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Phenol biodegradation at high organic loads in a complete sludge reactor by activated sludge

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Abstract

The presence of phenols in industrial discharges have phenol concentrations between 35 and 400 mg/L. But there are extreme cases, with concentrations of 30 to 80 g/L. The methods used for its treatment include: degradation, elimination and recovery. Its biological treatment is complicated because it causes the inhibition of microorganisms. The objective of this work was to evaluate the effect of the organic load on the phenol biodegradation rate of an industrial effluent. An activated sludge reactor was used at 1.76 and 2.81 days of hydraulic retention time (HRT), 25±0.5°C and air flow of 2.5 L/min, without pH control. The results showed a HRT of 1.76 days, with an organic load of 11.2±0.3 kg COD/m^{3}·d (run I), a lower COD removal rate, compared with the phenol biodegradation rate (30 and 41%). At 7.97±0.12 kg COD/m^{3}·d (experiment II) and the same HRT, the COD removal rate was higher compared to phenol biodegradation (56 and 37%). And with 3.3±0.2 Kg COD/m^{3}·d) (experiment III), the rate of removal of COD as of phenol biodegradation was 34% at 2.81 days of HRT.

Phenols, Toxicity, Biodegradation of Phenol, Activated Sludge, Catechol

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Introduction

Phenol and its by-products are some of the most widely studied organic pollutants due to their toxicity to living things and the environment. Phenol and its byproducts are some of the most extensively studied organic pollutants due to their toxicity to living beings and the environment (Agencia de Protección al Ambiente de EEUU, ATSDR, 2016).

Phenolic pollutants discharged from different kind of industries, such as pesticides, paints, polymer resins, paper, and petrochemistry have been found in their effluents (Ahmaruzzaman, 2008; Lin et al., 2009). As well as those discharged by the oil processing industry and refineries, which contain various compounds; Fats and oils, sulfates, suspended solids, cyanides, nitrogen compounds and heavy metals such as chromium; Iron, nickel, copper, molybdenum, selenium, vanadium and zinc. Organic compounds, such as: aliphatic and aromatic hydrocarbons, sulfur compounds, nitrogen compounds and phenols in concentrations higher than 30,000 mg/L (Wake, 2005). Effluents from coke ovens and pharmaceuticals have phenol concentrations between 10 and 300 mg/L (Lepik and Tenno, 2011; Pramparo et al, 2012). However, the presence of phenol in wastewater has been reported with concentrations up to 10,000 mg/L (Krastanov et al, 2013).

According to section 313 of Title III Emergency Planning decreed by the EPA (2015), phenol by having in its structure a benzene ring, i considered a pollutant priority due to its high toxicity and low biodegradability, in addition to being considered carcinogenic, mutagenic and teratogenic. it is easily absorbed by the skin and mucous membranes of animals and humans.

Its toxicity directly affects a great variety of organs and tissues, mainly lungs, liver, kidneys and genitourinary system (Olujimi et al., 2010).

Finding in upper freshwater and marine organisms, minimum values of CL (lethal concentration) or EC (critical exposure) in crustaceans and fish between 3 and 7 mg/L phenol (ATSDR, 2016).

In the literature, treatment technologies for phenolic compounds are addressed: the first consists of chemical processes oxygenation with reagents such as hydrogen peroxide (Qayyum et al., 2009) and the second of the adsorption and extraction processes (Lin and Juang, 2009; Smith et al., 2009). Other technologies contemplated chemical coagulation (Ozbolge et al., 2002), solvent extraction (Yang et al., 2006), membrane techniques (Kujawski et al., 2004), microfiltration (Wei et al., 2004), inverse osmosis (Ipek et al., 2004), biodegradation (El–Naas y Makhlouf, 2008), chemical oxidation (Suarez-Ojeda et al., 2007) and electrochemistry (Yavuz et al., 2006).

The conventional processes in the detoxification of this type of compounds have proved to be efficient, but they present certain disadvantages and limitations, due to the fact that, at concentrations of 10 mg/L phenol, causes inhibition of microorganisms. Their treatment through these processes requires extreme operating conditions, leading to the formation of intermediates, which, far from solving the problem, contribute to water pollution. Under this scenario, its toxic effect has been extensively studied using microorganisms (bacteria, fungi, protozoa, and algae) and numerous vertebrates and aquatic invertebrates.

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Therefore, prokaryotic and eukaryotic organisms under aerobic respiration conditions can carry out its degradation with oxygen being the final acceptor of electrons, using carbon molecules to produce CO_2, H_2O and energy in the form of ATP (Rodríguez, 2003).

