

Electro-separation of microalgal culture from wastewater

Rafiee, Poorya; Tong, Yen Wah; Hosseini, Maryam; Ebrahimi, Sirous

DOI

[10.1016/j.bcab.2019.101402](https://doi.org/10.1016/j.bcab.2019.101402)

Publication date

2019

Document Version

Accepted author manuscript

Published in

Biocatalysis and Agricultural Biotechnology

Citation (APA)

Rafiee, P., Tong, Y. W., Hosseini, M., & Ebrahimi, S. (2019). Electro-separation of microalgal culture from wastewater. *Biocatalysis and Agricultural Biotechnology*, 22, [101402].
<https://doi.org/10.1016/j.bcab.2019.101402>

Important note

To cite this publication, please use the final published version (if applicable).
Please check the document version above.

Copyright

Other than for strictly personal use, it is not permitted to download, forward or distribute the text or part of it, without the consent of the author(s) and/or copyright holder(s), unless the work is under an open content license such as Creative Commons.

Takedown policy

Please contact us and provide details if you believe this document breaches copyrights.
We will remove access to the work immediately and investigate your claim.

Electro-separation of microalgal culture from wastewater

Poorya Rafiee¹, Yen Wah Tong^{2*}, Maryam Hosseini³, Sirous Ebrahimi^{1,4*}

¹Biotechnology Research Centre, Faculty of Chemical Engineering, Sahand University of Technology, Tabriz, Iran

²Department of Chemical Engineering, National University of Singapore, Singapore

³Faculty of Chemical Engineering, Azarbaijan Shahid Madani University, Tabriz, Iran

⁴Department of Biotechnology, Delft University of Technology, Delft, Netherlands

(s.ebrahimi@tudelft.nl)

Abstract

For further applications of microalgae such as bio-products, microalgal harvesting from its culture medium (e.g. wastewater) must be studied. This becomes more essential when investigating whether or not cells can stay viable to be recycled into the system. Microalgae culture, wastewater, and a mixture of both were separately electrocoagulated at wastewater Chemical Oxygen Demand ranging 66-2700 mg.l⁻¹ and biomass dry weights between 1-8 g.l⁻¹. The mixed culture contained species of *C. Vulgaris*, *S. Obliquus*, *B. Braunii*, *B. Sudeticus*, and *A. Falcatus*, since mixed culture technique can reduce the expenses in industrial scales by eliminating the costly sterilization strategies necessary to avoid contamination. The mixed samples were successfully separated with the efficiencies between 44-87% and 70-80% at different Chemical Oxygen Demand and biomass dry weights, respectively.

In addition, it was shown that growth elements of carbon and nitrogen, although at lower rates, were consumed confirming the viability of the cells after electrocoagulation. The consumption rates for electrocoagulated samples were smaller than non-electrocoagulated samples only by 16, 12, and 31% in carbon, nitrate and ammonium concentrations,

24 respectively. According to the obtained results electrical separation of microalgae could
25 effectively harvest microalgae from wastewater without affecting the viability of the
26 biomass.

27 **Key words:** Electrocoagulation, Harvest, Microalgae, Mixed culture, Wastewater

28 **1. Introduction**

29 Renewable energy and treatment of wastewater are two topics of immense importance in the
30 current century. In one hand, the concerns over fossil fuels consumption grow every day, and
31 renewable biofuels seem to be a promising substitute. However, oil crops and waste oil
32 cannot provide the current demand for fuel, and microalgae can be a significant aid as
33 feedstock for biofuel production (Chisti 2007, Christenson and Sims 2011). Microalgae can
34 provide human with a more promising source for biofuel, bio-methane, and many other
35 currently oil-based materials like bio-plastic and fertilizers, needless to mention the cosmetic,
36 medical, and food industries that can benefit from microalgae bioproducts (Chiellini, Cinelli
37 et al. 2008, Roeselers, Van Loosdrecht et al. 2008, Barros, Gonçalves et al. 2015).

