Efﬁciency of an up-ﬂow Anaerobic Sludge Blanket reactor coupled with an electrochemical system to remove chloramphenicol in swine wastewater

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ABSTRACT

The application and design of treatment systems in wastewater are necessary due to antibiotics’ potential toxicity and resistant genes on residual efﬂuent. This work evaluated a coupled bio-electrochemical system to reduce chloramphenicol (CAP) and chemical oxygen demand (COD) on swine wastewater (SWW). SWW characterization found CAP of \(10 \, \mu g/L\) and 17,434 mg/L of COD. The coupled system consisted of preliminary use of an Up-ﬂow Anaerobic Sludge Blanket Reactor (UASB) followed by electrooxidation (EO). The UASB reactor (primary stage) was operated for three months at an organic load of 8.76 kg of COD/m³d and 50 mg CAP/L as initial concentration. In EO, we carried out a 2² (time operation and intensity) factorial design with a central composite design; we tried two Ti cathodes and one anode of Ti/PbO₂. Optimal conditions obtained in the EO process were 240 min of operation time and 1.51 A of current intensity. It was possible to eliminate 44% of COD and 64.2% of CAP in the preliminary stage. On bio-electrochemicals, total COD and CAP removal were 82.35 and \(99.99\%\), respectively. This coupled system can be applied to eliminate antibiotics and other organic pollutants in agricultural, industrial, municipal, and other wastewaters.

Key words: chloramphenicol, degradation, electrooxidation, organic matter, UASB, wastewater

HIGHLIGHTS

• Preliminary application of an anaerobic Reactor (UASB) allowed a reduction of 62.4% initial concentration of chloramphenicol.
• The optimization of the electrooxidation process, for chloramphenicol (CAP) removal, reduces operation time and current intensity.
• UASB + EO combined treatment allowed 82.35 and \(>99.9\%\) of COD and CAP elimination.
• The coupled system proved to be helpful for the CAP treatment in swine wastewater
1. INTRODUCTION

Swine farming is one of the most critical activities for meat production worldwide. With population growth, meat as a source of protein has intensified (Xu et al. 2019). Among factors that have favored the increase in the consumption of pork meat are the accessible price (in comparison with its bovine counterpart) and the increase in consumer confidence towards pork as a healthy source of animal protein (FAO 2018). In 2018, pork production was around 118.8 million metric tons, which is estimated to increase (FAO 2018; Nagarajan et al. 2019). Intensification of this activity will increase the generation of wastewater, which includes solids, organic materials, nitrogen, and other additives such as antibiotics (Cheng et al. 2018). These compounds are widely used in developed countries to treat diseases and promote growth (Kemper et al. 2008; Miyata et al. 2011; Pan et al. 2011; Chen et al. 2017; Cheng et al. 2018). Several studies have mentioned that from doses added, between 50 and 90% is excreted into the environment through wastewater, inducing the prevalence of bacteria and resistant genes (Ben et al. 2008; Pan et al. 2011; Cheng et al. 2018; Wang et al. 2019). China is the largest producer and consumer of veterinary antibiotics globally, with annual use of >84,000 tons (Chen et al. 2017; Guo et al. 2019; Xu et al. 2019). The ranges for the common pharmaceutical pollutants in surface water and treated water are 100 and 50 ng/L, respectively (WHO 2012).

Tetracyclines are antibiotics that have been found in the highest concentration in swine residual effluents (Pan et al. 2011; Chen et al. 2017; Cheng et al. 2018; Wang et al. 2019). Oxytetracycline has been detected in concentrations of 6.18–25.36 μg/L in swine effluents (Chen et al. 2017; Cheng et al. 2018) and raw wastewater (solid + liquid), in concentrations ranging from 1.61 to 20.19 mg/kg (Wang et al. 2019). Likewise, enrofloxacin from <limit detection to 1.09 mg/kg. Antibiotics such as florfenicol and chloramphenicol (CAP), belonging to the phenolic group, have not been detected in this type of effluent. However, CAP concentrations have been found above 28.4 ng/L in urban water supplies in Shanghai, China (Chen et al. 2015), and >0.103 mg/L in aquaculture wastewater effluent in Sonora, Mexico (Molina-Avila 2015). In general, antibiotics concentration and hormones in swine wastewater vary with sampling locations and analysis methods applied (Cheng et al. 2018). The CAP elimination in wastewater effluents is important due to its direct effects on humans, such as aplastic anemia (Zhang et al. 2013). It tends to bioaccumulate in the tissues of animals destined for consumption (Romero-Soto et al. 2018). CAP is a banned
antibiotic in many countries, but it is still used and found in wastewater due to its low cost, which leads to bacterial resistance problems (Tan et al. 2018).

Various efforts have been made to eliminate antibiotics by applying biological treatments, some with low yields because antibiotics kill or inhibit bacterial growth, affecting these systems (Huang et al. 2021). Likewise, eliminating these compounds becomes more complex in the case of wastewater in the presence of high concentrations of organic matter (Huang et al. 2021). Zheng et al. (2018) used a sequential anaerobic digestion system with intermittent aeration for the elimination of 11 antibiotics in swine wastewater, finding 87.9% elimination at a load of 0.17 ± 0.041 kg COD/m³d, of which 30.4% was due to the absorption of sludge and 57.5% to biodegradation. Cheng et al. (2018) used a combined biological filter (aerated/anaerobic) to eliminate nine antibiotics detected in pig wastewater with concentrations up to 0.192 mg/L, reaching eliminations of >82%. On the other hand, the up-flow Anaerobic Sludge Blanket reactor (UASB) is widely used to treat swine wastewater. It can eliminate concentrated effluents of organic matter to high-rate (Torkian et al. 2003; Kim et al. 2013; Pérez-Pérez et al. 2016; Mainardis & Goi 2019; Mainardis et al. 2020; Oliveira et al. 2020; Vassalle et al. 2020) and antibiotics in different grades. Antibiotic removal depends on the initial concentrations, antibiotics classes, types of bioreactors, and operating conditions (Cheng et al. 2018). Sorption and biodegradation are two of the most important mechanisms for eliminating antibiotics from wastewater (Cheng et al. 2018).

