Nonionic Linear Alcohol Ethoxylated removal in an Anaerobic Fluidized Bed Reactor

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Abstract
Nonionic surfactants have received much attention from scientists in recent decades, mainly because of their high consumption and disposal into sewers. This experiment was carried out in an anaerobic fluidized bed reactor for a range of nonionic surfactants (Linear Alcohol Ethoxylate – Genapol® C-100) at concentrations from 10 to 120 mg/L added to a synthetic substrate. The reactor was operated in five phases: inoculation (535±121 mg/L of COD), biomass adaptation (600±70 mg/L of COD), Phase I (4.7mg/L of LAE and 623±65mg/L of COD), Phase II (22.5 mg/L of LAE and 735±87 mg/L of COD), Phase III (51.4 mg/L of LAE and 697±68 mg/L of COD), Phase IV (107.4 mg/L of LAE and 845±87 mg/L of COD) and Phase V (97.9 mg/L of LAS and 882±126 mg/L of COD). The organic matter and LAE removal efficiencies' averages were 88% and above 99.9%, respectively, throughout the reactor operation. The populations of the Archaea and Bacteria Domains similarity were 74% and 59%, respectively, between reactor samples for phases IV (with a co-substrate) and V (without a co-substrate).

Keywords
Genapol® C-100, PCR/DGGE, Methanogenic Archaea, LAE

INTRODUCTION

Surfactants are compounds commonly used in industrial processes, cleaning and personal care products. They are major sources of industrial, commercial and domestic pollution. Nonionic surfactants are consumed at a large scale, and they release toxic substances caused by their degradation (SCOTT and JONES, 2000; PETROVIC et al., 2004). Alcohol ethoxylated and alkylphenol ethoxylated are the nonionic surfactants with the highest consumption rate in Europe, accounting for approximately 74% of the market share in this category (COELHO et al., 2009). The mechanism of biodegradation and the removal of these compounds from effluents, both in batch and in continuous systems, has been extensively studied. The purpose of this study was to evaluate the removal and degradation of LAE (Linear Alcohol Ethoxylate, Genapol® C-100) in an anaerobic fluidized bed reactor operated continuously and filled with biomass immobilized on sand as a support material.

MATERIAL AND METHODS

The anaerobic fluidized bed reactor was composed of acrylic (1.256 L) 4 cm in internal diameter and 100 cm in height. This system was filled with 306 g of sand and operated with a hydraulic retention time (HRT) of 18 hours at 30ºC for all phases. The reactor was inoculated with sludge from a UASB reactor used to treat swine waste and fed with synthetic substrate (sucrose, yeast extract, sodium bicarbonate and salt solution) (TORRES, 1992; DUARTE, 2006) and nonionic surfactant LAE (Genapol® C-100 SIGMA-ALDRICH) addition. However, in the last phase (V) the sucrose was removed from the feed. The reactor was operated in five phases (Table 1) beyond the adaptation of the biomass period. Physical chemical properties, such as COD, pH, alkalinity, solids according to the APHA (2005) and volatile organic acids (PENTEADO, 2011), were analyzed. LAE quantification was carried out by high performance liquid chromatography (HPLC–*Shimadzu System*) using fluorescence detector RF-10AXL and C8 reverse phase column (Supelco). The most

probable number (MPN) was determined using the multiple-tubes technique (APHA 2005) with samples taken from the end of the reactor operation for phases IV and V. As in previous analysis (MPN), the PCR/DGGE technique (Bacteria and Archaea Domains) was performed in the two final phases of the reactor operation. For the Archaea Domain, the *primers* 1400R and 1100FGC (KUDO, 1997) were used, and for the Bacteria Domain, the *primers* 1401R and 968FGC (NUBEL, 1996) were used.