With the use of microorganisms, decontamination is achieved by effectively eliminating these compounds, since bacteria, protozoa, and other major microorganisms disintegrate most organic compounds such as phenol, by reactions that use oxygen, transforming it in substances rich in energy and in substances of lower energy. The treatment of industrial effluents with high concentrations of phenol by activated sludge is an alternative for the control, correction and regeneration of the ecosystems affected by discharges of this nature.

The aerobic pathway has shown that there is a common metabolic pathway for this type of compound and even for those not so close to the phenolic compound family as biphenyls (Autenrieth et al, 1991). During which, the oxygen is consumed, and by the action of the enzyme phenol monooxygenase, an ⁻OH group is added to the phenolic ring resulting in the formation of catechol. In this way, the catechol ring can be broken in two different ways, by the *ortho* route and by the meta route. During the *ortho* route, the catechol bond 1-2 is ruptured to produce the muconic acid (Figure 1).

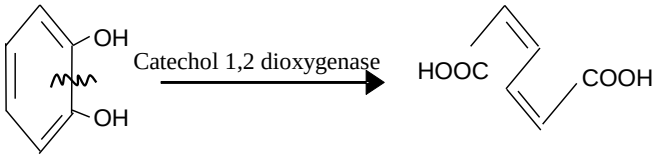

Figure 1 Ortho rupture of the phenolic ring

By the meta route, the ring rupture occurs between carbons 2-3, to give rise to the formation of the 2-hydroximuconic semialdehyde (figure 2).

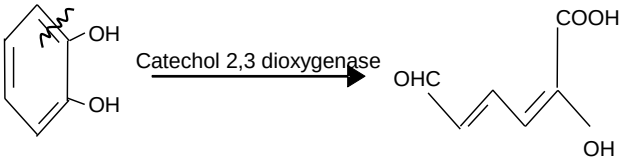

Figure 2 Meta rupture of the phenolic ring

In this route, after the rupture of catechol catalyzed by the enzyme catechol 2,3-dioxigenase, the 2-hmas can be degraded in two different ways as shown in Figure 3. In the first, the 2-hmas is oxidized to give rise to the formation of 2- hydroximuconic acid, which is subsequently decarboxylated through the action of two enzymes, an isomerase and a decarboxylase, to form 2-oxopenta-4-enoic acid. For its part, the 2-hmas is fractionated by hydrolysis to produce the acids; formic and 2-oxopenta-4-enoic acid. The latter degraded by hydration under the action of an aldolase to 4-hydroxy-2-oxopentanoic acid and then to acetaldehyde and pyruvic acid respectively, which continue to oxidize upon incorporation into the Krebs cycle (Ramírez, 2005).

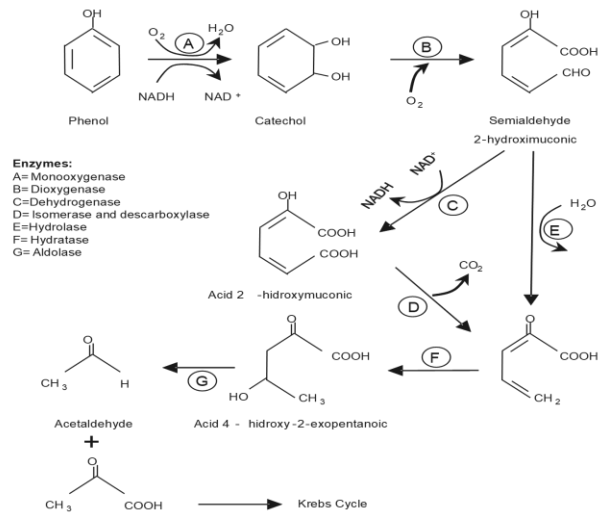

Figure 3 Meta pathway of aerobic phenol biodegradation (Shalaby, 2003)

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Genes that encode the pathway enzymes *meta* in the bacteria, are part of large plasmids such as TOL, while those encoding the pathway enzymes *orto*, most are located in the DNA (Selesi et al., 2010).

The success of phenol biodegradation in wastewater, It depends on the strategy employed during the acclimation of the bacterial consortium (biomass) of the biological reactor to the presence of this type of toxic compounds, what results a key factor in achieving the highest rate of biodegradation (Terreros et al., 2016). Since its bactericidal effect of phenol, Is based on the ability of the compound to dissociate within the cells, provoking that the functions of the cytoplasmic membrane are disrupted, causing cell death (Tay et al., 2005).

