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Selection of a Separation Method Used for Harvesting of Microalgae from Aqueous Solutions

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Harvesting of microalgae from aqueous solutions is still a bottleneck for biotechnologies using microalgae as a source of metabolites. The aim of the article is to provide an overview of four microalgae separation methods and highlight their core advantages, disadvantages, efficiencies and energy and economy requirements for various concentrations of microalgae suspensions and different microalgae strains. These four separation methods are centrifugation, coagulation/flocculation, flotation and membrane technologies. Possible optimizations of the separation methods inducing more effective harvesting are proposed as well. Based on the data included in the article, final conclusions are presented and the separation methods are compared. Flotation harvesting efficiency is more than 75 % when chemicals enhancing the process are applied. Harvesting efficiency of coagulation and flocculation achieves more than 80 %. Harvesting efficiency of membrane technologies and centrifugation is more than 90 %. The energy requirements are in the range of 0.07 to 11.1 kWh m^{-3} of the permeate or microalgal suspension volume and 0.09 to 9.5 kWh kg^{-1} of the dry weight of harvested biomass, depending on the separation method.

1. Introduction

Existing separation methods can be applied to harvest microalgae but they need further research attention and optimization to fulfil the industry demand for high-concentrated biomass slurries at competitive price. Around 20 to 30 % of the total cost of the biomass production is the harvesting of microalgae (Molina Grima et al., 2003). Muradov et al. (2015) reports, 50 % of the total cost of the biodiesel produced by microalgae is attributed to the harvesting process. According to Fasaei et al. (2018), the operational costs of harvesting are in the range of 0.5 to 2 € kg^{-1} of the microalgae biomass and the energy consumption varies between 0.2 to 5 kWh kg^{-1} of the microalgae biomass.

In the following paragraphs, an overview of core parameters of four separation methods is given. The information presented can serve as a starting point before a more in-depth insight into the field of microalgae harvesting. More detailed articles about the topic are, for example, those by Barros et al. (2015), Yin et al. (2020) and Brennan and Owende (2010).

2. Centrifugation

Centrifugation is the most applied separation method for microalgae harvesting due to its high harvesting efficiency (Zhao et al., 2020) which is more than 90 % (Dassey and Theegala, 2013). One of the major disadvantages of this method is its high energy consumption. A different separation method can be applied prior to centrifugation to pre-separate most of the water from the microalgae suspension at lower costs. Centrifugation also exposes microalgae cells to high shear forces which can damage the microalgae cells and cause the leakage of intracellular polymeric substances into the microalgal suspension. Centrifugation used for microalgae harvesting was studied in detail by Belohlav and Jirout (2019).

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3. Flotation

Harvesting by flotation is suitable for small and unicellular microalgae (Pragya et al., 2013). Advantages of flotation separation methods are small space requirements and short operating time (Kurniawati et al., 2014). The disadvantage of dissolved air flotation and dispersed air flotation is the requirement for chemical additions to induce and enhance the entire separation process. These chemicals might contaminate the final microalgae biomass. Harvesting efficiencies are summarized in Table 1. In Table 1, these abbreviations are used: (BBF) buoy-bead flotation, (BDAF) ballasted dissolved air flotation, (DAF) dissolved air flotation, (DiAF) dispersed air flotation, (EF) electrolytic flotation.

Table 1: Flotation efficiencies.