Due to the incomplete degradation of antibiotics in the biological phase, it is necessary to add a post-treatment to achieve their mineralization; this post-treatment can be electrooxidation, which is easily automatized and does not require the addition of chemical compounds (Moreira et al. 2017; Garcia-Segura et al. 2018; Romero-Soto et al. 2018). On the electrooxidation process, organic matter is directly oxidized through the -OH radicals that are generated on the anode surface (more than 90%); likewise, other oxidizing agents are generated indirectly, such as: HClO, H₂S₂O₈, H₂O₂, and organic matter is mineralized to CO₂ and H₂O (Drogui et al. 2007; García-Gómez et al. 2014; Romero-Soto et al. 2018). Existing different electrodes materials are used for electrochemical oxidation; notably, the Ti/PbO₂ electrodes are highly efficient, economical and lead release during electrolysis is negligible (Li et al. 2021).

Hou et al. (2019) investigated antibiotics elimination in UASB reactor coupled anoxic-oxic tank and advanced oxidation technologies (UV irradiation, ozonation, Fenton, and Fenton/UV, separately) for the elimination of antibiotics in pharmaceutical wastewater with an organic load of 6.5 kg/m³d. In this work, the elimination of CAP and COD from swine wastewater in the presence of a high load of organic matter (8.76 kg/m³ d) was investigated using a system integrated by a UASB reactor coupled to electrooxidation to achieve complete degradation of the pollutants present. For this reason, the objective of this research was to evaluate the degradation efficiency of CAP and COD by a UASB system coupled to electrooxidation.

2. MATERIALS AND METHODS

2.1. Experimental unit and operation conditions

Preliminary treatment was conducted in an 800 mL cylindrical UASB, made of Plexiglass material, as shown in Figure 1. The dimensions of the anaerobic reactor were: diameter of 7 cm, height of 23 cm, and a conical base of 5 cm in height. For experimental development, the anaerobic reactor was inoculated with 180 g of anaerobic biomass from a region brewing at the south of Sonora, Mexico. A peristaltic pump (Masterflex®) was used to hold a load of 4.38 kg of COD/m³ d (HRT of 35.6 h); during acclimatization for 66 days (at this stage, the reactor was not monitored) and 8.76 kg of COD/m³ d (HRT of 17.8 h) during monitoring (for 97 days). The same batch of water sampled was used in the influent during experimentation to maintain the equal organic load. Likewise, prior COD analyzes were carried out on the influent, and if this decreased, the HRT was adjusted. In the second stage, a cylindrical electrochemical reactor (Figure 1) made of plexiglas with a diameter of 9 cm, height of 22 cm, and a conical base of 5 cm in height with a working volume of 1,000 mL and three mesh electrodes were used. An anode of titanium/lead dioxide (Ti/TiPbO₂) and two cathodes of titanium (Ti) placed cathode-anode-cathode, with an inter-electrode distance of 1 cm, were used. Ti/TiPbO₂ anode was used because it is economical material and can achieve efficiencies as high as the boron-doped diamond (BDD) and other materials such as Ti/SnO₂ have been reported that presented passivation on the anode surface during the oxidation of some organic pollutants (García-Gómez et al. 2014). Electrodes dimensions were all 15 cm in length and 6.5, 4.5, and 2.5 cm in diameter, with 888, 706 and 342 cm² of active surface area, respectively. The current intensity was applied using a BK Precision® of Triple Output DC Power.
Supplies, model 1673 (Yorba Linda, California, USA). The flow was operated by peristaltic pumps (Masterflex®) in ascendant recirculation (45 mL/min). All experiments were conducted at room temperature at 25 ± 2 °C.

2.2. Swine effluent

Swine wastewater was collected in a general effluent from a farm in the north of Obregon city in the state of Sonora, Mexico; it was kept at a temperature of 4 °C before being used. The swine farm does not have a treatment system and the effluents are discharged into the nearest drain. For the UASB reactor, the residual water was characterized previously and enriched with chloramphenicol (firstly diluted and subsequently raw swine wastewater was used). During the acclimatization stage, the solution was prepared using a proportion of 1:2 of swine wastewater and distilled water at pH of 7, enriched with different doses of chloramphenicol (CAP, ≥99.0% Sigma Aldrich, USA) weekly, from 5 to 50 mg/L (during two months) hold a load of 4.38 kg of COD/m³d since its installation. The suspension was agitated with a stirring bar for at least 1 hour to ensure complete dissolution. After the acclimatization, raw wastewater was added to an anaerobic reactor with a CAP concentration of 50 mg/L (intermediate concentrations from Tan et al. (2018) and Chen et al. (2015) studies), a load of 8.76 kg of COD/m³d. The effluent of this stage was collected and preserved at a temperature of 4 °C for later use as an influent of the electrooxidation process (EO) under optimized operating conditions. Samples were collected in the influent and effluent of the biological system and effluent of the EO treatment for further analysis.