Biodegradation by biological processes, involves many factors. Among them; temperature, pH, because each microorganism has a specific range of temperature and pH for his growth (pH=7.0) (El-Naas et al., 2009).

Whilst extreme values of pH (less than 3 or greater than 9) are inhibitory for growth (Agarry et al., 2008). Other important factors are; dissolved oxygen concentration, concentration of substrate (Trigo et al., 2009) and the organic loading rate (Liu et al, 2003a). That is to say, a relatively high organic load facilitates the formation of anaerobic granules in a system UASB. In contrast, there are evidence to suggest that aerobic granules can be formed over a wide range of organic charge rates, ranging from 2.5 to 15 Kg COD/m^{3}·d. With an increase in the organic load of 3 to 9 Kg COD/m^{3}·d, the size of the aerobic granules increases 1.6 to 1.9 mm (Liu et al., 2003a).

Therefore, an increase in the organic loading rate can raise the rate of growth of the biomass, but in turn, can reduce the strength of the structure of the microbial community.

However, the physical characteristics of the aerobic granules, depend on this velocity. Which may affect their morphology, density, specific gravity and volumetric index of the sludge (VIS). The physical strength of aerobic granules may decrease with increasing speed of organic loading. At high loading speeds, it can occur partial loss of the integrity and consequently, the disintegration of its granular structure (Liu et al., 2003b).

Among the bacterial strains that degrade phenol may be mentioned: *Acinetobacter calcoaceticus, Pseudomonas pickettii*l, *Klebsiella oxytoca*, *Ralstonia eutropha, Burkholderia cepacia* G4, *Pseudomonas putida* (Kwon and Yeom, 2009), *Rhodococcus opacus* (Matera et al., 2010), *Alcaligenes* cepa TW1 (Essam et al., 2010). Levaduras como: *Candida tropicalis*, *Rhodotorula rubra* y *Trichosporon cutaneum* (Komarkova et al., 2003).

Algae (*Ochromonas dánica*) and fungus (Actinomicetes, *Nocardia hydrocarbonoxydans*) with activity to biodegrade phenol (Busca et al., 2008).

The application of microbial consortia in biological processes, presents some advantages over pure cultures. Between them, the increase in the degradation rate of phenol, by allowing the bacterial consortium exceed any event that limits the complete biodegradation of phenol (Cordova et al., 2009).

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For its part, the presence of more than one organic compound in the system can affect the functions of biodegradation. Many studies have reported inhibitory effects or increase in phenol degradation systems by adding glucose or phenolic compounds. When placing a co-substrate, it can improve, reduced or not affect the biodegradation of contaminants such as phenol (Bajaj et al., 2008). For example, Arutchelvan et al (2006), observed that by adding dextrose as co-substrate in ranges of 0.2 to the 0.8% (p/v), decreased phenol degradation efficiency. Since dextrose because it is a carbon source of easy biodegradation, the microorganism ignores phenol as a source of carbon, and in consequence, the period of its degradation, was delayed.

Acclimatization of any type of microorganism to the presence of phenol for its biodegradation, is a key factor (Terreros et al., 2016). Yoong (2000) mentions that at concentrations greater than 1300 mg/L of phenol during the treatment of wastewater by activated sludge, provoke complete inhibition of the system.

Silva et al (2002) used an SBR reactor, achieving a phenol biodegradation of the order of 99%.

Bevilaqua (2002) studied a conventional aerobic system coupled to an enzymatic treatment using tyrosinase as an enzyme, and observed degradation performance of the 75% with a residue of 420 mg/L of phenol in a reaction time of 4 hours with 46 Um/L of tyrosine and 50 mg/L of chitosan as coagulant).

Jiang et al (2004) investigated the possibility of treating phenolic wastewater with aerobic granular sludge, which showed an excellent capacity to degrade it to a concentration of 500 mg/L.

Tay et al (2005) used four reactors inoculated with aerobic granules, with feed sequenced with acetate as the main source of carbon, with a mass loading of 3.8 $Kg/m^{3}\cdot d$. Subsequently, the reactors were fed with different loads phenol (0.6, 1.2 and 2.4 $Kg/m^{3}\cdot d$). The reactor whith a load of 0.6 $kg/m^{3}\cdot d$, completely degraded the phenol at 60 min, while the reactor with the charge of 1.2 $Kg/m^{3}\cdot d$, it completely removed in 90 min. But, with 2.4 $Kg/m^{3}\cdot d$ present problems removing it.