Type	Microalgae	Conditions (mg L^{-1})	pH	Time (s)	Efficiency (%)	Reference
BBF	*Chlorella vulgaris*	microspheres: 700		180	96.16	Xu et al. (2018)
BBF	*Chlorella vulgaris*	microspheres: 700	9	180	98.43	Xu et al. (2018)
BBF	*Scenedesmus obliquus*	microspheres: 550 ferric chloride: 50	7.5	120	75.38	Zou et al. (2018)
BBF	*Scenedesmus obliquus*	microspheres: 550 chitosan: 30	7.5	120	88.52	Zou et al. (2018)
BBF	*Chlorella vulgaris*	microspheres: 550 ferric chloride: 50	7.5	120	83.77	Zou et al. (2018)
BFF	*Chlorella vulgaris*	microspheres: 550 chitosan: 20	7.5	120	92.47	Zou et al.(2018)
BDAF	*Scenedesmus obliquus*	$Al_2(SO_4)_3$: 30 glass beads: 300	5, 7, 9	600	≥99	Ometto et al. (2014)
BDAF	*Chlorella vulgaris*	$Al_2(SO_4)_3$: 6 glass beads: 300	5, 7, 9	600	≥99	Ometto et al.(2014)
BDAF	*Arthrospira maxima*	$Al_2(SO_4)_3$: 77 glass beads: 300	5, 7, 9	600	≥99 ~80 (pH 9)	Ometto et al. (2014)
DAF	*Chlorella vulgaris*	$Al_2(SO_4)_3$: 10	5, 7, 9	600	≥99	Ometto et al.(2014)
DAF	*Arthrospira maxima*	$Al_2(SO_4)_3$: 134	5, 7, 9	600	≥99	Ometto et al. (2014)
DAF	*Scenedesmus obliquus*	$Al_2(SO_4)_3$: 40	5, 7, 9	600	≥99	
DiAF	*Chlorella vulgaris*	saponin: 100	neutral	1,200	22.5	Kurniawati et al.(2014)
DiAF	*Scenedesmus obliquus*	saponin: 100	alkaline	1,200	22.5	Kurniawati et al. (2014)
DiAF	*Chlorella vulgaris*	chitosan: 5 saponin: 20	neutral	1,200	>93	Kurniawati et al. (2014)
DiAF	*Scenedesmus obliquus*	chitosan: 5 saponin: 20	alkaline	1,200	>93	Kurniawati et al.(2014)
EF	*Chlorella vulgaris*	4 V		78	12.2	Wei et. al. (2020)
EF	*Chlorella vulgaris*	5 V		78	39.2	Wei et. al. (2020)
EF	*Chlorella vulgaris*	6 V		78	95.7	Wei et. al. (2020)
EF	*Chlorella vulgaris*	7 V		78	~95.7	Wei et. al. (2020)
EF	*Chlorella vulgaris*	4 V		282	95	Wei et. al. (2020)
EF	*Chlorella vulgaris*	5 V		162	95	Wei et. al. (2020)
EF	*Chlorella vulgaris*	6 V		78	95	Wei et. al. (2020)
EF	*Chlorella vulgaris*	7 V		78	95	Wei et. al. (2020)

4. Coagulation and flocculation

The term coagulation is considered when salts are applied to destabilize the microalgal suspension, whereas the term flocculation is considered when polymers are applied. According to their origin, flocculants are divided into two groups: inorganic and organic. Various coagulants and flocculants for microalgae harvesting have been researched. Some coagulants and flocculants referred in the literature are summarized in Table 2. Coagulation and flocculation (CF) have been applied in wastewater treatment for many years and their properties are well-known and understood. CF applied for wastewater treatment can provide a good model to study interactions between coagulants or flocculants and microalgae cells. Considering the microalgae harvesting, microalgae biomass is the desired product and water is the waste product while the opposite is true for wastewater treatment.

CF provide a simple and fast separation and achieve high harvesting efficiencies. CF also require low energy input, mainly for agitation of microalgae suspensions and coagulants/flocculants. Coagulants and flocculants combined with the agitation induce the destabilization of microalgal suspensions and the formation of coagula or flocs. The disadvantages of CF are the possibility of contamination of final biomass by coagulants or inorganic flocculants. These separation methods are also sensitive to pH and suitable chemicals might be required to set the proper pH for CF. The chemicals can also contaminate final biomass. Organic flocculants are more environmentally friendly and more biodegradable than coagulants and inorganic flocculants. However, organic flocculants have to be charged before flocculation to become cationic and to be able to destabilize microalgal suspensions (Anthony and Sims, 2013).

Table 2: Coagulation and flocculation efficiencies.

