Application of Biosurfactant Formulated as an Alternative Collector in a Bench Scale-Induced Saturation Tower for the Treatment of Industrial Effluents

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Dissolved air flotation technology is the most efficient and economical solution for the treatment of industrial effluents, using biosurfactants to replace chemical surfactants, due to the tensioactive properties, greater stability and biodegradability. The present work aims to present an efficient solution through the new DAF technology, configured in the Induced Saturation Tower (IST) in stages, in bench scale. To increase the efficiency of the process, an anionic polymer as flocculating and a microbial biosurfactant was used in the treatment of oily effluents. In this sense, the potential of the biosurfactant produced by Pseudomonas cepacia CCT6659, cultivated in an economic culture medium composed of industrial waste, as alternative collector, was evaluated. The tests were performed in the Tower after homogenization between biosurfactant/oil/water. The oily effluents without the addition of the biosurfactant were used as controls. The samples were fed to the top of the tower and interacted successively with upward flows of air microbubbles at different stages. Samples from the treated effluents were collected to evaluate the percentage of oil removal by spectrophotometry and turbidimetry. The most expressive results demonstrated an increasing capacity of 80% of contaminant removal during 20 minutes of test. The initial turbidity of the industrial effluent was 288.6 NTU, and soon after the addition of the mixing of the collectors (polymer + biosurfactant) to the effluent, there was a considerable reduction of the turbidity to 101 NTU. Therefore, the use of biosurfactants as auxiliaries in flotation, associated to a new DAF technology, is a promising alternative for the industrial sector, avoiding the negative environmental impacts caused by the oily waters generated by the industries.

1. Introduction

Environmental problems have become increasingly routine, causing surface and groundwater pollution due to two major factors: population growth and industrial growth (Karlapudi et al., 2018). The increase in activities related to the petroleum and electricity sector are mainly responsible for the production of oily waters and their disposal, requiring systems for the treatment and separation of these wastes (Chaprão et al., 2018). In this context, one of the main techniques of separation of water and oil that has been successfully highlighted in the treatment of effluents is the process of by dissolved air flotation (DAF), which consists of the separation of particles through adhesion of bubbles, allowing the reuse of the involved phases economically (Rocha e Silva et al., 2017; Albuquerque et al., 2012). Currently, horizontal floats with macro bubble action are used. However, the innovation presented in this work comprises a vertical float in stages with microbubble performance and probably nanobubbles. These microbubbles are more reactive because they reach a larger surface area, raising the separation efficiency.

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The flotation uses mostly chemical surfactants to enhance the separation of the suspended material (Albuquerque et al., 2012). Surfactants are amphipathic molecules containing hydrophilic and hydrophobic moieties that partition at the oil / water or air / water interface. These characteristics allow surfactants to reduce surface and interfacial tension and form microemulsions (Santos et al., 2016). In order to replace synthetic and toxic surfactants, some alternatives have been highlighted, such as the use of biosurfactants in flotation processes, because they are biodegradable and with low toxicity, mitigating environmental impacts (Almeida et al., 2016). In the face of the challenges presented and the needs of development and improvement of the currently known techniques, this paper aims to propose effective solutions through technology of DAF. A new bench scale flotation system, configured in a stages saturation tower with the action of microbubbles, using a microbial biosurfactant as an alternative collector in the treatment and control of oily residues in the industrial area.

2. Materials and Methods

2.1 Synthetic Oily Effluent and Industrial Effluent

Knowing that 20 ppm is the maximum amount of oil allowed in industrial effluents by CONAMA, the values of concentration of oil in water were established at 150 ppm. A commercially available lubricant oil for use in engines SAE 20W-50 (Petrobras, Brazil) was used as a hydrophobic contaminant. The oily industrial Wastewater was obtained from a thermal power plant, located in the city of João Pessoa, State of Paraíba, Brazil. The effluent has a composition of approximately 500 ppm (5 %) of heavy oil (OCB1 fuel oil, with a kinematic viscosity of 620 cSt, at most 1 % sulphur (p/p), a maximum of 2 % of water and sediment (v/v), flash point to 66 °C, fluidity point greater than 24-27 °C, maximum of 200 mg/kg of vanadium, PETROBRAS in Brazil) and a yellowish color resulting from the routine activities of the plant (Soares da Silva et al., 2018).

2.2 Cationic Polymer

The water-soluble cationic polymer was used as a collector to increase the efficiency of the solid particulates in the industrial effluent. It is used as a flocculant in sewage treatment plants or industrial tributaries and is available commercially (FAXON QUÍMICA LTDA).