2.3. Experimental design for EO

CAP degradation in the EO process with raw wastewater was performed using a response surface methodology (RSM). Treatment time (X₁) and current intensity (X₂) were the independent variables of the model. X₁ and X₂ were selected because preliminary studies were the variables with a major effect on response variables. Likewise, in a real wastewater treatment application, it is difficult to change the temperature and pH to be treated when there are large volumes of water and when working with biological systems; for this reason, these factors were not evaluated (Romero-Soto et al. 2018). For the time range, values below and above the Chen et al. (2015) study were taken with a current intensity between 4 and 7.5 times lower than that investigation (1 A (2.7 mA/cm²) and 2.5 A (6.8 mA/cm²)) to verify if the antibiotic could be removed at a lower current intensity. The domain for X₁ was 60–240 min (U₁,0 = 150 min), and for X₂ was 1–2.5 A (U₁,0 = 1.75 A). Two levels had been assigned to each factor (2² plan), leading to 13 experiments comprised of four runs for the factorial design and nine runs for the central composite design, including five replicates at the center point and four runs for the extreme high and extreme low (Table 1). CAP and COD removal (percentage) and energy consumption were the three investigated responses (response variables). Design Expert® 7 (version 7.0.0) was used to generate the quadratic polynomial model. The optimal operating conditions were applied in the residual effluent of the biological treatment as post-treatment.
CAP removal (R) and energy consumption (E) were calculated using Equations (1) and (2):

\[
R(\%) = \frac{C_0 - C_f}{C_0} \times 100
\]

\[
E = \frac{I \times U \times t}{V} \times 10^{-3}
\]

where \(C_0\) is the initial concentration of CAP (mg/L), \(C_f\) is the final concentration of CAP (mg/L), \(I\) is current intensity (A), \(U\) is electrical potential (V), \(t\) is treatment time (h), and \(V\) treated water volume (m\(^3\)).

The following second-order equation gives the predicted response in all experimental fields (Equation (3)) (García-Gómez et al. 2014; Romero-Soto et al. 2018; Yahiaoui et al. 2018):

\[
Y = b_0 + \sum_{i=1}^{k} b_{i}X_i + \sum_{i=1}^{k} b_{i}^2X_i^2 + \sum_{i=1}^{k} \sum_{j=1}^{k} b_{ij}X_iX_j
\]

where \(Y\) is the experimental response and \(b_0\) is the average of the experimental response.

Coefficients \(b_i\), \(b_{ii}\), and \(b_{ij}\) are the linear, quadratic, and interaction effects between factors \(i\) and \(j\) for the response \(Y\).

### 2.4. Analytical details

Chloramphenicol analysis was performed by HPLC (Agilent™, 1260, USA). Liquid chromatography separation was carried out in a ZORBAX 300-extender-C18 column 4.6 \times 150 mm, 3.5 \(\mu\)m (Agilent™, USA). For CAP, methanol: water, 65:35 (v/v) with a flow rate of 1 mL min\(^{-1}\) at \(\lambda\) 280 and 225 nm in a retention time (RT) of 1.99 min was used. Two mL of sample and 2 mL of buffer solution (methanol: water, 65:35 (v/v)) were mixed, stirred and after 30 min of repose, 1 mL of the supernatant was taken and left to evaporate over 12 hours, the dried sample was reconstituted with 1 mL of methanol HPLC (JT Baker™) and filtered with a membrane of 0.22 \(\mu\)m (Merck Millipore, USA). The antibiotic detection limit was 10 \(\mu\)g/L. Chemical oxygen demand (COD), NO\(_x\) (nitrate NO\(_3\) and nitrite NO\(_2\)), ammonium (NH\(_4^+\)), and orthophosphate (PO\(_4^{3-}\)) were
determined using the standard methods (APHA 1999). For the colorimetric analysis, a brand spectrophotometer (Thermo Scientific® Waltham, MA) was used. The pH was measured by a pH meter (HI 2550, HANNA® Instruments).

3. RESULTS AND DISCUSSION

3.1. Physicochemical characterization of swine wastewater

Swine wastewater was characterized before treatment; Table 2 shows these results. Nutrient concentration in swine wastewater depends on the animal weight, livestock practices (feeding, frequency of cleaning, etc.), and treatment applied before being discharged to receiving sources (Garcia-Sanchez et al. 2016). In this study, the ammonia nitrogen and total COD concentration were 386.76 ± 46 mg-N/L and 17,000 ± 450 mg/L, respectively. The sampled swine farm has a complete cycle; it has bellies, obtains piglets and stallions, and fattens them until sent to the slaughterhouse (closed cycle); which means that there is the presence of different types of organic waste (drugs) and nutrients. Likewise, the high concentrations of organic matter show that the biodigester used to treat the general farm effluent is not working properly (is not wholly under anaerobic conditions and hydraulic residence times have not been established). Then, the farm wastewater effluent is directly deposited in the closest receiving sources without treatment. However, the values obtained are low compared with those found by Zhao et al. (2016), who found 14,000–20,000 mg/L and 2,500–4,000 mg-N/L of COD and total ammonia nitrogen, and the results of Garcia-Sanchez et al. (2016), who reported concentrations in the range of 17,958 ± 1,571 to 20,007 ± 2,478 mg/L of total COD and 656 ± 195 to 1,038 ± 216 mg-N/L of NH₄-N. On the other hand, the presence of antibiotics in swine wastewater has been poorly or not reported. Garcia-Sanchez et al. (2013) detected concentrations of tylosin from 20.2 to 32.4 μg/L in swine wastewater in Mexico. In this study, CAP was not detected; nevertheless, Molina-Avila (2015) found 105 ng of CAP/L in aquaculture wastewater effluent in the south of the Sonora, Mexico, and Chen et al. (2015) reported concentrations of CAP up to 28.4 ng/L in urban water supplies in Shanghai, China. Oxytetracycline was the antibiotic mainly detected, with a concentration of 114 ng/L, which is a kind of tetracycline antibiotic, and had been detected in other studies in a concentration ranging from 2 ng/L to 68 mg/L in stream waters in a small catchment area with livestock farms (Matsui et al. 2008). This study focused on CAP only because this antibiotic is already prohibited in many countries due to its adverse health effects, such as aplastic anemia, and because it is bioaccumulative and has been found in the tissues of animals destined for human consumption (Zhang et al. 2013; Romero-Soto et al. 2018; Tan et al. 2018). CAP is still used in aquaculture and pig farms due to its low cost, and harmful concentrations of this antibiotic have been reported in aquacultural and swine farm effluents, increasing the presence of resistant bacteria (Chen et al. 2015; Romero-Soto et al. 2018).