38 On the other hand, the shortage of fresh water has led to universal attempts to find sustainable
39 water management strategies. Bio-treatment using microalgae has received attention since the
40 removal of the nutrients is less expensive and more environmental friendly compared to
41 conventional chemical methods (Hoffmann 1998, Christenson and Sims 2011, Abdel-Raouf,
42 Al-Homaidan et al. 2012)

43 As a result, it would be a promising idea to use microalgae to treat the wastewater of its
44 nutrients and generate biofuel and other bioproducts. Nevertheless, the most costly stage of
45 microalgae-based technology would be its harvesting from the liquid phase reaching to 20-
46 60% of the total cost (Sander and Murthy 2010, Nguyen, Le et al. 2019). Many strategies,

47 including centrifugation, coagulation, ultrasonic, pH change, filtration, etc., have been
48 applied to separate the microalgae from the liquid phase (Fayad, Yehya et al. 2017, Nguyen,
49 Le et al. 2019). Electrocoagulation (EC) is one of the most widely applied strategies to
50 harvest microalgae (Gao, Yang et al. 2010, Uduman, Qi et al. 2010) and to treat different
51 wastewater (Gao, Yang et al. 2010). Researches have reported up to 95% of the microalgae
52 removal by electrocoagulation (Uduman, Qi et al. 2010). Furthermore, electrocoagulation has
53 been successfully applied to treat various wastewater with perfect efficiencies (Sahu,
54 Mazumdar et al. 2014). In these studies, microalgae was separated mainly from growth
55 medium dissolved in water, and other separation mediums like wastewater have been rarely
56 discussed (Udom, Zaribaf et al. 2013). In one of the very rare studies on algae harvesting
57 from wastewater, the chemical coagulation was applied as the harvesting technique (Udom,
58 Zaribaf et al. 2013). In addition, one major bottleneck in microalgae application is the low
59 productivity of the culture in terms of product formation and biomass. Besides, many
60 microalgal products are secondary metabolites which are produced at the cost of growth
61 limitation. If these metabolites can be removed continuously from the cells, the biomass can
62 be re-used to produce the high-value compounds (Hejazi and Wijffels 2004). Therefore, the
63 viability of cells at different stages of industrial operations can be very important. This must
64 be added to the fact that the viable biomass can always be recycled and used as inoculum for
65 the next growth generation. However, there have rarely been studies to investigate the effect
66 of harvesting techniques on the cell viabilities. In one study, the chemical coagulation seems
67 to have had no effect on the cells viability (Papazi, Makridis et al. 2010), although no
68 investigation has been found to inspect electrocoagulation for similar results.

69 The harvesting of a mixed culture of microalgae from wastewater using electrocoagulation
70 has been rarely focused in literature. In addition, there has been no study to inspect the
71 viability of microalgal cells after electrocoagulation. Therefore, this study aims to investigate

72 the efficiency of EC for harvesting a mixed culture of microalgae from an industrial
73 wastewater medium. In addition, the effect of EC on the microalgal growth was investigated
74 through a series of viability experiments.

75

76 **2. Materials and Methods**

77 ***2.1. Microalgae medium and cultivation***

78 A mixed culture containing *C. Vulgaris*, *S. Obliquus*, *B. Braunii*, *B. Sudeticus*, and *A.*
79 *Falcatus* was prepared and inoculated into a 4-liter cylindrical photobioreactor (PBR) filled
80 with autoclaved 3N-BBM+V (modified Bold Basal Medium with 3-fold Nitrogen and
81 Vitamins) upto 3.5 liters. The 3N-BBM+V medium consisted of macro-nutrients: 0.75 g
82 NaNO₃, 0.025 g CaCl₂.2H₂O, 0.075 g MgSO₄.7H₂O, 0.075 g K₂HPO₄.3H₂O, 0.175 g
83 KH₂PO₄, 0.025 g NaCl and micro-nutrients: 4.5 mg Na₂EDTA, 0.582 mg FeCl₃.6H₂O, 0.246
84 mg MnCl₂.4H₂O, 0.03 mg ZnCl₂, 0.012 mg CoCl₂.6H₂O, 0.024 mg Na₂MoO₄.2H₂O, 1.2 mg
85 Thiamine hydrochloride as well as 0.01 mg Cyanocobalamin, per liter of DI water (Guo and
86 Tong 2014). All chemicals were purchased from Sigma–Aldrich (Singapore). The PBR was
87 illuminated using four 13W 6700K florescent lamps and aerated with a mixed flow of air and
88 CO₂ (1.75 LPM air and its 5% CO₂ flow) with an aeration rate of 0.5 vvm. In addition to the
89 air flow, the content of the culture flask was magnetically stirred to provide good mixing
90 under room temperature. When a dry weight (DW) of 2 g.l⁻¹ was obtained, the algal culture
91 was used for the subsequent electrocoagulation. The required microalgae were diluted or
92 concentrated depending on the desired DW values using distilled water or centrifugation,
93 respectively.

