The authors suggested that the hydrophobicity of microalgae played a crucial role in the electroflotation process. Among the tested microalgae, the harvesting efficiency of *Tribonema* sp. reached up to 96.2%. However, a significant decrease in the harvesting efficiency for *Scenedesmus* sp. (70.1%) and *Pandorina* sp. (10%) was observed.

2.5. Other Harvesting Methods

Apart from filtration, flotation, and flocculation techniques used in isolation for harvesting microalgae, combinations of one or two of these methods with modifications using recent technologies have been developed (Table 7).

Table 7. Microalgal biomass recovery by some other techniques.

Microalgae	Place	Methods	Recovery (%)	References
Chlorella sp.	Republic of Korea	Coagulation ($FeCl_3$ and $Fe_2(SO_4)_3$)	99%	[107]
	Republic of Korea	1 mM of $FeCl_3$ and 0.5% of H_2O_2	90%	[108]
	Romania	Activated algae granules	99%	[109]
	India	Coagulation, *M. oleifera*	95.76%	[110]
	Prague, Czech Republic	Magnetic particles (diethylaminoethyl and polyethylenimine)	90%	[111]
		Co-flocculation/air flotation (helix tube flocculation reactor)	94%	[112]
	Brazil	Sedimentation	97.8%	[113]
	Iran	sedimentation	66.00%	[99]
	Greece	Magnetic harvesting (microwave-synthesized naked magnetite (Fe_3O_4) particles)	99%	[114]
Aurantiochytrium sp.	Republic of Korea	Centrifugation	90%	[107]
Nannochloris sp.	USA	Centrifugation with high flow rate	90%	[115]
Nannochloropsis maritima	China	Magnetic nanoparticles, Fe_3O_4 nanoparticles	95%	[116]
Scenedesmus sp.	Spain	Adsorbents of magnetite-based nanoparticles (Fe_3O_4 NPs)	95%	[117]
	India	Coagulation, *M. oleifera*	95.76%	[110]
Desmodesmus brasiliensis	China	Foam separation (natural surfactant cocamidopropyl betaine)	93.6%	[118]
Synechocystis sp.	India	Coagulation, *M. oleifera*	95.76%	[110]
Spirulina sp.	India	Coagulation, *M. oleifera*	95.76%	[110]

Activated algae granules comprising *Chlorella* sp. were developed to replace microalgae–bacteria flocs. The granulation processes occurred in the presence of *Phormidium* sp., and a recovery efficiency of >99% was obtained through fast sedimentation of the granules [109]. Among the different harvesting methods, the pH-stimulated sedimentation technique is an inexpensive and simple one. More than 97.8% harvesting efficiency was attained for *C. sorokiniana* under the optimal sedimentation conditions of 250/s velocity gradient, 10 s mixing time, and pH 12 [113]. A thermal-tolerant species, *C. pyrenoidosa*, was easily harvested when cultured at 40 °C when compared with culturing at low temperatures. The improvement in harvesting microalgal cells at high temperatures might be ascribed to the increment in cell size and reduction in cell surface charge [87]. One study indicated that an emulsion consisting of cooking oil and cetyltrimethylammonium bromide was utilized for harvesting *C. vulgaris*. Further, the emulsion concentration and pH were adjustable for harvesting *C. vulgaris* based on the technological requirements [54]. Magnetic beads with diethylaminoethyl and polyethylenimine were investigated in connection with harvesting *C. vulgaris*. Both magnetic beads exhibited optimal harvesting efficiencies of >90%, but efficient detachment was attained only for diethylaminoethyl magnetic beads [111].