2.3 Obtaining the Microbial Biosurfactant

The bacterium Pseudomonas cepacia CCT 6659, obtained from the culture bank of the André Tosello Research and Technology Foundation, located in the city of Campinas - São Paulo, was used as a microorganism producing the biosurfactant. Cultures were peaked every 30 days and kept in sloped test tubes with the solid Nutrient Agar (NA) medium under refrigeration at 5 °C. Young cultures of the bacteria were transferred to an Erlenmeyer containing 50 mL of BHI and maintained under 150 rpm orbital shaking for a period of 10-14 hours at 28 °C to obtain a D.O. of 0.7 (corresponding to an inoculum of 10^7 U.F.C./mL) at 600 nm. The production of biosurfactant was carried out according to Soares da Silva et al. (2017), used a mineral medium composed of: 0.5 g/L de KH₂PO₄, 1 g/L de K₂HPO₄, 0.5 g/L de MgSO₄.7H₂O, 0.1 g/L de KCl e 0.01 g/L de FeSO₄.7H₂O, supplemented with 2.0 % canola oil frying and residual 3.0 % cornsteep liquor as substrates, pH 7.0, at a temperature of 28 °C, during 60 hours of cultivation, under 250 rpm agitation and an inoculum of 1.5 %. The proposed biosurfactant is recognized by your excellent performance in bioremediation processes.

2.4 Formulation of biosurfactant

The cell free metabolic liquid containing the biosurfactant obtained after centrifugation for biomass removal was subjected to addition of 0.2 % potassium sorbate as preservative and conditioned under sterile conditions in a sealed container at room temperature (Soares da Silva et al., 2018).

2.5 Evaluation of the tensioactives properties of biosurfactant

The surface tension of the biosurfactant was measured on a KSV Sigma 700 (Finland) tensiometer using the NUOY ring. Subsequently the biosurfactant was extracted to determine its Critical Micellar Concentration (CMC) (Rocha e Silva et al., 2014). After extraction, the product was treated with a base and crystallized for maximum removal of impurities. For the determination of the emulsification activity, samples of the formulated and isolated biosurfactant added in hydrophobic compound (motor oil) were analyzed according to the methodology described by Cooper and Goldenberg (1987). The dispersion or aggregation capacity of petroleum by products was simulated in the laboratory by contaminating samples of distilled water with engine oil. The tests were conducted by adding the formulated biosurfactant and isolated to motor oil in proportions (1:2, 1:8 and 1:25 v / v). The results were observed and measured visually (Saeki et al., 2009).
2.6 Evaluation of the biosurfactant as an alternative collector in the Tower of Induced Saturation (IST)

The tests were performed in an Induced Saturation Tower (IST) in Stages in bench scale, according to schematic representation (Figure 1). A 34 L volume of oily effluent added with 600 mL of formulated biosurfactant and 0.8 g/L of polymer was homogenized for approximately 5 minutes to obtain a uniform water/oil/manifold distribution. The oily effluent without the addition of the biosurfactant was used as control. The sample was fed to the top of the tower and interacted in countercurrent and, successively, with upward flows of air microbubbles in the bases of the different stages. The formed bubble particle aggregate was then transported to the upper section of each stage, due to the geometric shape of the stage cap, exiting through a foam collector conduit and solids. Samples of the treated effluent were collected after 5, 10, 15 and 20 minutes of process to evaluate the percentage of oil removal. The samples were evaluated by spectrophotometry using a calibration curve prepared with a standard solution of the oil at 5000 mg/L. Samples of the industrial effluent after 5 minutes of process were also used to evaluate the turbidity.

2.7 Statistical analysis of the data obtained

The data collected will be expressed as a mean ± standard deviation of the tests performed in triplicate. A statistical analysis of variance of ANOVA will be applied to determine a significance, where values of p <0.05 will be analyzed.

3. Results and discussion

The majority of biosurfactants produced by bacteria have demonstrated the capacity to reduce surface tension to values around 28 mN/m (Soares da Silva et al., 2017).

The formulated biosurfactant produced by *Pseudomonas cepacia* CCT 6659 was able to reduce the surface tension of the medium from 65 mN/m to 26 mN/m with a Critical Micellar Concentration (CMC) of 0.6 g/L. The results of the emulsification activity demonstrated a high affinity of the microbial surfactant to the hydrophobic compound (motor oil) evaluated (Table 1). The formulated biosurfactant emulsified up to 95.5% of the residual motor oil, demonstrating potential to be applied at oil contaminated sites. The ability to emulsify hydrocarbons depends on the hydrophobic compound as well as the structure of the biosurfactant, which is

*Figure 1: Induced Saturation Tower in Stages in bench scale.*
influenced by the substrate used. This capacity is related to the compatibility between the structure of the biosurfactant and the hydrocarbon, which will allow the stabilization or not of the microscopic droplets of the emulsion (Silva et al., 2014).