94 ***2.2. Wastewater***

95 A food industry wastewater was used with an initial Chemical Oxygen Demand (COD) of
96 20000 mg.l⁻¹. This concentration was later diluted to obtain the desired COD values for the
97 harvesting experiments using distilled water. Although the set-up was not aimed to perform
98 in a sterile condition, the wastewater was autoclaved in order to make sure that no other
99 micro-organism existed at the start of the experiment.

100 ***2.3.Electrocoagulation cell***

101 The EC cell consisted of a 250-mililiter beaker equipped with Aluminum electrodes
102 connected to a DC Power supply. The sample volume was 200 milliliters, and EC time was 5
103 min. Each sample was left to settle for 5 min before sampling. The whole sample, without
104 modification, was later left for further microalgal growth. The current density for all
105 experiments was 250 A.m⁻², and the interelectrode distance was 1cm. The EC experiments
106 were performed for microalgae (MIC), wastewater (WW), and the mixture of both (MWW).
107 In case of microalgae and wastewater mix (MWW) the ratio was 1:9, respectively. In pure
108 microalgae and pure wastewater experiments, the distilled water was replaced with similar
109 ratios. Each EC experiment was performed in duplicates to ensure the reproducibility of the
110 results.

111 ***2.4.Analytical Methods***

112 For each set of harvesting experiments, the Chemical Oxygen Demand COD was measured
113 before and after the electrocoagulation was run. The COD was measured using dichromate
114 according to standard methods (Baird, Bridgewater et al. 2012). All tests were performed
115 three times and an average value was reported.

116 The dry weight (DW) was reported by measuring the difference between the weights of a
117 dried filter before and after addition of 5 milliliters of sample. To dry the filter before and

118 after microalgae addition, it was kept in an oven at 105 °C for a day and then cooled in a
119 desiccator (Baird, Bridgewater et al. 2012).

120 For determining the dissolved nitrogen, the ammonium and nitrate tests were measured by
121 phenate and spectrophotometric methods, respectively (Baird, Bridgewater et al. 2012). All
122 tests were performed three times and an average value was reported.

123 **3. Results**

124 ***3.1. The effect of wastewater concentration***

125 The results of COD removal by electrocoagulation based on varying initial wastewater COD
126 concentrations for WW and MWW are depicted in Figure 1. In WW and MWW experiments,
127 with higher COD values the removal efficiency started to decrease. In WW experiments, the
128 recovery values for the CODs of 82, 266, 543, 827, and 2748 mg.l⁻¹ were 100, 88, 87, 67, and
129 39%, respectively.

130 In addition, for MWW experiments, the recovery values were 87, 79, 77, 50, and 44%,
131 respectively. To ensure consistency of the resulted trend for removal efficiency through COD
132 results, Optical Density (OD) of the samples before and after the EC run were also measured
133 and recovery was calculated in terms of OD values (Zongo, Maiga et al. 2009, De Godos,
134 Guzman et al. 2011) (See supplementary file).

135 ***3.2. The effect of microalgal concentration***

136 When the initial dry weight of microalgae was changed, the recovery rate maintained at high
137 values. These results have been illustrated in the Figure 2. The initial wastewater COD was
138 measured to be between 193 and 263 mg.l⁻¹ and after the EC run, the COD removal for WW
139 varied between 74 and 92% (not shown in the graph). For microalgae, the initial dry weights

140 were 1, 2, 4, and 8 g.l⁻¹. The removal efficiencies for MIC were 96, 89, 76, and 90% for 1, 2,
141 4, and 8 g.l⁻¹.

142 The MWW only had a slight change, since no big drop in removal of microalgae culture had
143 occurred. Except for microalgal cell density of 1 g.l⁻¹, where the removal was 68% the three
144 other cell concentrations were measured to be 80%. Here, too, OD of the samples were also
145 measured and patterns were compared with the data from COD analysis (refer to
146 supplementary data).