Melo et al (2005) studied the biodegradation of phenol in a system of biological reactors; one discontinuous and one rotating, fed at 2 different flow rates: 2 y 4 L/h. Confirming that the performance of the system, improved to increased availability of dissolved oxygen and stirring speed, which favored the transfer coefficient of the oxygen mass, achieving a better degradation of phenol.

Hossein et al. (2006) used a packaged bubble bioreactor for the treatment of phenolic residues, finding 100% of its elimination at a loading rate of 33120 $mg/m^{2}\cdot hr$.

Marrot et al. (2006) studied the biodegradation of high phenol concentration by activated sludge in a membrane bioreactor with phenol concentrations of 0.5 to 3 g/L. Reporting that the activated sludge process cannot stand the phenol at an excessive loading speed.

Bajaj et al. (2008) in a reactor with a cycle of 360 minutes of operation (260 minutes under aerobic conditions and 100 minutes under anoxic conditions). They found a 50% removal of phenol present in synthetic wastewater with an initial concentration of 5.17 g/L of phenol.

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Contreras et al. (2008) used respirometric techniques to study the effect of pH, phenol and dissolved oxygen concentrations during the kinetics of phenol degradation by activated sludge, with a purity of 99%, a hydraulic retention time (HRT) of 24 h and air flow of 2 L/min and dissolved oxygen concentration (DO) above 5 mg/L. Observing that both the pH and the concentration of dissolved oxygen have a significant effect on phenol degradation.

Farooqi et al (2008), studies on biodegradation of phenols and m-cresols by upflow anaerobic sludge blanket and aerobic sequential batch reactor. The results indicates that anaerobic treatment by UASB and aerobic treatment by SBR can be successfully used for phenol/cresol mixture, representative of major substrates in chemical and petrochemical wastewater and the results shows proper acclimatization period is essential for the degradation of m - cresol and phenol. Moreover, SBR was found as a better alternative than UASB reactor as it is more efficient and higher concentration of m cresols can be successfully degraded.

Donoso-Bravo et al. (2009) using synthetic phenol water with a concentration of 5 gCOD/L and a content of; 10, 25 and 40% phenol as carbon source at a concentration of 400 mg/L in ASBR reactors. They found a biodegradation efficiency of the order of 30%.
Chérif et al (2011) studied a model to explain the specific growth rate of microorganisms associated with substrate consumption and the effect of an inhibitor on the degradation of phenol with a purity of 99% in a batch reactor of 2 L of working volume at 25±1°C with a phenol concentration range of 0-793 mg/L, biomass concentration of 0.74 to 6.7 g/L and air flow of 2.5 L/min.

Finding that the model-based design of fed-batch feeding strategies allowed the phenol degradation to be conducted separately under the modes of substrate limitation and substrate inhibition.

Fernandez et al (2013), studied the aerobic biodegradation of a mixture of mono substituted phenols in a sequencing batch reactor. The PNP and o-cresol mixture was also biodegraded although some transitory accumulation of intermediates occurred (mainly hydroquinone and catechol) o chlorophenol was not biodegraded and resulted in inhibition of o-cresol and PNP biodegradation and complete failure of the SBR within a few days. The biomass had very good settling properties when a settling time of 1 min was applied: sludge volume index (SVI_5) below 50 mL/g, SVI_5/SVI_{30} ratio of 1 and average particle size of 200μm.

Athar et al (2015) studied in an SBR reactor, the treatment of synthetic residual water with phenol. Reporting that by increasing the phenol concentration at 2000 mg/L during the acclimatization phase, affects the activity of biomass due to its toxicity.

Inglezakis et al (2016), Studied the inhibitory effect of cyanide, phenol and 4-nitrophenol in a activated sludge reactor, evaluating the degree of inhibition on aerobic oxidation of organic matter, nitrification and desnitrification. Using cyanide, phenol and 4-nitrophenol of 0.2 to 1.7 mg/L, 4.8 to 73.1 mg/L, and of 8.2 to 73.0 mg/L respectively. And they report that was highly toxic cyanide, inhibiting the autotrophic activity of biomass by more than 50%, as well the heterotrophic activity and of the desnitrification, to low concentrations (1.0 a 1.7 $mgCN^-/L$).

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Mentioned that bacteria autotrophic were more sensitive to phenol that the aerobic heterotrophs, while denitrifying bacteria, were more resistant to phenol. In this context, the use of mixed microbial cultures, principally by activated sludge, is an excellent alternative for phenol biodegradation (Badia-Fabegrat et al., 2014; Pradeep, 2015).