To improve the dissolved air flotation approach, a co-flocculation or air flotation method was established for harvesting *Chlorella* sp. 64.01 biomass. In a co-flocculation device, an ejector and a helix tube flocculation reactor were used. In this process, aerated flocs were more stable by encapsulating micro-bubbles into microalgae flocs [112]. It was reported that wastewater from the oxidized dye (methylene blue and methyl orange) was evaluated for its ability to be used as a coagulant to harvest microalgae. A higher harvesting efficiency of >90% was achieved for both methylene blue and methyl orange at a 5:1 ratio (dye wastewater:cell culture) [108]. Natural coagulants prepared from *different plant* species were utilized for harvesting microalgal biomass. Among them, the highest harvesting efficiency was attained for *M. oleifera* extract (8 mg/mL), and its harvesting efficiency improved to 95.76% with the addition of chitosan (0.75 mg/mL) [110]. For harvesting *Scenedesmus* sp. from wastewater, magnetite-based nanoparticles (Fe_3O_4 NPs) were used. A harvesting efficiency of >95% was achieved for these nanoparticles [117].

Fe_3O_4 nanoparticles were developed to harvest *N. maritima* biomass, and its recovery efficiency reached above 95% at 120 mg/L. The growth medium obtained from magnetic separation was also reused and similar biomass production was achieved [116]. For harvesting *C. vulgaris*, iron oxide magnetic microparticles were also used. The acid treatment process was performed to completely demagnetize the harvested algae [111]. In another study, microwave-assisted naked magnetite (Fe_3O_4) particles were utilized to harvest *C. vulgaris* and a 99% harvesting efficiency was attained at pH 3.0. After the treatment, recovered particles could be recycled a minimum of five times without affecting their harvesting effectiveness [114].

Although several techniques have been used to collect microalgal biomass, each technique has some advantages as well as disadvantages in terms of recovery percent, time, cost-effectiveness, laboratory or industrial applications, etc. (Table 8). Of these, only certain methods have been employed to transfer technologies from the laboratory level to large-scale level.

Table 8. Advantages and disadvantages of different microalgae harvesting techniques.

Harvesting Methods	Advantages	Disadvantages	References
Filtration	✓ Easy harvesting from 10 min to 24 h ✓ Biomass recovery 76–100% ✓ Inexpensive and chemical-free method ✓ Highly suitable for industrial-level cultivation	✓ Fouling and clogging issues for small algal cells ✓ Mainly suitable for large algal cells ✓ The filter membrane should be cleared at regular intervals	[14,15,17,19,22,26]
Flotation	✓ Different types of flotation approaches with short harvesting time ✓ Biomass recovery 80–98.73% ✓ Large-scale harvesting method ✓ Affordable cost	✓ Certain chemical surfactants are expensive ✓ Suitable for some specific algal species	[33,39,40,42]
Chemical flocculation	✓ Predominantly, sulfate and chloride flocculants are used ✓ Biomass recovery 82–99% ✓ Commercial-scale use ✓ Inexpensive, rapid, and easy harvesting process	✓ Removal of chemicals from algal cells is required ✓ Chemicals may be toxic to algal cells	[57–59]
Electro-flocculation	✓ Suitable for all species of microalgae ✓ Biomass recovery 90% to 98% ✓ Chemical-free method ✓ Rapid and easy harvesting method	✓ High energy consumption ✓ Metal electrodes are expensive	[45,66,67]
Bio-flocculation	✓ Chiefly, chitosan and microorganisms are used ✓ Biomass recovery 80% to 99.68% ✓ Eco-friendly and non-toxic ✓ Rapid, industrial-level, and inexpensive technique ✓ Culture medium can be reused	✓ Contamination due to other microorganisms	[79,84,85,90]

Table 8. *Cont.*

Harvesting Methods	Advantages	Disadvantages	References
Electrochemical techniques	✓ Rapid ✓ Chemical-free and easy harvesting techniques ✓ Biomass recovery 36.6% to 99%	✓ Require high energy consumption ✓ Metal electrodes are expensive	[98,99,103]
Coagulation	✓ Chemical and bio-coagulants are used ✓ Biomass recovery 95.7% to 99% ✓ Rapid, industrial-level, and easy harvesting process	✓ Chemicals may be toxic ✓ Chemical coagulants are expensive ✓ Difficult to remove the coagulant from algal cells	[25,108,109]
Sedimentation	✓ Biomass recovery 66% to 97% ✓ Inexpensive, simple, and less energy needed	✓ Appropriate for particular species ✓ Time-consuming process	[99,113]
Centrifugation	✓ Biomass recovery 90% ✓ Rapid and small-scale laboratory technique	✓ Expensive method ✓ Cell damage due to high speed	[25,115]
Magnetic harvesting	✓ Magnetic nanoparticles and Fe_3O_4 nanoparticles are used ✓ Biomass recovery 95% ✓ High harvesting efficiency with short time	✓ Expensive method	[116,117]