147 ***3.3. The viability tests***

148 Two separate sets of microalgae samples, electrocoagulated (EC) and non-electrocoagulated
149 (non-EC), were studied for the consumption of important nutrients for a 7-day period. All
150 growth conditions were as described above. To study the nitrogen consumption, ammonium
151 and nitrate tests were performed on daily basis, and the COD test was applied to study the
152 consumption of carbonic compounds. The results of COD, nitrate, and ammonium tests can
153 be found in figures 3, 4, and 5, respectively. Figure 3 shows that carbon sources in the non-
154 EC sample were consumed at a rate of 17.72 mg.l⁻¹.day⁻¹ while it was consumed at the rate of
155 14.89 mg.l⁻¹.day⁻¹ in EC sample. In other words, the COD was removed at least 60% in both
156 EC and non-EC samples.

157 On the other hand, the consumption of nitrate was measured to investigate consumption of
158 the nitrogen source for growth. The results are depicted in Figure 4. The nitrate consumption
159 rates were measured to be 2.52 and 2.21 mg.l⁻¹.day⁻¹ for non-EC and EC samples,
160 respectively. Based on the initial nitrogen concentrations, dissolved N was removed by 35-
161 40% from the mediums.

162 Since ammonium is a different nitrogen source present in wastewater, its consumption rate
163 was also monitored. Figure 5 shows the ammonium consumption within a 7-day period.
164 While ammonium consumption rate is $0.638 \text{ mg.l}^{-1}.\text{day}^{-1}$ for non-EC sample, it was 0.440
165 $\text{mg.l}^{-1}.\text{day}^{-1}$ for the EC sample. Results can be interpreted as the removal of 15-21% of
166 ammonium from the mediums.

167 **4. Discussion**

168 Although electrocoagulation has been applied for years even at industrial scale for
169 wastewater treatment and recently for biomass separation, the involved mechanisms have
170 been seriously argued. The current theory states that EC involves several sequent stages
171 (Moreno-Casillas, Cocke et al. 2007): first, the metal ions are generated. Then, the metal ions
172 hydrolysis occurs and metal hydroxides and polyhydroxides form. Water is simultaneously
173 electrolyzed producing small bubbles of oxygen at the anode and hydrogen at the cathode.
174 Next, the particles are destabilized, the emulsions are broken and then come together to
175 aggregate and form flocs. Finally, chemical reactions and precipitation can occur including
176 hydroxyl ions forming precipitate with particles. These mechanisms, though affected by
177 biomass/wastewater concentration, individually or collectively provided both colloidal
178 (wastewater) and biological (microalgae) separations.

179 ***4.1. The effect of wastewater concentration***

180 At constant conditions like current density and time, the falling trend of removal efficiency
181 with higher initial concentration was observed which is in agreement with the results in other
182 studies (Aoudj, Khelifa et al. 2010). The removal efficiency is quite comparable to many
183 studies in the literature (Olguín 2012, Fernandes, Pacheco et al. 2015), although the
184 efficiencies often vary widely from one study to another, since the exact composition of
185 wastewater complicates the comparison. In one study, for example, on the pulp and paper

186 industry effluent, with an initial COD of 620 mg.l⁻¹, the COD removal efficiency at the same
187 current density was reported to be around 50% (Sridhar, Sivakumar et al. 2011). Apart from
188 the chemical composition, the 3-centimeter interelectrode distance has decreased the
189 efficiency compared to the current study value where the electrode gap was 1 cm. With
190 increasing the distance, a decrease in the amount of anode dissolution will occur, and the ions
191 need to transfer a longer distance for interaction to form flocs. Thus, with less flocs
192 formation, COD removal will decrease (Khandegar and Saroha 2012). One study used natural
193 flocculants of Ecotan and Tanfloc to harvest microalgal culture from a pre-treated urban
194 wastewater set-up. The optimal biomass recovery was reported to be 92 and 90% for Ecotan
195 and Tanfloc, respectively. A dose amounts of 10 and 50 mg.l⁻¹ were, respectively, used for
196 these two natural flocculants (Gutiérrez, Passos et al. 2015). As that study reports, the COD
197 of the set-up influent was 250 mg.l⁻¹ on average (Passos, Solé et al. 2013, Gutiérrez, Passos et
198 al. 2015), which is quite comparable with the WW and MWW results in this study, especially
199 since no optimization was aimed and practiced here. Yet, in another study on harvesting
200 bacterial and microalgal cultures from a piggery wastewater, seven different coagulants and
201 flocculants were tested including two conventional coagulants of FeCl₃ and Fe₂(SO₄)₃, and
202 five commercial polymeric flocculants such as Chitosan. The researchers tested different
203 doses of these chemicals. The best removal efficiencies were generally for FeCl₃ and
204 Fe₂(SO₄)₃. Efficiencies higher than 90% all occurred for high doses of coagulants/flocculants,
205 between 150-250 mg.l⁻¹. The wastewater tested here, too, was far less (=202 mg.l⁻¹) than the
206 maximum amount of COD that microalgal biomass was introduced to in the current study
207 (De Godos, Guzman et al. 2011).