Table 1. Phases of operation of the anaerobic fluidized bed reactor

	Phases						
	Inoculation	**Adaptation**	**I**	**II**	**III**	**IV**	**V**
Duration (Days)	20	40	93	89	68	90	73
COD influent (mg/L)	535±121	600±70	623±65	735±87	697±68	845±87	882±126
OLV (mgCOD/L/d)	715	802±94	834±86	991±112	932±91	112±116	118±169
LAE influent (mg/L)	-	-	4.7	22.5	51.4	107.4	97.9

COD – Chemical Oxygen Demand, OLV – Organic Load Volumetric, LAE – Linear Alcohol Ethoxylate (Genapol® C-100)

RESULTS AND DISCUSSION

The pH remained constant throughout the reactor operation. The influent pH average was 7.5±0.4 to 7.0±0.1, and the effluent average was 8.0±0.1 to 7.9±0.2 for the adaptation and phases I to V. The influent alkalinity varied from 273±16 mg$CaCO_3$/L (Phase III) to 310±33 mg$CaCO_3$/L (Phase V), and the effluent varied from 359±37 mg$CaCO_3$/L (adaptation) to 386±36 mg$CaCO_3$/L (Phase I). The OLV ranged from 802±94 mgCOD/L/d (Adaptation) to 1180±169 mgCOD/L/d (Phase V). Throughout the reactor, the operation of the COD influent was 600±70 mg/L (Adaptation) and 882±126 mg/L (Phase V) (Figure 1). Although the LAE addition generated low OLV between the phases, this compound altered the values of the COD influent, mainly in the phases of higher surfactant concentration (phases IV and V). The effluent COD ranged from 50±18 mg/L (Phase II) to 145±93 mg/L (Phase V). In Phase I, the organic matter removal efficiency had a greater variation, most likely due to the surfactant addition and, therefore, the biomass adaptation to the compound. This efficiency varied from 83% (Phase V) to 93% (Phase II). Even though this efficiency was high throughout the reactor operation, after Phase II, there was a decrease in these values, demonstrating that the addition and the increase of the LAE concentration altered the microbial biofilm in the reactor.