The objective of this work was to evaluate the effect of the organic load on the rate of removal of COD and phenol from an industrial effluent by activated sludge, varying the hydraulic retention time, in order to achieve maximum removal.

Materials and methods

Sampling

There were 3 batches of 10 L of phenolic wastewater. The method used for sampling is described in the standard (NMX-AA-003-1980).

Analytical techniques

For the evaluation of the main parameters in the industrial effluent phenol feed (influent) of processed water (effluent) and control system studied, the following analytical techniques were used:

The pH was evaluated by a potentiometer (Conductronic PC18). COD, total solids (TS), volatile solids (VS) were determined according to standard method (APHA, 2016). The determination of phenol, was performed by the colorimetric method of the 4-aminoantipyrine according to the Mexican standard NMX-AA-050-SCFI-2001 using a UV-VIS Spectrometer Perkin Elmer Lambda XLS model computer.

Experimental design

A complete mixing aerobic reactor was used, with a design volume of 4.28 L, useful volume of 3.87L, with dimensions: Length: 28 cm, width of 17.5 cm and depth of 13 cm, divided into 4 sections (A, B , C and D). Section "A", where the biological reaction is carried with dimensions: Length of 18.3 cm, width of 17.5 cm and depth of 13 cm. Section "B" (sludge return zone) to section "A", with a length of 10.2 cm, width of 6.6 cm and depth of 12.4 cm. Section "C" (sedimentation zone) with a length of 7 cm, width of 6.6 cm and depth of 12.3 cm and section "D" (zone of clarification) with a length of 17.5 cm, width of 2.5 cm and depth of 10.4 cm (figure 1), to have a hydraulic behavior in cascade. The system used was equipped with a thermostat to maintain the temperature at 25±0.5°C, fine bubble diffusers and air pump.

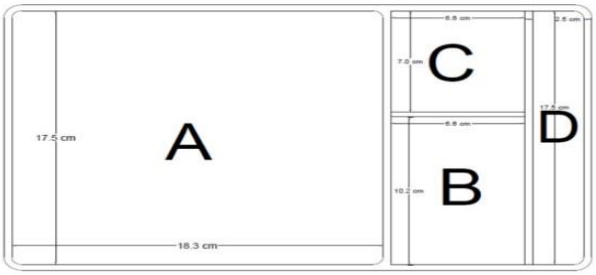

Figure 4 Schematic of the activated sludge reactor

Inoculum

The biomass used as an inoculum was collected from an activated sludge reactor at the Cerro de la Estrella municipal wastewater treatment plant in Mexico City, with a concentration of 12.6 g/L of TS and 9.6 g/L of VS.

Alimentation (influent)

Before analyzing the results, Table 1 shows the averages of the main parameters evaluated in 3 lots of 10 L of phenolic residual water, through calibration curves. A significant dispersion is observed in the data, due to the variability of phenolic wastewater during the different sampling periods.

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Parameter	Experiment		
	I X±S	II X±S	III X±S
COD (g/L)	19.73±3.36	14.04±1.9	9.27±1.7
Phenol (g/L)	2.81±0.35	3.21±0.7	3.18±0.3
pH	6.56±0.19	6.65±0.14	6.66±0.12
TS	0.32±0.2	0.25±0.1	0.26±0.1
VS	0.19±0.1	0.19±0.08	0.17±0.1
X: arithmetic average, S: standard deviation			

Table 1 General characteristics of industrial residual water with phenol (influent)

Operating conditions of the reactor

The table 2 shows the operating conditions of the reactor with continuous feed flow, to the different runs in which the experiment was carried out.

Experiment	I X±S	II X±S	III X±S
Bv (kgCOD/m^3·d)	11.2±0.3	7.97±0.12	3.3±0.18
HRT (days)	1.76	1.76	2.81
Q_{air} (L/min)	2.5	2.5	2.5
X: arithmetic average, S: standard deviation			

Table 2 Operating conditions of the activated sludge reactor

Results and discussion

Characteristics of phenolic wastewater after treatment. The table 3 shows the averages of the main parameters evaluated for the water treated by the activated sludge reactor, during the experiment under the tested operating conditions.