3.2. Statistical analysis

CAP degradation in raw wastewater by EO was evaluated using a central composite design. The central composite matrix explores the whole experimental domain and determines the coefficients of a mathematical second-order polynomial equation. There are two quantitative variables: treatment time (U₁, min) and current intensity (U₂, A), which control the amount of •OH radicals and other oxidants produced in the reactor (García-Gómez et al. 2014; Chen et al. 2015; Table 2 | Physicochemical parameters in swine wastewater

| Parameters | (mg/L) |
|------------|--------|
| Ammonium   | 386.76 |
| Phosphate  | 0.60   |
| Nitrite    | 47.23  |
| Total COD  | 17,434 |
| **Antibiotics** |      |
| Chloramphenicol* | <0.01 |
| Oxytetracycline | 0.114 |
| Enrofloxacin  | 0.084 |
| Florfenicol  | 0.030  |

*Detection limit 0.01 mg/L.
Romero-Soto et al., 2018). CAP removal ($Y_1, \%$), COD removal ($Y_2, \%$), and energy consumption ($Y_3, \text{kWh/m}^3$) were the dependent factors (responses). Thirteen experiments were carried out and the results are given in Table 1. These tests were comprised of five experiments carried out at the center of the experimental domain (central point), four experiments corresponding to the factorial design ($2^3$), and four other experiments carried out around the experimental field (low and high extremities).

The corresponding second-order polynomial equations models are given by Equations (4)–(6) for CAP and COD removal and energy consumption, respectively:

$$Y_1 = 83.05 + 17.21X_1 + 12.20X_2 - 4.84X_1X_2 - 3.69X_1^2 - 2.26X_2^2$$  \hspace{1cm} (4)

$$Y_2 = 59.14 + 10.30X_1 + 4.11X_2 + 1.42X_1X_2 + 5.42X_1^2 + 1.30X_2^2$$  \hspace{1cm} (5)

$$Y_3 = 17.32 + 10.65X_1 + 9.34X_2 + 5.70X_1X_2 - 0.019X_1^2 + 0.91X_2^2$$  \hspace{1cm} (6)

Coefficients of equation models were calculated using the half difference between the arithmetic average of the response values when the variable is code at the levels $-1$ and $+1$ (García-Gómez et al., 2014; Romero-Soto et al., 2018). Thus, a positive coefficient will positively affect the response, while a negative coefficient will have a negative effect on the response.

Coefficient $b_o = 83.05$ represents the average CAP removal obtained from all the experiments. The coefficient $b_1 = +17.21$ corresponding to the operation time ($X_1$) indicates that the CAP removal increased on average by 34.42% ($2 \times 17.21$) when the experimental time varied from 60 to 240 min; likewise, it is the most influential variable on the antibiotic removal. The second one is the current intensity ($X_2$). According to coefficient $b_2 = +12.20$, the elimination of the antibiotic increased on average by 24.40% ($2 \times 12.20$) when the intensity passed from 1 to 2.5 A.

For the COD removal, $b_o = 17.32$ was the average removal obtained from all the experiments. The coefficient $b_1 = +10.30$ indicates only 20.60% COD removal increases when the experimental time varied from 60 to 240 min, and $b_2 = +4.11$ suggests that 8.22% of the organic matter is eliminated when the current increased from 1 to 2.5 A.

For energy consumption, $b_o = 39.14$ was the average energy consumed in all the experiments. The coefficient $b_1 = +10.65$ indicates that 21.3 kWh/m$^3$ of energy consumption increases when the experimental time is varied from 60 to 240 min; while $b_2 = +9.34$, means an energy consumption of 18.68 kWh/m$^3$ when the current increased from 1 to 2.5 A.

The CAP and COD removal and energy consumption is shown by the response surface plot depicted in Figure 2. As observed, the removals could reach $\geq 99.99\%$ if the times in both response variables increased. The maximum reduction achieved was 99.75 and 67.84% and the minimum was 48.26 and 29.012% for the CAP and COD removal, respectively. The best result for CAP removal (99.75%) was: 150 min and 2.81 A with an energy consumption of 32.32 kWh/m$^3$. In comparison, the removal of COD (67.84%) was at 240 min and 2.5 A with an energy consumption of 44 kWh/m$^3$ (experiment 13 and 4, Table 2). Analysis of variance (ANOVA), given in Table 3, shows that CAP F-value of 37.11 implies the model is significant. On CAP removal A and B are substantial variables because the $p$-value is <0.0001, and its F value is more meaningful (58.57 and 116.62) than the F value of the model (37.11). On COD removal, only time (B) is significant because the $p$-value is 0.0022 (<0.05), and the F value (22.09) is higher than the F value of the model (6.24). Intensity (A) is not significant ($p$-value <0.05). On energy consumption, A and B are significant, $p$-value is <0.0001.

The coefficient correlation value ($R^2 = 0.9636$) means that the empirical model could not explain only 3.64% of the total variation. 'Prob > F' less than 0.05 indicates model terms are significant. A low dispersion of data observed in Figure 3(a)

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**Figure 2** | (a) Effect of the experimental time and current in the CAP removal, (b) effect of electrolysis time and current intensity on COD removal, and (c) effect of variables evaluated in energy consumption with Ti cathodes and a Ti/PbO$_2$ anode.
means a good fit of the removals obtained and the predicted. For COD removal, the coefficient correlation value ($R^2 = 0.8168$) implies that the empirical model could not explain 18.32% of the total variation, which means the Lack of Fit is significant. $R^2$ should be at least 0.80 for a good fit of a model (Joglekar & May 1987). High dispersion of the data is observed in Figure 3(b) due to the low correlation coefficient obtained. For energy consumption, the F-value of 2485.38 implies the model is significant. There is only a 0.01% chance that an F-value this large could occur due to noise. The coefficient correlation value ($R^2 = 0.9994$) means that the empirical model could not explain 0.06% of the total variation. It implies low dispersion of data, observed in Figure 3(c), which means a good fit for the obtained and predicted energy consumption.