208 The decrease in COD removal can be associated to the present compounds. In an EC process,
209 “the COD may increase” due to the reaction of some compounds such as acids with the metal
210 ions to form soluble products which remain in the solution. On the other hand, soluble and

211 miscible compounds that do not react with metal ion can completely “keep the COD
212 unchanged”. However, organic salts can form insoluble compounds with metal hydroxide
213 which leads to “partial removal of the COD” from the medium. Since these compounds
214 usually consist the main body of municipal and industrial wastewater (Moreno-Casillas,
215 Cocke et al. 2007) with higher concentration of such compounds at more concentrated
216 wastewater, less COD can be removed from the medium accordingly.

217 ***4.2. The effect of microalgal concentration***

218 Except for 8 g.l⁻¹ sudden increase, the falling pattern was expected due to increase in cell
219 density. This falling pattern can be associated with the adequacy of metal ions to remove the
220 excessive algae along with the decrease in the reaction rate in EC process. (Gao, Yang et al.
221 2010). It was already reported that there is no linear correlation between the concentrations of
222 microalgae and the removal efficiency (Tenney, Echelberger et al. 1969, De Godos, Guzman
223 et al. 2011). However, the non-linear correlation between the cell concentration and removal
224 efficiency may be attributed to algogenic organic matter (AOM). The negative effect of AOM
225 on coagulation has been addressed before (Zhuang, Wu et al. 2016). On the other hand, the
226 algae cell itself, in the category of suspended solid particles, can be removed with high
227 efficiencies due to the in-situ-generated coagulants (Moreno-Casillas, Cocke et al. 2007).
228 The 8-gram microalgal sample was concentrated using centrifugation of four similar 2-gram
229 samples in a way that the growth culture medium was removed after being centrifuged and
230 replaced with and mixed in a fresh growth medium together. Consequently, the AOM in the
231 four samples had been removed and therefore its negative effect on the coagulation process
232 had been mitigated.

233 The results obtained from this study are quite comparable with other studies, given the fact
234 that the cell density in those studies was either much lower than present research (<1 mg.l⁻¹)
235 (Vandamme, Pontes et al. 2011) or reported in cell count (Gao, Yang et al. 2010, Wong, Ho

236 et al. 2017). In one of the rare studies on harvesting microalgae from wastewater, six
237 chemicals were used to harvest *Chlorella* at both wild and lab-cultured species from
238 wastewater. These chemicals included two reagents of alum and ferric chloride, cationic
239 polymer, anionic polymer, and natural polymers. The best removal efficiency was achieved
240 by ferric chloride and alum in which microalgal culture could be harvested by 93 and 91%
241 efficiency, respectively. It is worth mentioning that to obtain these efficiencies, 122 mg.l⁻¹ of
242 ferric chloride and 140 mg.l⁻¹ of alum were used (Udom, Zaribaf et al. 2013). These amounts
243 of additive chloride and sulfate ions yet again bring in the conventional debate over the
244 benefits of electrocoagulation over coagulation. In addition, in the noted study, no separate
245 data were provided on the flocculation of the wastewater itself especially because the carbon
246 source was provided through CO₂ flow. In another study the effect of biomass concentration
247 on the removal efficiency was tested. In this study, two commercial flocculants, namely
248 Drewfloc-447 and Chemifloc CV-300, were applied. For both flocculants, almost nothing
249 happened when the concentration of biomass doubled. On the other hand, when the initial
250 concentration of biomass was halved, the removal efficiency rose by 50% in Drewfloc-447
251 case and fell by 12% (De Godos, Guzman et al. 2011). Although, the mixed rising and falling
252 patterns associated with concentration change have been also observed in the current study,
253 these patterns are more moderate. This difference seems to be the result of a mixed culture,
254 since in mentioned work, only a pure culture of *C. Sorokiniana* was investigated.
255 Results of harvesting at both different biomass and wastewater concentrations show that
256 although biological features can help decrease or increase the efficiency, in terms of
257 coagulation both colloidal and biological particles act similarly. These results are perfectly in
258 accordance with previous studies (Pieterse and Cloot 1997).
259 For the MWW values, the measures were more uniform. MWW values for recovery
260 efficiency for all the dry weights, except for 1 g.l⁻¹, were measured to be approximately 80%.