Parameter	Experiment		
	I X±S	II X±S	III X±S
COD (g/L)	13.65±3.16	6.24±1.6	6.08±0.9
Phenol (g/L)	1.67±0.4	2.01±0.37	2.1±0.27
pH	6.31±1.06	6.68±0.28	6.54±0.25
TS	1.07±0.5	0.39±0.1	0.52±0.1
VS	0.72±0.3	0.28±0.1	0.4±0.1
X: arithmetic average, S: standard deviation			

Table 3 General characteristics of treated wastewater (effluent)

Biological degradation of phenol in an activated sludge reactor

The figure 5 shows the behavior of the biomass during phenol degradation. The dark rhombus represent the volatile solids of mixed liquor from the aerobic reactor, which was inoculated with activated sludge from the Cerro de la Estrella municipal wastewater treatment plant in Mexico City, with a concentration of 12.6 g/L TS and 9.6 g/L of VS. During the first days of operation of the reactor, the biomass was acclimated to the phenol degradation of the industrial residual water with a phenol concentration of 0.48±0.025 g/L. However, once it starts with phenolic industrial wastewater feed with a phenol concentration of 2.81±0.35 g/L at an organic load of 11.2±0.3 KgCOD/m^3·d, it is appreciated a substantial loss of biomass from the reactor. Behavior similar to that reported by Buitron et al (2005).

Aerobic granules can be formed in a wide range of organic loading, ranging from 2.5 to 15 KgCOD/m^3·d, increased the size of the aerobic granules of 1.6 to 1.9 mm with organic loads of 3 to 9 Kg COD/m^3·d (Liu et al., 2003a). So the physical characteristics of the aerobic granules depend on the loading speed, which may affect its morphology, density, specific gravity and volumetric index of sludge (VIS). In addition to high speeds of organic loading, it may occur a loss of integrity cell and by consequently, the disintegration of the granular structure (Liu et al., 2003b). Therefore, the behavior of the reactor biomass of activated sludges during their stabilization in contact with phenol, Is contrasted with that reported in the literature. However, from day 36 of operation and throughout the experiment, the biomass showed stable behavior to the extent that the organic load was reduced feeding, with an average concentration of volatile solids of the mixed liquor (VSML) of 1.9±0.36 g/L, sedimentability of 225±5 mL/775mL of supernatant, in an imhoff cone.

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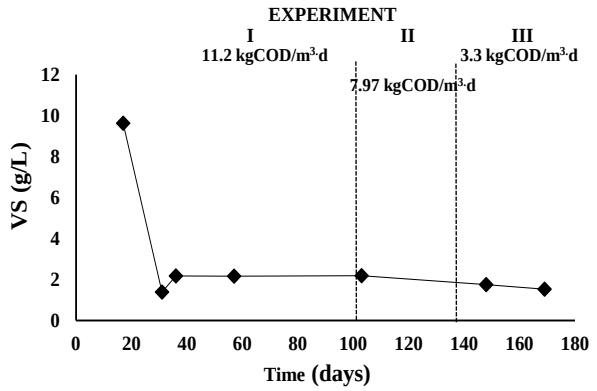

Figure 5 Variation of biomass with time

The figure 6 shows the profile of volatile solids (VS) in the influent and effluent of the reactor during the study to 1.76 days of HRT (experiments I - II) and 2.81 days of HRT (experiment III).

The clear rhombus represent the VS in the influent and the dark rhombu, the VS in the effluent and it is observed that at the moment of starting the feeding of the activated sludge reactor to higher organic load (experiment I), leads to a loss of biomass, due to the degree of phenol toxicity although that in a mixed culture activated sludge has a greater capacity of degradation of this type of toxic compounds, as the phenol (Shalaby, 2003). It has been reported that bacteria of the genus *Bacillus spp*., *Micrococcus spp*. y *Pseudomonas spp*. tolerate concentrations of 10 to 25 g/L of phenol (Yang et al., 2005). *Pseudomonas putida* with a capacity of degradation phenol of 500 to 600 mg/l in 48 hours of incubation. However, once the biomass has acclimated to the presence of phenol as the only source of carbon, was observed from the experiments II and III, a stable behavior, reaching average values 0.34±0.1 g/L. In this context, to avoid loss of biomass from the reactor, in addition to what is mentioned in the literature, on the factors involved in its biodegradation (Trigo, 2009), as temperature, pH, dissolved oxygen, substrate concentration, among others (Agarry, 2008).

As well as the organic loading speeds (Liu et al., 2003a). It is very important to have a biomass acclimatization strategy (Terreros et al., 2016). To allow the increase of the organic load, without affecting the metabolism, Nor the reproduction of the aerobic bacterial consortium, much less cause its cell death (Luo, 2009).