It is well established that microalgae are considered excellent feedstocks for the production of biodiesel, bioethanol, and biogas due to their excessive growth rates, higher lipid content, and nontoxicity. In particular, different species of *Chlorella*, *Scenedesmus*, and *Saccharina* have been widely investigated for producing biohydrogen. The highest hydrogen recovery was obtained from *C. vulgaris* [119]. Among the different harvesting techniques, coagulation, filtration, and centrifugation are the most important approaches for biofuel application. These techniques could be employed alone or in combinations to improve microalgal harvesting efficiency. Further, the harvesting efficiency for gravity sedimentation techniques can be improved by flocculants [3]. Further, electro-flocculation is an effective process for harvesting microalgae with higher harvesting efficiency. The flotation technique is also considered to be an effective harvesting process for microalgae-based biofuel industries. The flotation approach registered significant harvesting efficiency above 88.8% [4].

Various harvesting approaches were employed for improving microalgal biomass recovery [120]. Of these, the flocculation method is highly suitable for large-scale harvesting of microalgae. Although a high flocculation efficiency is obtained while using inorganic chemical substances, the chemical flocculants are difficult to fully separate from the growth medium, which results in water pollution. Bio-flocculants like chitosan can be used in place of chemicals. However, the cost of polymers is high for industrial applications [121,122]. One study found that pH-induced flocculation by NaOH appeared to be an effective, inexpensive, and environmentally friendly technique due to the reusability of the growth medium [29]. Membrane technology appears to be one of the most important microalgae harvesting technologies in terms of simplicity, cost-effectiveness, low energy consumption, and higher biomass recovery. A recent advancement in the harvesting of microalgae is magnetic nanoparticles. A nanoparticle-associated approach showed a great microalgal harvesting efficiency at the laboratory level. However, energy-intensiveness is the major disadvantage in terms of large scale. A recent study demonstrated that the combination of flocculation and filtration approaches significantly improved the performance of microalgae harvesting. Under optimum operating conditions, the combined technology significantly reduced the estimated total cost to 0.139 USD/kg when compared with the filtration method without the association of flocculation (0.206 USD/kg) [123].

3. Conclusions and Future Perspectives

For commercializing microalgae and their products, the selection of appropriate harvesting techniques is a challenging task. The results of previous studies demonstrated that *Chlorella* species are the most-studied microalgae due to their various industrial applications. Some of the other industrially important microalgae are *Scenedesmus* sp., *Nannochloropsis* sp., *Dunaliella* sp., and *Tetraselmis* sp. Various factors are involved in harvesting microalgae biomass, including the harvesting method, microalgal species, pH of the culture medium, medium composition, chemical residue after harvesting, nature of water bodies, etc. Each harvesting method has its own merits and demerits. Of the different techniques, the flocculation process using chemical or natural flocculants has been widely used to harvest different microalgae. In particular, natural organic flocculants (bioflocculants) are utilized for cost-effectiveness and reduced contamination level in the harvested biomass. To some extent, combinations of two or more harvesting approaches could be employed for effective microalgal harvesting like coagulation and flocculation or filtration and centrifugation. However, these techniques are time-consuming and expensive. Electrochemical methods have been used to avoid/reduce chemical contaminations in microalgal biomass, but these processes are more energy-consuming and highly expensive for large-scale operations. Recently, nanoparticles have been extensively utilized for harvesting microalgae biomass. The present review offers some key suggestions for the development of cost-effective techniques to harvest microalgae biomass.

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