Coagulant/Flocculant (C/F)	Dosage of C/F (g L^{-1})	Microalgae	Con. (g L^{-1})	Eff. (%)	Reference
$AlCl_3$	0.5	*Chlorella minutissima*	N/A	>80	Papazi et al. (2010)
$Al_2(SO_4)_3$	0.1	*Scenedesmus.* sp.	0.54	>90	Chen et al. (2013)
$Ca(OH)_2$	0.4	*Scenedesmus* sp.	0.54	90	Chen et al. (2013)
$FeCl_3$	0.15	*Scenedesmus* sp.	0.54	97.32	Chen et al. (2013)
$Fe_2(SO_4)_3$	0.75	*Chlorella minutissima*	N/A	>80	Papazi et al. (2010)
PDADMAC	0.005	*Chlorella vulgaris*	0.3	90	Gerchman et al. (2017)
Zetag 7650 + $Al_2(SO_4)_3$	0.05 $Al_2(SO_4)_3$ 0.005 Zetag	*Tetraselmis suecica*	0.42	>99	Danquah et al. (2009)
cationic guar gum	0.1	*Chlorella* sp.	0.89	92.15	Banerjee et al. (2013)
cationic guar gum	0.04	*Chlorella* sp.	0.78	94.5	Banerjee et al. (2013)
cationic inulin	0.06	*Botryococcus* sp.	N/A	88.61	Rahul et al. (2015)
cationic locust bean gum	0.055	*Chlorella* sp.	1.32	96.98	Kumar et al. (2019)
cationic locust bean gum	0.040	*Muriellopsis* sp.	0.86	96.64	Kumar et al. (2019)
cationic locust bean gum	0.030	*Scenedesmus* sp.	0.79	97.42	Kumar et al. (2019)
cationic nanocellulose	0.07	*Chlorella vulgaris*	0.35	89	Vandamme et al. (2015)
cationic starch	0.04	*Chlorella protothecoides*	0.44	84	Letelier-Gordo et al. (2014)
cationic starch	0.04	*Chlorella protothecoides*	0.56	89	Letelier-Gordo et al. (2014)
cationic starch	0.04	*Chlorella protothecoides*	0.77	90	Letelier-Gordo et al. (2014)
chitosan	0.08	*Scenedesmus* sp.	0.54	>95	Chen et al. (2013)
chitosan	0.005	*Chlorella sorokiniana*	0.48	>99	Xu et al. (2013)
chitosan	0.03	*Nannochloropsis gaditana*	N/A	>80	Şirin et al. (2013)
chitosan	0.04	*Phaeodactylum tricornutum*	N/A	>90	Şirin et al. (2012)
Moringa oleifera seed flour	1	*Chlorella vulgaris*	N/A	89	Teixeira et al. (2012)
poly γ-glutamic acid	0.02	*Chlorella vulgaris*	1.2	>80	Zhang and Hu (2012)
poly γ-glutamic acid	0.02	*Chlorella protothecoides*	1.2	~90	Zhang and Hu (2012)
poly γ-glutamic acid	N/A	*Nannochloropsis oculata*	N/A	>90	Zhang and Hu (2012)

Note: Con.- concentration of the microalgal suspension, Eff.- harvesting efficiency.

5. Membrane technologies

Membrane technologies (MT) are less sensitive to pH than coagulation or flocculation and no pH adjustments of microalgae suspensions are required. Separation by MT is not induced by chemicals and produced biomass is free of any chemical contamination. MT are gentler to microalgae cells in comparison with centrifugation. MT are mainly affected by fouling. Fouling must be removed regularly and membranes destroyed by fouling must be replaced. In general, higher fluxes induce higher fouling rates. A crucial parameter is critical flux above which fouling occurs. Below the value of critical flux, flow through the membrane remains constant in time. High pressure drops were applied to overcome fouling and membranes were rapidly destroyed by fouling. Nowadays, MT are generally applied in large scales due to new materials, enhanced manufacturing technologies and decreased costs of membranes.

Considering the microalgae harvesting, fouling is caused by the accumulation of intracellular polymeric substances produced by microalgae cells and extracellular polymeric substances present in microalgal suspensions. The fouling formation might be influenced by the membrane charge (Rossi et al., 2004). Neutral polyacrylonitrile membranes are more fouling resistant than charged polyacrylonitrile membranes (Rossi et al., 2004). The appropriate choice of membrane materials also influences the formation of fouling. For example, polyvinylidene fluoride (PVDF) membranes induce the fouling formation (Zhao et al., 2020).

One of the options to reduce fouling might be the application of vibrated modules, submerged aerated systems, stirrers or agitators. These solutions cause shear stress along the surface of membranes. As a consequence of the shear stress, fouling is removed from the surfaces. In general, the shear stress along the membranes can be induced hydraulically, pneumatically or mechanically. Another approach mitigating or eliminating fouling is to apply relaxation or backwashing.

For adsorption-based fouling, chemical cleaning is needed (Rossi et al., 2004). The origin of the adsorption-based fouling is organic or inorganic. The complete removing of organic adsorption-based fouling is difficult because it is probably caused by the connection of organics with a membrane polymeric matrix via cationic bonds (Bilad et al., 2013). The inorganic adsorption-based fouling can be eliminated from the membranes surface by the application of citric acid but this approach still needs further research (Bilad et al., 2013).

The achieved harvesting recovery might be expressed by the concentration factor (COF), the harvesting efficiency (HE) or by the retentate concentration (REC). The following values of harvesting recovery were achieved: HE of 90 to 95 % for hollow fiber membranes and *Nannochloropsis* sp. (Bhave et al., 2012), REC of 46.6 g L^{-1} for tangential flow filtration, *Tetraselmis suecica*, initial concentration of 0.6 g L^{-1} (Danquah et al., 2009), COF in the range of 50 to 245 for pressure filters (Molina Grima et al., 2003), COF in the range of 2 to 180 for vacuum filters (Molina Grima et al., 2003), HE of 99 to 100 % for submerged filtration, PVDF, and *Chlorella vulgaris* (Bilad et al., 2012) and HE of 98 to 99 % for submerged filtration, PVDF, and *Phaeodactylum tricornutum* (Bilad et al., 2012).