261 **4.3. The viability tests**

262 It must be noted that small difference in the initial values of COD in both samples can be due
263 to the COD reduction that normally occurs due to electro-oxidation, electrocoagulation, etc.
264 (Moreno-Casillas, Cocke et al. 2007).

265 In one study on the growth of a *Chlorella* on wastewater, the COD was removed by 90% over
266 the course of 14 days. In addition, 90% of the total nitrogen and 93% of ammonium were
267 removed at the same interval (Li, Chen et al. 2011). Since the cell concentration in both
268 studies were almost similar, the COD removal can be attributed to the difference between the
269 microalgal species. While *C. Vulgaris* is only one of the microalgae species present in the
270 current study, in the mentioned research the microalgal medium mainly contained *Chlorella*
271 which is known to be a very good mixotrophic, meaning that it can feed both on CO₂ and
272 organic sources (Martínez, Camacho et al. 1997). As a result, the cell dry weight in that study
273 has multiplied by a factor of 12 from 0.1 to 1.2 g.l⁻¹ within the experiment time (Li, Chen et
274 al. 2011).

275 In another study, in which cultivation of bacterial and microalgal biomass was investigated
276 on a piggery wastewater, the COD was removed by a range between 49 and 78% for
277 *Chlorella* consortium, *S. obliquus*, *Chlorococcum sp.*, and *C. sorokiniana* species. In
278 addition, the consumption of N-NH₄⁺ was also investigated. The N-NH₄⁺ removal was
279 reported to be between 77 and 81% (De Godos, Guzman et al. 2011).

280 These data from COD, nitrate and ammonium consumption rates collectively states that
281 although the consumption rates slightly differ from each other, yet confirm the consumption
282 of carbon and nitrogen sources meaning that a great number of microalgae are viable and
283 growing. In addition, the slight reduction in consumption rates of these sources may indicate
284 a part of biomass culture has been inactivated due to oxidative stress, production of harmful

285 oxidants, and/or irreversible membrane permeabilization caused by EC (Wei, Elektorowicz et
286 al. 2011). The confirmation of biomass viability in the current study is in agreement with
287 previous work on bacteria (Wei, Elektorowicz et al. 2011). Studies show that other methods
288 of biomass harvesting can lead to similar conclusions with cell viability. In one case,
289 researchers used three methods of centrifugation to harvest 9 different species of microalgae.
290 The most vulnerable species in that study suffered only from 12% of biomass viability
291 (Heasman, Diemar et al. 2000).

292 **5. Conclusion**

293 In this study, a mixed microalgal culture was successfully harvested from a wastewater
294 medium with high recovery efficiency. These recovery efficiencies continued to maintain at
295 high rates even at high concentrations of wastewater and microalgae. The results showed that
296 the growth nutrients represented by COD, ammonium and nitrate were all consumed,
297 although slightly smaller than non-electrocoagulated samples, in the course of a 7-day re-
298 culturing after the electrocoagulation. These results confirm that cells were viable after the
299 harvesting process. Therefore, electrocoagulation can be used to harvest microalgae from
300 wastewater without the risk of disrupting of the microalgal cells.

301 **6. Conflict of Interest**

302 This research did not receive any specific grant from funding agencies in the public,
303 commercial, or not-for-profit sectors.

304 **7. References**

305 Abdel-Raouf, N., A. Al-Homaidan and I. Ibraheem (2012). "Microalgae and wastewater
306 treatment." *Saudi journal of biological sciences* 19(3): 257-275.
307 Aoudj, S., A. Khelifa, N. Drouiche, M. Hecini and H. Hamitouche (2010).
308 "Electrocoagulation process applied to wastewater containing dyes from textile industry."
309 *Chemical Engineering and Processing: Process Intensification* 49(11): 1176-1182.

