Microalgae are considered as one of the most promising alternatives for the integrated use of agro-industrial water residues and the production of metabolites of high industrial interest. This is due to algae can grow on wastewater which in turn can reduce the emission of nutrients to rivers and lakes. However, the greatest scientific-technological barrier is the concentration and separation of the biomass produced. There are several processes used at different levels (from laboratory to industrial scale) such as flocculation, centrifugation, flotation, etc. These can be very expensive or can (possibly) contaminate the biomass. Unlike the previous ones, electrofloation has been proposed as a cost-efficient method, nevertheless its final efficiency will depend heavily on the type of alga and culture medium. Taking into account the above, the present project aims to design an electrofloation system for the concentration and harvest of microalgae biomass.

The effect of several factors (pH, time, voltage and distance between the electrodes) and for types of materials (Copper, Aluminium, Iron and Steel) on biomass recovery efficiency from a culture of *Chlorella vulgaris* UTEX 1803 was evaluated by the implementation of a Design of experiments (4³ non-factorial design) using STATISTICA 7.0.

Results show that, the materials with higher concentration efficiency were cooper and aluminium with 40 and 80% respectively, and the most relevant factors were distance between electrodes (1-2 cm), time (>20 min) and Voltage (>15V). In order to increase the efficiency of the overall process a new 4⁵ experimental factorial design was proposed using as factors distance between electrodes, time, voltage and agitation. Results show that agitation positively affects the total efficiency until reaching a total concentration of the biomass (100%). It was found that a voltage close to 50V and a time greater than 25 min positively affect the final efficiency of the copper and aluminium electrodes, however aluminium has the highest efficiency (> 95%) compared to copper (<85%).

1. Introduction

Microalgae are microorganisms with high photosynthetic efficiency, high biomass production and a relatively fast growth rate that play a key role in CO₂ capture and oxygen production on different ecosystems (Zenoussi et al., 2015). According to Baierle et al. (2015), about 1.8 kg of CO₂ are required for the production of 1 kg of biomass (dry weight) of microalgae. One of the main attractions of this type of microorganisms is the variety of metabolites, which have favorably impacted the development of industries such as pharmaceuticals, nutraceuticals and even the biofuel industry (Chisti et al., 2007).
One of the main technological barriers in large scale production is the lack of an efficient and affordable method for the concentration and separation of the biomass produced from the culture medium. This occurs due to the small cell size (0.1-3 μm) and that its surface is negatively charged and is loaded with algogenic organic matter (AOM) that protects the cell wall. These two factors contribute to keeping the cells dispersed in the medium (Henderson et al., 2008).

Currently there are several methods for harvesting microalgae. Centrifugation is a widely used method, this technology allows to concentrate the biomass of the majority of microalgae species known today. However, the efficiency of this process depends on three key factors: the rate at which the biomass sediments, the residence time of the biomass inside the centrifuge and the sedimentation distance (Molina et al., 2003). However, this technology has difficulties such as high operating time and high initial investment. In addition, the energy cost and its long-term maintenance (depreciation, materials and other associated costs) can represent up to 25% of the total operation (Henderson et al., 2008). Another technique applied to concentration is coagulation and / or flocculation; this has been a technology widely applied in the treatment of drinking water as well as the treatment of wastewater from different sources and can also be applied to the concentration of microalgae (Molina et al., 2003). There are other technologies such as filtration, sedimentation, bubble flotation, etc. (Table 1). However, most of the above have high costs, low separation efficiency and can irreversibly affect the quality of biomass (Misra et al., 2015).

Table 1: Comparison of technologies for algae concentration

<table>
<thead>
<tr>
<th>Type of Technology</th>
<th>Benefits</th>
<th>Difficulties</th>
</tr>
</thead>
<tbody>
<tr>
<td>Filtration</td>
<td>Economically viable and profitable</td>
<td>low efficiency and / or productivity</td>
</tr>
<tr>
<td>Centrifugation</td>
<td>Efficiency of 100%, maintains lipid quality and is a fast procedure</td>
<td>High costs and energy demand</td>
</tr>
<tr>
<td>Chemical coagulation</td>
<td>Economically viable, efficiency close to 100%, it is a simple and fast method.</td>
<td>Excess ions (SO$_4^{-2}$ and Cl$^{-1}$) generate irreversible changes to biomass</td>
</tr>
<tr>
<td>Floating by bubbles</td>
<td>Economically viable and profitable</td>
<td>Notoriously unstable</td>
</tr>
<tr>
<td>Electroflocculation/ flotation</td>
<td>Operates at low energy consumption, does not require the addition of any chemical flocculant; Applicable to a wide variety of microalgae.</td>
<td>Technology understudied</td>
</tr>
</tbody>
</table>