6. Economy and energy requirements

The energy evaluation of selected separation methods is summarized in Table 3. In Table 3, the following abbreviations are used: (C) coagulation, (CEN) centrifugation, (EC) electro-coagulation-flocclation, (FIL) filtration, (FLOT) flotation, (HW) hollow fibers, (MMV) magnetically induced membrane vibration, (MT) membrane technologies, (PVDF) polyvinylidene fluoride, (TFF) tangential flow filtration, (TM) tubular membranes.

Table 3: Energy evaluation.

Method	Specifications	Microalgae	Energy requirements	Reference
CEN		N/A	8 kWh m^{-3}	Danquah et al. (2009)
C	Zetag 7650 + Al$_2$(SO$_4$)$_3$	*Tetraselmis suecica*	0.07 kWh m^{-3}	Danquah et al. (2009)
ECF		*Chlorella vulgaris*	1.3 to 9.5 kWh kg^{-1}	Vandamme et al. (2011)
ECF		*Phaeodactylum tricornutum*	0.2 to 0.4 kWh kg^{-1}	Vandamme et al. (2011)
FIL	pressure filters	N/A	0.88 kWh m^{-3}	Danquah et al. (2009)
FIL	vacuum filters	N/A	5.9 kWh m^{-3}	Danquah et al. (2009)
FLOT	electrolytic, 4 V	*Chlorella vulgaris*	0.1 kWh kg^{-1}	Zou et al. (2018)
FLOT	electrolytic, 6 V	*Chlorella vulgaris*	0.09 kWh kg^{-1}	Wei et al. (2018)
FLOT	electrolytic, 7 V	*Chlorella vulgaris*	0.15 kWh kg^{-1}	Wei et al. (2020)
MT	PVDF, MMV, 4 modules	*Dictyosphaerium* sp.	2.9 kWh m^{-3}	Zhao et al. (2020)
MT	PVDF, MMV, 1 module	*Dictyosphaerium* sp.	11.1 kWh m^{-3}	Zhao et al. (2020)
MT	HF plus TM	*Nannochloropsis* sp.	0.3 to 0.7 kWh m^{-3}	Drexler and Yeh (2014)
MT+CEN	PVDF membrane	*Chlorella vulgaris*	0.84 kWh m^{-3}	Bilad et al. (2012)
MT+CEN	MMV	*Chlorella vulgaris*	0.77 kWh m^{-3}	Bilad et al. (2013)
TFF		*Tetraselmis suecica*	2.06 kWh m^{-3}	Danquah et al. (2009)

In general, the estimation of operational costs is scale-dependent and the real cost can be significantly different for pilot- and full-scale systems. Based on the data by Ometto et al. (2014), the following operational costs were estimated. For dissolved air flotation, the operational costs are: 0.2 £ m^{-3} d^{-1} (*Scenedesmus obliquus*), 0.06 £ m^{-3} d^{-1} (*Chlorella vulgaris*), 0.7 £ m^{-3} d^{-1} (*Arthrospira maxima*). For ballasted dissolved air flotation, the operational costs are 0.2 £ m^{-3} d^{-1} (*Scenedesmus obliquus*), 0.07 £ m^{-3} d^{-1} (*Chlorella vulgaris*), 0.4 £ m^{-3} d^{-1} (*Arthrospira maxima*). The following items, energy, coagulant aluminum sulphate and beads, were considered for the estimation of the operational costs.

7. Conclusions

Selection of a suitable separation method is complex and depends on many factors. If separated microalgae are unicellular and small, flotation may be applied. Flotation is usually induced by the addition of chemicals. In this case, the final biomass may be contaminated by these chemicals. Flotation harvesting efficiency is more than 75 % when chemicals enhancing the process are applied. Harvesting efficiency of coagulation and flocculation (CF) achieves more than 80 % but final biomass is contaminated when inorganic flocculants and coagulants are applied. Organic flocculants are more biodegradable than inorganic flocculants and coagulants but they have to be charged before they are applied. CF are also sensitive to pH. Membrane technologies and centrifugation are not sensitive to pH and are not induced by chemicals. Harvesting efficiency of membrane technologies and centrifugation is more than 90 %. Membrane technologies are gentler to microalgae cells in comparison with centrifugation. Energy consumption of centrifugation is high in comparison with CF. Membrane technologies are prone to fouling. Neutral polyacrylonitrile membranes are more fouling resistant than charged polyacrylonitrile membranes and polyvinylidene fluoride membranes.

The energy requirements are in the range of 0.07 to 11.1 kWh m^{-3} of permeate or microalgal suspension volume, and 0.09 to 9.5 kWh kg^{-1} of dry weight of harvested biomass, depending on the separation method.

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