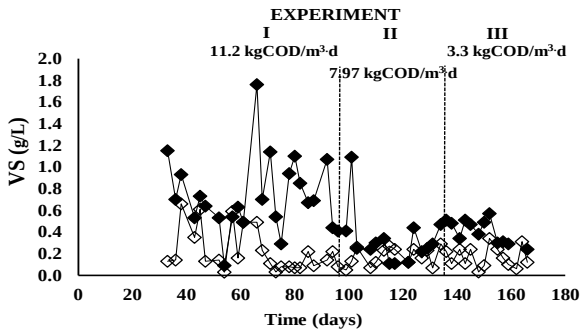

Figure 6 Variation of the concentration of solids with time

The figure 7 shows the performance of pH of the activated sludge reactor. The clear rhombus, represent the pH in the influent and the dark rhombus, the pH of the effluent. It can be seen that during the first 66 days of the experiment I, the pH of the reactor was very unstable reaching pH values of 5.56±0.79, which was probably due to the high organic load and to the toxic effect of the phenol on the bacterial population, perhaps causing the cellular death of a part of the initial biomass with which it was inoculated to the reactor when it was put into operation. And because organic matter is readily biodegradable, its availability as an easily biodegradable substrate caused the pH of the blended liquor to acidify. During the next days of operation, a tendency to rise to reach a mean value of pH of 6.93±1.06 (day 80 of operation). Finally, reach an average pH value of 6.31±1 at the end of this first run. During the experiments (II and III), the average pH values were of the order of 6.68±0.28 and 6.54±0.25 respectively. In response to the acclimatization of the biomass to the phenol of industrial wastewater with which it was fed to the activated sludge reactor throughout the experiment.

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This allowed the experiment to proceed properly, without any inhibitory effect or disturbance affecting reactor performance during phenol biodegradation under the tested operating conditions.

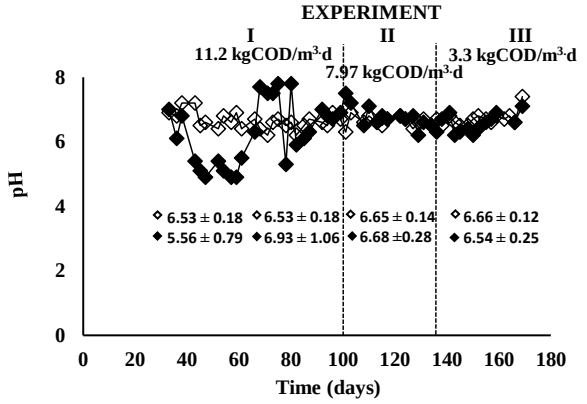

Figure 7 Variation of pH with time

In the figure 8, the efficiency of COD removal is shown, the clear rhombus represent the COD present in the influent and the dark rhombus, the COD in the effluent. It is observed that during the first 73 days of operation, due to the instability of the system, due to the phenol concentration of 2.81±0.35 g/L and organic load of 11.2±0.3 KgCOD/m^{3}·d (experiment I), The removal efficiency of COD was only 21%. However, as the biomass became acclimated to the presence of this toxic compound, an improvement in COD removal was observed in the last day of operation of this run, in a 51%. With the decrease of the organic load in 7.97±0.12 KgCOD/m^{3}·d, but higher phenol concentration (3.21±0.7 g/L) in the industrial residual water in the influent of the reactor (experiment II), a rate of COD removal of 62%. A significant change was observed in COD removal efficiency during the experiment III (34%) in which an organic load of 3.3±0.18 KgCOD/m^{3}·d was tested. The above may be explained as follows, that the biomass being exposed to the phenol of industrial wastewater, for a much longer time (HRT of 2.81 days), relative to the exposure time of the previous runs, its metabolic activity is Was affected, which is reflected in its ability to remove COD.

Because of its toxic nature, partial intoxication can result, and consequently partial loss of floc integrity and consequently the disintegration of its granular structure, thereby reducing its removal efficiency (Liu et al., 2003b).