### 3.3. Optimization of electrochemical system

The importance of the variables and their interactions was calculated using Pareto analysis. This analysis estimates the effect (percentage) of each variable studied on the response (García-Gómez et al. 2014). For this study, treatment time was the most
significant contribution to the CAP and COD removal with 75.16%, followed by the current intensity with 18.70%. The high contribution for the time and current intensity is due to these parameters controlling hydroxyl radicals and other oxidants produced during the experimentation, such as HClO, H2S2O8, H2O2, etc. (Drogui et al. 2007; García-Gómez et al. 2014; Moreira et al. 2017; Romero-Soto et al. 2018). Chemical reactions are shown in Equations (7)–(9):

\[
\begin{align*}
\text{Cl}^- + 2\text{H}_2\text{O} & \rightarrow \text{HClO} + \text{H}_2\text{O}^+ + 2e^- \\
2\text{SO}_4^{2-} + 2\text{H}^+ & \rightarrow \text{H}_2\text{S}_2\text{O}_8 + 2e^- \\
\text{O}_2^{(\text{dissolved})} + 2\text{H}^+ + 2e^- & \rightarrow \text{H}_2\text{O}_2
\end{align*}
\] (7)

A particular compromise was established to determine the optimum conditions of the process, CAP and COD removal, and the energy consumption (priority for CAP removal, followed by COD elimination and finally reduction energy consumption). The objective is more likely to maximize the CAP and COD removal than to minimize energy consumption. For optimization, there are five levels of importance in the statistical package (Design-Expert software), which were selected based on the order of priority of the response variables. Five points were assigned to CAP degradation, four to COD degradation, and three to energy consumption. Based on these preferences and other parameters of the model, the Design-Expert software could generate the following optimum conditions: treatment time = 240 min and current intensity = 1.51 A. The predicted CAP and COD removal and energy consumption were 92.64 and 52.23%, and 23.16 kWh/m3, respectively. Therefore, the overall desirability for this work is 0.667 as all responses are predicted to be within the desired limits. These conditions were applied in the effluent of the biological system.

There are different works where this technology has been applied to remove other contaminants (Table 4). CAP and COD removals achieved are high; however, the time and energy consumption is required too. Because swine wastewater had high organic matter concentration, which competes in removing other compounds, this proves that an increase in organic load may negatively impact antibiotic reduction (McAdam et al. 2011; Tran et al. 2013; Yoo et al. 2020).

### 3.4. Degradation of CAP and COD in the coupled biological-electrochemical system

A UASB reactor was used as a pretreatment, which was monitored for three months after acclimatization. The porcine wastewater was treated through an anaerobic stage and after by electrooxidation under optimal conditions (240 min and 1.51 A); this coupling was made to eliminate the high organic matter concentration found in the studied effluent using a UASB reactor followed by an electrooxidation system to complete antibiotics oxidation. Figure 4 shows the removals achieved by the UASB reactor after 97 days of operation. The initial concentration of CAP was 50 mg/L and 17,000 ± 75 mg/L of COD. CAP concentration in this study was high; however, we wanted to verify that biological systems could cope with high loads of antibiotics and organic matter and because these compounds were in the order of several mg/L in effluents of hospitals, pharmaceutical production, and aquaculture farms (Kummerer 2001; Chelliapan et al. 2006; Meng et al. 2015; Hou et al. 2019). This combined system can be used to treat other wastewater effluents.

The maximum COD removal in the UASB reactor was 44 ± 1.2% at an organic load of 8.76 kg of COD/m3d (shown in Figure 4), which is low compared with the results found by Zhao et al. (2016), who reached 83.6% in swine wastewater at organic load (VOL) of 2.29 kg/m3d (almost four times less than this work). Lo et al. (1994) obtained removal efficiency of COD from 95% in swine manure at a VOL of 1.65 kg/m3d. However, the COD removal decreased to 57% at VOL of 3.5 kg/m3d. Campos et al. (2005), with the same configuration at 20 h HRT and VOL of 1.42 kg COD/m3d, obtained removal efficiency of COD of 84.0%. The low efficiency obtained in the biological system may be because only 60% is a biodegradable matter (Andreadakis 1992). Likewise, a high concentration of ammonia can inhibit the activity of microorganisms and hence hinder the maximum organic loading rates in the anaerobic processes (Zhao et al. 2016). García-Sanchez et al. (2016) mentioned that the presence of antibiotics does not affect COD removal if the reactor is acclimated. Zaiat et al. (2001) and Sanchez et al. (2005) recommended increasing the solids retention time (SRT) to improve the organic matter removal due to the UASB not being suitable for the treatment of pig manure, based on poor yield for low HRT and high VOL. The main mechanisms of removal of veterinary antibiotics in anaerobic conditions are sludge sorption and biodegradation. Greater than 60% of antibiotics in the influent are biodegraded, 24% are adsorbed by sludge, and 15% of the antibiotics remain in the effluent (Zheng et al. 2018). The combined system of UASB and EO reached 82.35 and >99.9% of COD and CAP removal, respectively (shown in Figure 5). No studies have coupled this type of biological system with EO to
Table 4 | Summary of the previously reported results for the electrooxidation of antibiotics