310 Baird, R. B., L. Bridgewater, L. S. Clesceri, A. D. Eaton and E. W. Rice (2012). Standard
311 methods for the examination of water and wastewater, American public health association.

312 Barros, A. I., A. L. Gonçalves, M. Simões and J. C. Pires (2015). "Harvesting techniques
313 applied to microalgae: a review." *Renewable and Sustainable Energy Reviews* 41: 1489-
314 1500.

315 Chiellini, E., P. Cinelli, V. I. Ilieva and M. Martera (2008). "Biodegradable thermoplastic
316 composites based on polyvinyl alcohol and algae." *Biomacromolecules* 9(3): 1007-1013.

317 Chisti, Y. (2007). "Biodiesel from microalgae." *Biotechnology advances* 25(3): 294-306.

318 Christenson, L. and R. Sims (2011). "Production and harvesting of microalgae for wastewater
319 treatment, biofuels, and bioproducts." *Biotechnology advances* 29(6): 686-702.

320 De Godos, I., H. O. Guzman, R. Soto, P. A. García-Encina, E. Becares, R. Muñoz and V. A.
321 Vargas (2011). "Coagulation/flocculation-based removal of algal–bacterial biomass from
322 piggery wastewater treatment." *Bioresource technology* 102(2): 923-927.

323 Fayad, N., T. Yehya, F. Audonnet and C. Vial (2017). "Harvesting of microalgae *Chlorella*
324 *vulgaris* using electro-coagulation-flocculation in the batch mode." *Algal Research* 25: 1-11.

325 Fernandes, A., M. Pacheco, L. Ciríaco and A. Lopes (2015). "Review on the electrochemical
326 processes for the treatment of sanitary landfill leachates: present and future." *Applied*
327 *Catalysis B: Environmental* 176: 183-200.

328 Gao, S., J. Yang, J. Tian, F. Ma, G. Tu and M. Du (2010). "Electro-coagulation–flotation
329 process for algae removal." *Journal of Hazardous Materials* 177(1-3): 336-343.

330 Guo, Z. and Y. W. Tong (2014). "The interactions between *Chlorella vulgaris* and algal
331 symbiotic bacteria under photoautotrophic and photoheterotrophic conditions." *Journal of*
332 *applied phycology* 26(3): 1483-1492.

333 Gutiérrez, R., F. Passos, I. Ferrer, E. Uggetti and J. García (2015). "Harvesting microalgae
334 from wastewater treatment systems with natural flocculants: effect on biomass settling and
335 biogas production." *Algal research* 9: 204-211.

336 Heasman, M., J. Diemar, W. O'connor, T. Sushames and L. Foulkes (2000). "Development of
337 extended shelf-life microalgae concentrate diets harvested by centrifugation for bivalve
338 molluscs—a summary." *Aquaculture Research* 31(8-9): 637-659.

339 Hejazi, M. A. and R. H. Wijffels (2004). "Milking of microalgae." *TRENDS in*
340 *Biotechnology* 22(4): 189-194.

341 Hoffmann, J. P. (1998). "Wastewater treatment with suspended and nonsuspended algae."
342 *Journal of Phycology* 34(5): 757-763.

343 Khandegar, V. and A. K. Saroha (2012). "Electrochemical treatment of distillery spent wash
344 using aluminum and iron electrodes." *Chinese Journal of Chemical Engineering* 20(3): 439-
345 443.

346 Li, Y., Y.-F. Chen, P. Chen, M. Min, W. Zhou, B. Martinez, J. Zhu and R. Ruan (2011).
347 "Characterization of a microalga *Chlorella* sp. well adapted to highly concentrated municipal
348 wastewater for nutrient removal and biodiesel production." *Bioresource technology* 102(8):
349 5138-5144.

350 Martínez, M. E., F. Camacho, J. Jiménez and J. Espinola (1997). "Influence of light intensity
351 on the kinetic and yield parameters of *Chlorella pyrenoidosa* mixotrophic growth." *Process*
352 *Biochemistry* 32(2): 93-98.

353 Moreno-Casillas, H. A., D. L. Cocke, J. A. Gomes, P. Morkovsky, J. Parga and E. Peterson
354 (2007). "Electrocoagulation mechanism for COD removal." *Separation and purification*
355 *Technology* 56(2): 204-211.