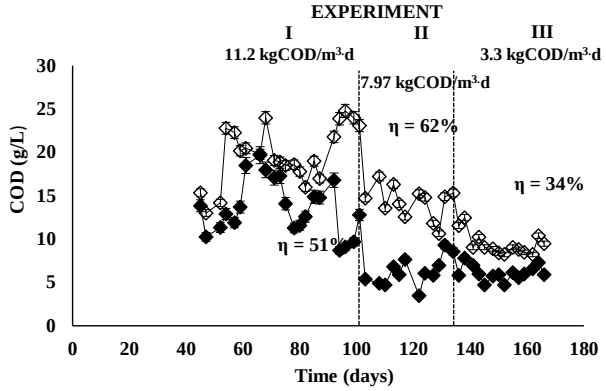

Figure 8 Variation of COD concentration with time

For his part, figure 9 shows the behavior of the reactor on the biodegradation of phenol, to a hydraulic retention time of 1.76 days (experiment I and II) and 2.81 days (experiment III) with an air flow of 2.5 L/min. The clear rhombus represent the phenol present in the influent and the dark rhombus, the phenol remaining in the reactor effluent. During the development of the experiment under the tested operating conditions, it was observed that as the organic load decreased from 11.2±0.3 to 3.3±0.18 KgCOD/m^{3}·d, the rate of phenol biodegradation also decreased. From 48% of phenol biodegradation in the first days of reactor operation, to only 29% phenol biodegradation, at the end of the experiment (experiment III). The decrease in the phenol biodegradation capacity of the mixed culture of the activated sludge reactor, given the toxic nature of phenol and the time of exposure of the bacterial consortium against this toxic compound (HRT of 2.81 days) in relation to that tested in the previous races.

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I present the same effect discussed above, regarding the removal rate of COD, derived from the metabolic affectation of aerobic microorganisms, because of the toxic nature of phenol.

In this context, to achieve an adequate biodegradation of phenol present in industrial wastewater, it is necessary to use the best biomass acclimatization strategy (Terreros et al., 2016), as well as to make dilutions (Ba et al, 2014), in order to achieve its maximum rate of biodegradation, since it is reported in the literature that under aerobic respiration conditions, phenol can become a harmless compound because some aerobic bacteria use it as a source of carbon and energy (Ahmed et al, 2012). The phenol biodegradation results obtained in this study, under the proven operating conditions of the reactor, are in line with what has been reported in the literature. Since at low concentrations of phenol (250 mg/L), it results in 100% degradation. However, as the concentration increases (from 300 to 500 mg/L), the removal rate decreases by 94%, and for concentrations between 800 and 900 mg/L, the degradation profile decreases until reaching values of 93% (Busca et al, 2008), that is, at higher phenol concentration, lower rate of biodegradation.

While at concentrations of more than 1300 mg/L phenol, an inhibitory effect on the biomass is provoked and even, the total loss of the microbial activity can take place (Yoong et al, 2000). With these results, it is possible to treat industrial wastewater with phenol concentrations of 2.8 to 3.2 g/L and organic loading rates in a range of 7 to 11±0.2 Kg COD/m^3·d, at 1.76 days of HRT and 2.5 L/min of air.

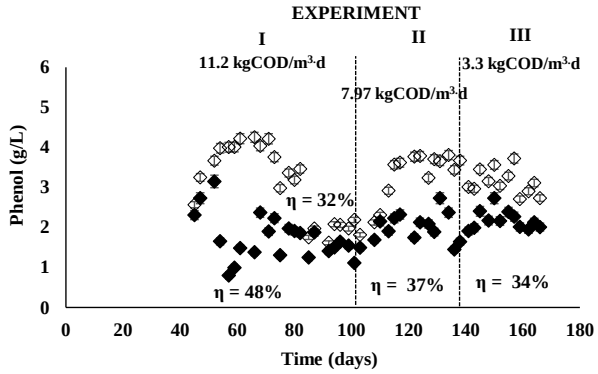

Figure 9 Variation of phenol concentration with time

Conclusions

The strategy used for the acclimatization of biomass from the biological reactor that was used in this research work, was a key factor in achieving biodegradation of phenol during the treatment of phenolic residual water of the resin industry. This allowed the reactor to perform adequately, without the need to use a co-substrate (glucose, fructose, etc), or to conduct encapsulation of the bacterial consortium, as mentioned in the literature, in spite of it, it was possible to biodegrade the phenol present in the wastewater, under the operating conditions of the reactor.

With the results obtained, is demonstrated, that it is possible to treat industrial wastewater with high concentrations of phenol, at high rates of organic load, in relation to the methods traditionally used for this purpose.

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On the other hand, in addition to using the best strategy for the acclimatization of biomass, dilutions are recommended to reduce the effects of inhibition and intoxication of the bacterial population, in order to avoid any disturbance affecting the performance of the biological reactor and achieves, the maximum rate of biodegradation of this type of toxic compounds.

Solving in this way, a real problem of environmental pollution and human health, in addition to reducing the operating costs of the biological reactor during the achievement of said objective.

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