In recent years electroflocculation has emerged as an energy and environmentally more efficient possibility for water treatment, since it has been used successfully in the treatment of domestic and industrial wastewater (especially in mining) due to its simple reliability, the predictability of the results and the characteristic of being non-specific (Golzary et al., 2015, Lee et al., 2015). During the electroflocation process the metal ions are released by electrolytic oxidation of the anode material, these ions will serve as coagulating agents for the formation of microalgae flocs, at the cathode micro bubbles of oxygen and hydrogen are generated, helping the already formed flocs rise to the surface, these micro bubbles are generated due to oxidation and water reduction (Matos et al., 2013) and does not generate unnecessary anions such as SO$_4^{-2}$ and Cl$^{-1}$ that can generate irreversible modifications to the biomass. There are many factors that can influence the final efficiency of the process: electrode type (copper, aluminum, etc.) electrode size (submerged area), electrode gap, current (Amperes), treatment time, pH, conductivity of the microalgae suspension, etc. (Gao et al., 2010); however, until now, each of these variables have been evaluated independently without determining the possible correlation between one or several variables; so far to the author's knowledge there is no scientific article and/or study to establish the effect of all the variables together and their possible reaction in the final process efficiency. due to the above, the present work seeks to design and build a laboratory-scale electroflocation system offering alternatives and recommendations to perform microalgae concentration at a low cost, with simple but reliable equipment that can be replicated in other laboratories.

2. Materials and methods

2.1 Microalgae production

Chlorella vulgaris UTEX 1803 was purchased from the collection of strains from the University of Texas (Austin, Texas, USA). The algae were maintained in modified Bold Basal medium (Andersen et al., 2005) during 15 days in 500 mL GL 45 clear glass laboratory bottles previously steam sterilized (120°C, 60 min), each experiment was coupled to a bubbling aeration system with an air flow of 0.6 L/L of culture media.
2.2 Electroflotation experiments

In order to carry out the different experiments, a small equipment composed of three components (power source, reactor and electrodes) was constructed, which allow to obtain the necessary conditions for the concentration of microalgal biomass at the laboratory scale.

The power source was built by the construction of a simple and economical electrical circuit that allows to use four inexpensive 12V (1 Amp) power chargers in order to achieve a voltage close to 50V (figure 1). A 250 mL beaker with a working volume of 200 mL was used as reactor. Commercial sheets of copper, aluminium, stainless steel and steel were used as electrodes with a submerged area of 11.5 cm².

In order to determine the electroflotation efficiency of *C. vulgaris* a non-factorial central experiment design $4^3$ (4 factors and 3 levels) was applied using STATISTICA 7.0 software (Statsoft, 2004), using as variables: time, voltage, pH and distance between electrodes (Table 2). The efficiency of each experiment was calculated from the final optical density of each sample taken on a HACH DR1900 spectrophotometer at 550 nm and using Eq (1).

![Electrical circuit](image)

**Figure 1. Electrical circuit for connection of 4 12V (1 Amp) power chargers**

![Experimental setup](image)

**Table 1: Design of experiments for Algae Electroflotation**

<table>
<thead>
<tr>
<th>Time (min)</th>
<th>Voltage (V)</th>
<th>pH</th>
<th>Distance between electrode (cm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>5</td>
<td>4</td>
<td>4</td>
<td>2</td>
</tr>
<tr>
<td>10</td>
<td>7</td>
<td>7</td>
<td>3</td>
</tr>
<tr>
<td>15</td>
<td>10</td>
<td>10</td>
<td>4</td>
</tr>
</tbody>
</table>

$$Efficiency\,(\%) = \left( \frac{Initial\ OD_{550nm} - Final\ OD_{550nm}}{Initial\ OD_{550nm}} \right) \times 100$$  \hspace{1cm} (1)

3. Results and discussion

3.1 Electrodes efficiency

Results shown that aluminum (63.9%) and Iron (35.4%) were the materials with the highest efficiencies compared to copper (32.8%) and steel (17.4%) (Figure 2). However, during the process iron and steel electrodes underwent a rapid and continuous oxidation process that affected the biomass and modified the color of the medium (changing from green to red); the above can irreparably contaminate the biomass. Unlike the above, the copper electrodes and aluminum showed no change in coloration of biomass or the medium.