356 Nguyen, T. D. P., T. V. A. Le, P. L. Show, T. T. Nguyen, M. H. Tran, T. N. T. Tran and S. Y.
357 Lee (2019). "Bioflocculation formation of microalgae-bacteria in enhancing microalgae

358 harvesting and nutrient removal from wastewater effluent." *Bioresource technology* 272: 34-
359 39.

360 Olguín, E. J. (2012). "Dual purpose microalgae–bacteria-based systems that treat wastewater
361 and produce biodiesel and chemical products within a Biorefinery." *Biotechnology advances*
362 30(5): 1031-1046.

363 Papazi, A., P. Makridis and P. Divanach (2010). "Harvesting *Chlorella minutissima* using cell
364 coagulants." *Journal of applied Phycology* 22(3): 349-355.

365 Passos, F., M. Solé, J. García and I. Ferrer (2013). "Biogas production from microalgae
366 grown in wastewater: effect of microwave pretreatment." *Applied Energy* 108: 168-175.

367 Pieterse, A. and A. Cloot (1997). "Algal cells and coagulation, flocculation and sedimentation
368 processes." *Water Science and Technology* 36(4): 111-118.

369 Roeselers, G., M. Van Loosdrecht and G. Muyzer (2008). "Phototrophic biofilms and their
370 potential applications." *Journal of applied phycology* 20(3): 227-235.

371 Sahu, O., B. Mazumdar and P. Chaudhari (2014). "Treatment of wastewater by
372 electrocoagulation: a review." *Environmental science and pollution research* 21(4): 2397-
373 2413.

374 Sander, K. and G. S. Murthy (2010). "Life cycle analysis of algae biodiesel." *The*
375 *International Journal of Life Cycle Assessment* 15(7): 704-714.

376 Sridhar, R., V. Sivakumar, V. P. Immanuel and J. P. Maran (2011). "Treatment of pulp and
377 paper industry bleaching effluent by electrocoagulant process." *Journal of hazardous*
378 *materials* 186(2-3): 1495-1502.

379 Tenney, M. W., W. F. Echelberger, R. G. Schuessler and J. L. Pavoni (1969). "Algal
380 flocculation with synthetic organic polyelectrolytes." *Applied microbiology* 18(6): 965-971.

381 Udom, I., B. H. Zaribaf, T. Halfhide, B. Gillie, O. Dalrymple, Q. Zhang and S. J. Ergas
382 (2013). "Harvesting microalgae grown on wastewater." *Bioresource technology* 139: 101-
383 106.

384 Uduman, N., Y. Qi, M. K. Danquah, G. M. Forde and A. Hoadley (2010). "Dewatering of
385 microalgal cultures: a major bottleneck to algae-based fuels." *Journal of renewable and*
386 *sustainable energy* 2(1): 012701.

387 Vandamme, D., S. C. V. Pontes, K. Goiris, I. Foubert, L. J. J. Pinoy and K. Muylaert (2011).
388 "Evaluation of electro-coagulation–flocculation for harvesting marine and freshwater
389 microalgae." *Biotechnology and bioengineering* 108(10): 2320-2329.

390 Wei, V., M. Elektorowicz and J. Oleszkiewicz (2011). "Influence of electric current on
391 bacterial viability in wastewater treatment." *Water research* 45(16): 5058-5062.

392 Wong, Y., Y. Ho, H. Leung, K. Ho, Y. Yau and K. Yung (2017). "Enhancement of *Chlorella*
393 *vulgaris* harvesting via the electro-coagulation-flotation (ECF) method." *Environmental*
394 *Science and Pollution Research* 24(10): 9102-9110.

395 Zhuang, L.-L., Y.-H. Wu, V. M. D. Espinosa, T.-Y. Zhang, G.-H. Dao and H.-Y. Hu (2016).
396 "Soluble algal products (SAPs) in large scale cultivation of microalgae for biomass/bioenergy
397 production: a review." *Renewable and Sustainable Energy Reviews* 59: 141-148.

398 Zongo, I., A. H. Maiga, J. Wéthé, G. Valentin, J.-P. Leclerc, G. Paternotte and F. Lapique
399 (2009). "Electrocoagulation for the treatment of textile wastewaters with Al or Fe electrodes:
400 Compared variations of COD levels, turbidity and absorbance." *Journal of Hazardous*
401 *Materials* 169(1-3): 70-76.

402