The dispersion percentages of the formulated and isolated biosurfactant demonstrated an optimum dispersant capacity of the surfactant evaluated (Table 1), with dispersion indices of approximately 75.0%. It is evident that the biosurfactant produced by *P. cepacia* has stability in all the biosurfactant/motor oil relationships tested. The results indicate a good interaction between the biosurfactant and the motor oil employed, even at oil concentrations 25 times higher than that of the microbial surfactant.

**Table 1 – Percentages of emulsification and dispersion of motor oil by Pseudomonas cepacia biosurfactant formulated and isolated**

<table>
<thead>
<tr>
<th>Biosurfactant / Oil</th>
<th>Biosurfactant formulated</th>
<th>Biosurfactant isolated (CMC)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Emulsification</td>
<td>95.50 ± 0.07</td>
<td>90.12 ± 0.12</td>
</tr>
<tr>
<td>Dispersion ½</td>
<td>75.23 ± 0.03</td>
<td>65.16 ± 0.14</td>
</tr>
<tr>
<td>Dispersion 1/8</td>
<td>53.22 ± 0.05</td>
<td>45.58 ± 0.11</td>
</tr>
<tr>
<td>Dispersion 1/25</td>
<td>40.47 ± 0.11</td>
<td>30.92 ± 0.17</td>
</tr>
</tbody>
</table>

For the application of the biosurfactant produced by *P. cepacia* as an alternative collector in the treatment of oily effluents, the results demonstrated the high efficiency of oil removal from the synthetic and industrial effluents with and without the addition of formulated biosurfactant, associated to the action of the microbubbles in the Induced Saturation Tower (IST) in stages. Up to 80% of the contaminant was removed during the 20 minutes test (Figure 2). Both treatments presented satisfactory removal results. However, it is important to note that the test without addition of biosurfactant presented descending percentages of removal with the advancement of time. The test with the addition of the biosurfactant, demonstrated ascending percentages of removal as the residence time of the collector increases with respect to the effluent. From this analysis, it is necessary to emphasize that the dimensions of the flotation stages are limited, thus having a short residence time and consequently influencing at the time of action between biosurfactant / oil / microbubbles. For maximum removal efficiency, the study of a longer residence time needs to be considered. However, it was possible to obtain an excellent evolution of the results in the IST. Silva et al. (2018), investigated the separation of oil into water using a pilot scale horizontal (DAF) prototype, with and without the use of a microbial biosurfactant. The biosurfactant considerably improved the process, increasing the water/oil separation efficiency from 41.0% to 98.0%. Chaprão et al. (2018), using a microbial biosurfactant in a horizontal (DAF) prototype, achieved a 92% rate of oil removal in a synthetic effluent.

**Figure 2 – Results of the oil removal efficiency with and without the biosurfactant of *P. cepacia* as an alternative collector, associated with the action of the microbubbles in the Tower of Induced Saturation**
The results for the oil removal efficiency of the industrial effluent through the turbidity test proposed by Brazilian environmental regulator (CONAMA), showed very expressive values. The initial turbidity of the industrial effluent was 288.6 NTU, after addition and mixing of the collectors (polymer + biosurfactant) to the effluent, there was a considerable reduction of turbidity to 101 NTU (Figure 3). After the flotation process associated with the action of the microbubbles in the Tower of Induced Saturation (IST) in stages, the biosurfactant + polymer considerably reduced turbidity reaching 29 NTU, equivalent to 71.28 % of removal of contaminants in suspension in the industrial effluent. In this way, adapting to the standard demanded by CONAMA.

![Figure 3 – Results of the efficiency of turbidity reduction of the industrial effluent with and without the polymer + biosurfactant of P. cepacia as collectors, associated to the action of the microbubbles in the Tower of Induced Saturation](image)

### 4. Conclusions

The biosurfactant of *P. cepacia* presented excellent tensoactive properties and had its proven efficiency during its application in the Induction Saturation Tower (IST) on a bench scale, with the action of microbubbles. In this way, the biotensoativo revealed great potential to be used as an alternative collector in the treatment of oily effluents in the DAF system, allowing an increase in the separation efficiency of the process. Therefore, the innovative flotation system in staged, coupled with a biotechnological agent, demonstrated the ability to be applied in processes of separation of organic and inorganic pollutants generated in the industries.

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