Within the results it was possible to determine that the best operating conditions for the aluminum are a voltage of 10V a time greater or equal to 12 minutes and a distance between electrodes of 1-3 cm. In the case of copper, a voltage greater than 20V is required, in a time greater than 12 minutes and a distance of 1-2 cm. Finally, it can be emphasized that solutions with neutral pH (pH 7) obtained the best floculation efficiencies for all materials. Because the strain used (*C. vulgaris*) comes from fresh water and its culture pH is close to neutral, it is not necessary to make any previous adjustments.
According with Matos et al (2013), it is possible to reach efficiencies close to 100% by increasing retention time and voltage, therefore an optimization phase was proposed to improve the efficiency using aluminum and copper as electrodes. In order to increase the efficiency a new design of experiment was proposed (table 2), which includes an additional variable (Agitation) plus the best conditions established in the first experimental design.

![Figure 2. Efficiency of materials](image)

**Table 2: Design of experiments for Optimization of Algae Electroflotation**

<table>
<thead>
<tr>
<th>Time (min)</th>
<th>Voltage (V)</th>
<th>Agitation (rpm)</th>
<th>Distance between electrode (cm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>10</td>
<td>20</td>
<td>100</td>
<td>2</td>
</tr>
<tr>
<td>15</td>
<td>30</td>
<td>150</td>
<td>3</td>
</tr>
<tr>
<td>20</td>
<td>40</td>
<td>200</td>
<td>4</td>
</tr>
</tbody>
</table>

**3.2 Electroflotation Optimization**

From the new experiment design, it was possible to significantly increase the efficiency of both copper (from 32.8 to 88%) and aluminum (from 64 to 97%) (Figure 3). According to the Pareto chart (Figure 4), it was found that the variables that influence the efficiency of copper are voltage, time and distance between the electrodes; In addition, the variables voltage and time are directly related: longer (25 min) and voltage (> 45V) and a distance between 1-2 cm increases the overall efficiency of the process. Unlike the previous one, in aluminum it was found that only time and voltage affect the efficiency of the process (figure 5), where a time equal to or greater than 25 minutes and a voltage equal to or greater than 40 V. these results are congruent with those presented by Udhaya et al. (2014), where biomass from strains such as C. vulgaris KC492080, Chlorococcom sp KC492078 and Chlamydomonas sp KC492081 can be concentrated up to 90% using aluminum electrodes.

![Figure 3. Efficiency of Aluminum and Cooper.](image)
Although it was found that for the values evaluated the agitation does not present as a variable that influences positively or negatively in the process (figure 4 and 5) Zhou et al. (2016) showed that the agitation is an essential variable in the electroflotation of algal cells, since agitation allows the medium to be homogenized, forcing a larger number of cells to contact the electrodes and allowing flocs to form, in turn, agitation prevents the sacrificial electrode (anode) from filling up with cells.

4. Conclusions

In the first phase of experimentation it was found that the iron electrodes present a rapid oxidation after a few minutes, contaminating the microalgal biomass and making the electro-float inefficient; it was also found that the pH adjustment of the slope does not play an important role in the concentration of biomass.

By increasing the voltage and time it was possible to improve the final efficiency for the copper and aluminum electrodes, on the other hand it was found that agitation was not a decisive factor. However, the homogenization of the medium will allow all of the microalgal cells come in contact with the electrodes and reduce the time of reaction. Finally, the results shown that an inexpensive small equipment composed of three components (power source, reactor and electrodes) can be successfully used for the concentration of small amounts of algae culture without the necessity of expensive equipments. The latter implies the possibility of developing new projects that evaluate the possible effects of this type of technology on the quality of the...
biomass produced; in turn, will allow different laboratories in different parts of the world to test with other microalgae, thus permitting the creation of better equipment that increase the efficiency of microalgae production processes for different industries.

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Reference