| System treatment- Anode | Drug removal       | Concentration (mg/L) | Conditions                | Effluent                  | Removal obtained (%) | References                  |
|-------------------------|--------------------|----------------------|---------------------------|---------------------------|----------------------|-----------------------------|
| Electrooxidation and Ti/PbO₂ | Chloramphenicol (CAP) | 0.5                  | 0.65 A and 34 min         | Aquaculture wastewater    | <99.9                | Romero-Soto <i>et al.</i> (<2018)>|
| Electrooxidation and Ti/PbO₂ | Oxytetracycline OXY | 100                  | 30 mA/cm² and 6 h         | Synthetic water           | 98                   | Nunes <i>et al.</i> (<2016>) |
| Electrooxidation and Al-doped PbO₂ | CAP               | 100                  | 2.5 h and 30 mA/cm²       | Synthetic water           | 87.30                | Chen <i>et al.</i> (<2015>)   |
| Electrooxidation and BDD (Boron doped diamond) electrodes | Tetracycline (TTC) | 150                  | 3 A and 4 h               | Synthetic water           | >99                  | Brinzila <i>et al.</i> (<2012>) |
| Electrooxidation and Ti/PbO₂ | OXY                | 100                  | after 6 h, using 1.5 A    | Raw milk                  | 95                   | Kitazono <i>et al.</i> (<2012>) |
| Electrooxidation and Ti/PbO₂ | (OXY)              | 100                  | 1.5 A and 2 h             | Livestock wastewater      | >99.9                | Miyata <i>et al.</i> (<2011>) |
| Electrooxidation and Ti/RuO₂ | TTC               | 50 and 200           | 47.6 mA/cm², pH 3.9 and 60 min | Deionized water           | 89.1 and 82.2, respectively | Zhang <i>et al.</i> (<2009>) |
eliminate antibiotics in swine wastewater. However, the most significant degradation of organic matter was carried out in the biological stage; likewise, the CAP was eliminated mainly during the EO because microorganisms tend to utilize the easily degradable organic substances rather than the refractory antibiotics maintaining their metabolism (McAdam et al. 2011; Zheng et al. 2018). The perspectives of the combined system are that it can be applied in the elimination of antibiotics

**Figure 4** Monitoring of the UASB reactor during 97 days operation after the acclimatization with a load of 8.76 kg of COD/m³d and 50 mg of CAP/L. •• removal and reduction of COD, …… removal and reduction of CAP.

**Figure 5** Overall removal of COD and CAP in a combined UASB reactor–EO system.
and other pollutants in wastewater effluents of different activities with high organic loads such as aquaculture, pig farm, pharmaceutical, hospital, and others. Likewise, the coupling of the UASB system with an electrochemical reactor can contribute to reducing energy consumption through the use of biogas produced in the anaerobic digester. In addition, UASB allows reducing organic loads before being treated in an electrochemical system and is recommended to use two UASB reactors before the electrochemical system if the wastewater has a COD $\geq 6,500$ mg/L, as in this study. Another alternative to reduce energy consumption in advanced treatment is solar panels use as an energy supply, especially in zones with a larger area such as aquaculture and pig farms.

4. CONCLUSION

A coupled biological-electrochemical system for the elimination of CAP and COD in swine wastewater was used. The swine effluent sampled presented a CAP concentration below to limit detection, for which the antibiotic was added (50 mg/L of CAP). The UASB reactor (at an organic load of 8.76 kg of COD/m$^3$ d) reached only 44 and 64.2% of COD and CAP removal. Likewise, the bio-electrochemical combination system increased elimination to 82.35 and $>99.9\%$ of COD and CAP, respectively (using optimal conditions in EO process: treatment time $= 240$ min and current intensity $= 1.51$ A). Electrolysis time and current intensity are more significant variables in this type of effluent treatment due to the high organic matter. The methane capture from the anaerobic system and its conversion into electricity can be applied to an electrochemical system to reduce operating costs. This study underlines the effectiveness of UASB and electrooxidation for chloramphenicol removal in swine wastewater. It could treat other wastewater types such as municipal, agriculture, aquaculture, and industrial.

DATA AVAILABILITY STATEMENT

All relevant data are included in the paper or its Supplementary Information.