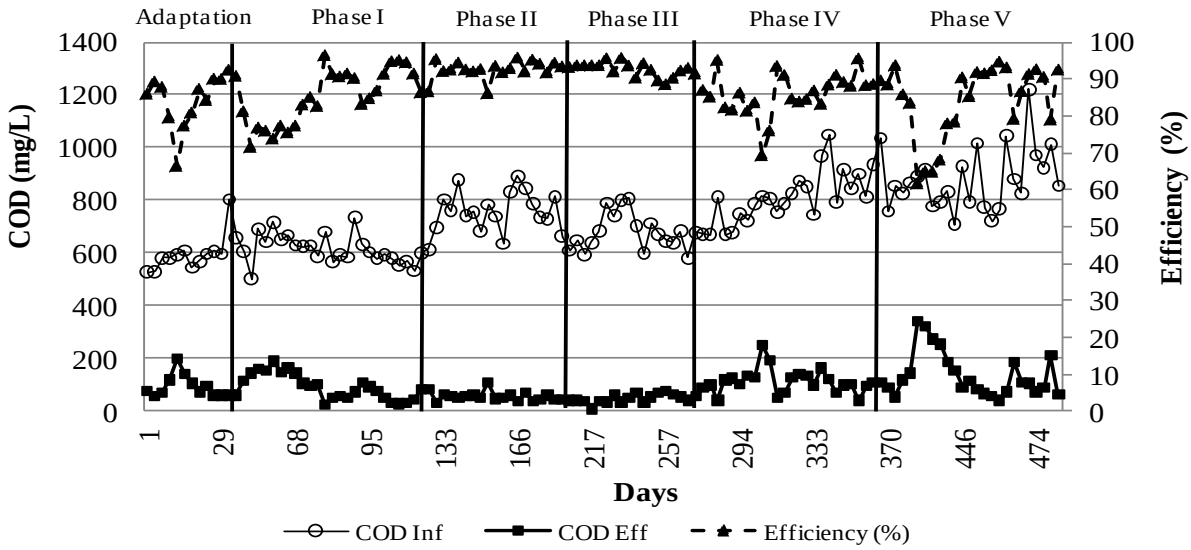

Figure1. Temporal variation of the COD influent, effluent and removal efficiency

There was a high removal efficiency of LAE (Genapol® C-100) throughout the reactor operation (Table 2). The influent averages varied from 4.7 mg/L (Phase I) to 107.4 mg/L (Phase IV), and the effluent values from Phase I to Phase V were below the detection limit of the quantification curve (5 mg/L). The LAE removal efficiency was above 99.9% for all the reactor operation phases. Therefore, there were high removal efficiencies for this surfactant, even in Phase V (without sucrose). HUBER et al. (2000) performed degradation tests with alcohol ethoxylates in anaerobic conditions in batch reactors. The authors proved that more than 99% of the LAE (dodecanol ethoxylate) degraded for the 5 mg/L and the 40 mg/L influent after 22 and 50 days, respectively.

Table 2. LAE quantification in the HPLC method

	Phases				
	I	**II**	**III**	**IV**	**V**
Duration (Days)	93	89	68	90	73
LAE Inf. (mg/L)	4.7	22.5	51.4	107.4	97.9
LAE Eff. (mg/L)	< LOQ	< LOQ	< LOQ	< LOQ	< LOQ

LAE – Linear Alcohol Ethoxylate (Genapol® C-100), Inf. – Influent, Eff. – Effluent, LOQ – Limit of Quantification. LAE calibration curve: 5 – 30 mg/L

The volatile fatty acids (VFA) production remained low throughout the reactor operation. The highest value was found in Phase I, where the average was 155 mg/L. This fact is justified by the short microbial selection due to the lower surfactant concentration used in this phase. There was little variation in the VFA values for phases IV and V, which had average values of 82 mg/L and 90 mg/L, respectively. However, the prevalence of the acids changed the percentage between these phases. In Phase IV (with a co-substrate), isobutyric acid was most abundant (29%), followed by succinic acid (13%). In contrast, in Phase V (without a co-substrate), succinic acid was prevalent (34%), followed by isobutyric acid (24%). These facts show that a co-substrate can change microbial populations and, therefore, acid production in the reactor. Changes were noted in the microbial populations in relation to the spatial location inside the reactor (the distributor - D, biofilm in the support material - SM, and the phase separator - PS). For the Archaea Domain, in Phase IV, a similarity of 98% was observed between the populations in the D and the PS and 90% between the populations in the D and the SM. For the Bacteria Domain, a similarity of 90% and

82%, respectively, was observed. In Phase V (without a co-substrate), for the Archaea Domain, a similarity of 89% was observed between the PS and the SM and a similarity of 82% was observed between the populations in the PS and the D. For the Bacteria Domain, there was a similarity of 85% between the populations of the D and the SM and a similarity of 76% between the populations of the PS and the SM. The hydrodynamic regime in a fluidized bed reactor is very turbulent. Thus, the biofilm formation in the SM is not thick and provides microbial selection when compared to the PS and the D, which have microorganisms that form biopolymers. The similarities found between Phases IV and V were 74% (Archaea) and 59% (Bacteria), proving that the sucrose absence selected the biomass throughout the reactor. In Phase V, 7.0E+12 cells/gTVS of total anaerobic bacteria (TAB) and 2.7E+9 cells/gTVS of methanogenic archaea (MA) were found. In the PS, 2.0E+13 cells/gTVS of TAB and 2.5E+10 cells/gTVS of MA were found in the same operation phase. In the PS, there was a greater TAB presence than in the MA because the PS was a facultative environmental (confirmed by the Resazurin test).

CONCLUSIONS

Because of the high recirculation rate, the anaerobic fluidized bed reactor (HDT of 18 hours) was adequate for Linear Alcohol Ethoxylate (LAE - Genapol® C-100) removal. The sucrose absence selected the microbial populations in the reactor, and these were capable of using the surfactant as the carbon source even at high concentrations. The sucrose absence also changed the acid concentration generated in the anaerobic process of the Linear Ethoxylated Alcohol degradation.

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