REFERENCES

Andreadakis, A. D. 1992 Anaerobic digestion of piggery wastes. Water Science and Technology 25, 9–16.
APHA 1999 Standard Methods for the Examination of Water and Wastewater 20th. Public Health Association, Washington, DC, USA.
Ben, W., Qiang, Z., Adams, C., Zhang, H. & Chen, L. 2008 Simultaneous determination of sulfonamides, tetracyclines and tiamulin in swine wastewater by solid-phase extraction and liquid chromatography-mass spectrometry. Journal of Chromatography A 1202 (2), 173–180.
Brinzila, C. I., Pacheco, M. J., Cifra, L, Ciobanu, R. C. & Lopes, A. 2012 Electrodegradation of tetracycline on BDD anode. Chemical Engineering Journal 209, 54–61.
Campos, C. M., Damasceno, L. H., Machizuki, E. T. & Botelho, C. G. 2005 Evaluation of the performance of the anaerobic sludge blanket reactor (UASB) at laboratory scale in the removal of the organic load of swine wastewater. Ciência e Agrotecnologia 29, 390–399.
Chellapan, S., Wilby, T. & Sallis, P. J. 2006 Performance of an up-flow anaerobic stage reactor (UASR) in the treatment of pharmaceutical wastewater containing macrolide antibiotics. Water Resources 40 (3), 507–516.
Chen, J., Xia, Y. & Dai, Q. 2015 Electrochemical degradation of chloramphenicol with a novel Al doped PbO2 electrode: performance, kinetics and degradation mechanism. Electrochimica Acta 165, 277–287.
Chen, J., Liu, Y. S., Zhang, J. N., Yang, Y. Q., Hu, L. X., Yang, Y. Y., Zhao, J. L., Chen, F. R. & Ying, G. G. 2017 Removal of antibiotics from piggery wastewater by biological aerated filter system: treatment efficiency and biodegradation kinetics. Bioresource Technology 238, 70–77.
Cheng, D. L., Ngo, H. H., Guo, W. S., Liu, Y. W., Zhou, J. L., Chang, S. W., Nguyen, D. D., Bui, X. T. & Zhang, X. B. 2018 Bioprocessing for elimination antibiotics and hormones from swine wastewater. Science of the Total Environment 621, 1664–1682.
Drogui, P., Blais, J. A. & Mercier, G. 2007 Review of electrochemical technologies for environmental applications. Recent Patents on Engineering 1 (3), 257–272.
FAO 2018 World Production and Trade of Pork in 2018. FAO, Rome, Italy.
García-Sanchez, L., Garzon-Zuniga, M. A., Buelna, G., Moeller-Chavez, G. E., Noyola, A., Aviléz-Flores, M. & Estrada-Arriaga, E. B. 2013 Occurrence of tylosin in swine wastewater in Mexico. Water Science and Technology 68 (4), 894–900.
García-Gómez, C., Drogui, P., Zaviska, F., Seyhi, B., Gortáres-Moroyoqui, P., Buelna, G., Neira-Sáenz, C., Estrada-alvarado, M. & Ulloa-Mercado, R. G. 2014 Experimental design methodology applied to electrochemical oxidation of carbamazepine using Ti/PbO2 and Ti/BDD electrodes. Journal of Electroanalytical Chemistry 732, 1–10.
García-Sanchez, L., Garzon-Zuniga, M. A., Buelna, G. & Estrada-Arriaga, E. B. 2016 Tylosin effect on methanogenesis in an anaerobic biomass from swine wastewater treatment. Water Science and Technology 75 (2), 445–452.
Garcia-Segura, S., Ocon, J. D. & Chong, M. N. 2018 Electrochemical oxidation remediation of real wastewater effluents – a review. Process Safety and Environmental Protection 115, 48–67.
Guo, N., Ma, X., Ren, S., Wang, S. & Wang, Y. 2019 Mechanisms of metabolic performance enhancement during electrically assisted anaerobic treatment of chloramphenicol wastewater. Water Resources 156, 199–207.
Hou, J., Chen, Z., Gao, J., Xie, Y., Li, L., Qin, S., Wang, Q., Mao, D. & Luo, Y. 2019 Simultaneous removal of antibiotics and antibiotic resistance genes from pharmaceutical wastewater using the combinations of up-flow anaerobic sludge bed, anoxic-oxic tank, and advanced oxidation technologies. *Water Resources* 159, 511–520.

Huang, A., Yan, M., Lin, J., Xu, L., Gong, H. & Gong, H. 2021 A review of processes for removing antibiotics from breeding wastewater. *International Journal of Environmental Resource and Public Health* 18 (9), 4909.

Joglekar, A. & May, A. T. 1987 Product excellence through design of experiments. *Cereal Foods World Journal* 32, 857–858.

Kemper, N., Färber, H., Skutlarek, D. & Krieter, J. 2008 Analysis of antibiotic residues in liquid manure and leachate of dairy farms in Northern Germany. *Agricultural Water Management* 95 (11), 1288–1292.

Kim, J., Kim, W. & Lee, C. 2013 Absolute dominance of hydrogenotrophic methanogens in full-scale anaerobic sewage sludge digesters. *Journal of Environmental Sciences* 25 (11), 2272–2280.

Kitazono, Y., Ihara, I., Yoshida, G., Toyoda, K. & Umetu, K. 2012 Selective degradation of tetracycline antibiotics present in raw milk by electrochemical method. *Journal of Hazardous Materials* 245, 112–116.

Kummerer, K. 2001 Drugs in the environment: emission of drugs, diagnostic aids and disinfectants into wastewater by hospitals in relation to other sources—a review. *Chemosphere* 45, 957–969.

Li, J., Guo, M., Shao, Y., Yu, H. & Ni, K. 2021 Electrocatalytic properties of a novel β-PbO2/Halloysite nanotube composite electrode. *ACS Omega* 6 (8), 5436–5444.

Lo, K. V., Liao, H. & Gao, Y. C. 1994 Anaerobic treatment of swine wastewater using hybrid UASB reactors. *Bioresource Technology* 47, 153–157.

Mainardis, M. & Goi, D. 2019 Pilot-UASB reactor tests for anaerobic valorisation of high-loaded liquid substrates in friulian mountain area. *Journal of Environmental Chemical Engineering* 7 (5), 103548.

Mainardis, M., Buttazzoni, M. & Goi, D. 2020 Up-flow anaerobic sludge blanket (UASB) technology for energy recovery: a review on state-of-the-art and recent technological advances. *Bioengineering (Basel)* 7 (2), 43.

Matsui, Y., Ozu, T., Inoue, T. & Matsushita, T. 2008 Occurrence of a veterinary antibiotic in streams in a small catchment area with livestock farms. *Desalination* 226 (1–3), 215–221.

McAdam, E. J., Bagnall, J. P., Soares, A., Koh, Y. K. K., Chiu, T. Y., Lester, J. N. & Cartmell, E. 2011 Fate of alkylphenolic compounds during activated sludge treatment: impact of loading and organic composition. *Environmental Science & Technology* 45 (1), 248–254.

Meng, L.-W., Li, X.-K., Wang, K., Ma, K.-L. & Zhang, J. 2015 Influence of the amoxicillin concentration on organics removal and microbial community structure in an anaerobic EGSB reactor treating with antibiotic wastewater. *Chemical Engineering Journal* 274, 94–101.

Miyata, M., Ihara, I., Yoshid, G., Toyod, K. & Umetu, K. 2011 Electrochemical oxidation of tetracycline antibiotics using a Ti/IrO2 anode for wastewater treatment of animal husbandry. *Water Science and Technology* 65 (3), 456–461.

Molina-Avila, B. G. 2015 *Caracterización de agua residual acuícola: Parámetros fisicoquímicos y antibióticos*. BS Thesis, Inst. Tecnol., Sonora, Obregón, México.

Moreira, F. C., Boaventura, R. A. R., Brillas, E. & Vilar, V. J. P. 2017 Electrochemical advanced oxidation processes: a review on their application to synthetic and real wastewaters. *Applied Catalysis B: Environmental* 202, 217–261.

Nagarajan, D., Kusmayadi, A., Yen, H. W., Dong, C. D., Lee, D. J. & Chang, J. S. 2019 Current advances in biological swine wastewater treatment using microalgae-based processes. *Bioresource Technology* 289, 121178.

Nunes, M. J., Monteiro, N., Pacheco, M. J., Lopes, A. & Ciriaco, L. 2016 Ti/beta-PbO2 versus Ti/PT/beta-PbO2: influence of the platinum interlayer on the electrodegradation of tetracyclines. *Journal of Environmental Science and Health, Part A* 51 (10), 839–846.

Oliveira, J. F., Fia, R., Fia, F. R. L., Rodrigues, F. N., Matos, M. P. & Siniscalchi, L. A. B. 2020 Principal component analysis as a criterion for monitoring variable organic load of swine wastewater in integrated biological reactors UASB, SABF and HSSF-CW. *Journal of Environmental Management* 262 (1–3), 215–221.

Pan, X., Qiang, Z., Ben, W. & Chen, M. 2011 Residual veterinary antibiotics in swine manure from concentrated animal feeding operations in Shandong Province, China. *Chemosphere* 84 (5), 695–700.

Pérez-Pérez, T., Pereda-Reyes, I., Oliva-Merencio, D. & Zaiat, M. 2016 Anaerobic digestion technologies for the treatment of pig wastes. *Cuban Journal of Agricultural Science* 50, 343–354.

Romero-Soto, I. C., Dia, O., Leyva-Soto, L. A., Drogui, P., Buelna, G., Diaz-Tenorio, L. M., Ulloa-Mercado, R. G. & Gortares-Moroyoqui, P. 2018 Degradation of chloramphenicol in synthetic and aquaculture wastewater using electrooxidation. *Journal of Environmental Quality* 47 (4), 805–811.

Sanchez, E., Borja, R., Travieso, L., Martin, A. & Colmenarejo, M. F. 2005 Effect of organic loading rate on the stability, operational parameters and performance of a secondary upflow anaerobic sludge bed reactor treating piggy waste. *Bioresource Technology* 96 (3), 335–344.

Tan, C., Dong, Y., Fu, D., Gao, N., Ma, J. & Liu, X. 2018 Chloramphenicol removal by zero valent iron activated peroxymonosulfate system: kinetics and mechanism of radical generation. *Chemical Engineering Journal* 334, 1006–1015.

Torkian, A., Eqbali, A. & Hashemian, S. J. 2003 The effect of organic loading rate on the performance of UASB reactor treating slaughterhouse effluent. *Resources, Conservation and Recycling* 40 (1), 1–11.

Tran, N. H., Urase, T., Ngo, H. H., Hu, J. & Ong, S. L. 2013 Insight into metabolic and cometabolic activities of autotrophic and heterotrophic microorganisms in the biodegradation of emerging trace organic contaminants. *Bioresource Technology* 146, 721–731.
Vassalle, L., Diez-Montero, R., Machado, A. T. R., Moreira, C., Ferrer, I., Mota, C. R. & Passos, F. 2020 Upflow anaerobic sludge blanket in microalgae-based sewage treatment: co-digestion for improving biogas production. *Bioresource Technology* 300, 122677.

Wang, R., Feng, F., Chai, Y., Meng, X., Sui, Q., Shen, M., Wei, Y. & Qi, K. 2019 Screening and quantitation of residual antibiotics in two different swine wastewater treatment systems during warm and cold seasons. *Science of the Total Environmental* 660, 1542–1554.

WHO 2012 *Pharmaceuticals in Drinking-Water*. World Health Organization (Water pollutants), Geneva, Switzerland, p. 8.

Xu, Z., Song, X., Li, Y., Li, G. & Luo, W. 2019 Removal of antibiotics by sequencing-batch membrane bioreactor for swine wastewater treatment. *Science of the Total Environmental* 684, 23–30.

Yahiaoui, I., Yahia Cherif, L., Madi, K., Aissani-Benissad, F., Fourcade, F. & Amrane, A. 2018 The feasibility of combining an electrochemical treatment on a carbon felt electrode and a biological treatment for the degradation of tetracycline and tylosin – application of the experimental design methodology. *Separation Science and Technology* 53 (2), 337–348.

Yoo, C. G., Meng, X., Pu, Y. & Ragauskas, A. J. 2020 The critical role of lignin in lignocellulosic biomass conversion and recent pretreatment strategies: a comprehensive review. *Bioresource Technology* 301, 122784.

Zaiat, M., Rodrigues, J. A., Ratusznei, S. M., Camargo, E. F. & Borzani, W. 2001 Anaerobic sequencing batch reactors for wastewater treatment: a developing technology. *Applied Microbiology and Biotechnology* 55, 29–35.

Zhang, H., Liu, F., Wu, X., Zhang, J. & Zhang, D. 2009 Degradation of tetracycline in aqueous medium by electrochemical method. *Asia-Pacific Journal of Chemical Engineering* 4 (5), 568–573.

Zhang, W., Sun, W., An, S., Xiong, B., Lin, K., Cui, X. & Guo, M. 2013 Acute and chronic toxic effects of chloramphenicol on Scenedesmus obliquus and Chlorella pyrenoidosa. *Water Environment Research* 85 (8), 725–732.

Zhao, B., Li, J., Buelna, G., Dube, R. & Le Bihan, Y. 2016 A combined upflow anaerobic sludge bed and trickling biofilter process for the treatment of swine wastewater. *Environmental Technology* 37 (10), 1265–1275.

Zheng, W., Zhang, Z., Liu, R. & Lei, Z. 2018 Removal of veterinary antibiotics from anaerobically digested swine wastewater using an intermittently aerated sequencing batch reactor. *Journal of Environmental Science (China)* 65, 8–17.

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