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Degradation of the antibiotic amoxicillin by photo-Fenton process – Chemical and toxicological assessment

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ABSTRACT

The influence of iron species on amoxicillin (AMX) degradation, intermediate products generated and toxicity during the photo-Fenton process using a solar simulator were evaluated in this work. The AMX degradation was favored in the presence of the potassium ferrioxalate complex (FeOx) when compared to $FeSO_4$. Total oxidation of AMX in the presence of FeOx was obtained after 5 min, while 15 min were necessary using $FeSO_4$. The results obtained with *Daphnia magna* biossays showed that the toxicity decreased from 65 to 5% after 90 min of irradiation in the presence of $FeSO_4$. However, it increased again to a maximum of 100% after 150 min, what indicates the generation of more toxic intermediates than AMX, reaching 45% after 240 min. However, using FeOx, the inhibition of mobility varied between 100 and 70% during treatment, probably due to the presence of oxalate, which is toxic to the neonates. After 240 min, between 73 and 81% TOC removal was observed. Different pathways of AMX degradation were suggested including the opening of the four-membered β-lactamic ring and further oxidations of the methyl group to aldehyde and/or hydroxylation of the benzoic ring, generating other intermediates after bound cleavage between different atoms and further oxidation to carboxylates such acetate, oxalate and propionate, besides the generation of nitrate and ammonium.

1. Introduction

The presence of pharmaceuticals in the environment has been reported as an emerging risk to the biotic environment. Among the pharmaceuticals, special attention is focused on antibiotics, since bacterial resistance and toxic effects on several organisms such as algae and crustaceous have been found not only at high concentrations, but also at low concentrations resulting in chronic effects (Andreozzi et al., 2004; Foti et al., 2009). The antibiotic AMX, a broad spectrum aminopenicillin antibiotic, widely used in human and veterinary medicine, was tested for toxicity on microalgal species (growth inhibition) and found to be nontoxic to *Pseudokirkneriella subcapitata* e *Closterium ehrenbergii* (EC_{50} 100 mg L^{-1}), but showed marked toxicity to the *Synechococcus leopolensis* (EC_{50} 2 μg $L^{-1)}$ (Andreozzi et al., 2004). This compound has been identified in municipal sewage treatment plant effluent at concentration of 13 ng L^{-1} in Italy (Castiglioni et al., 2006). So,

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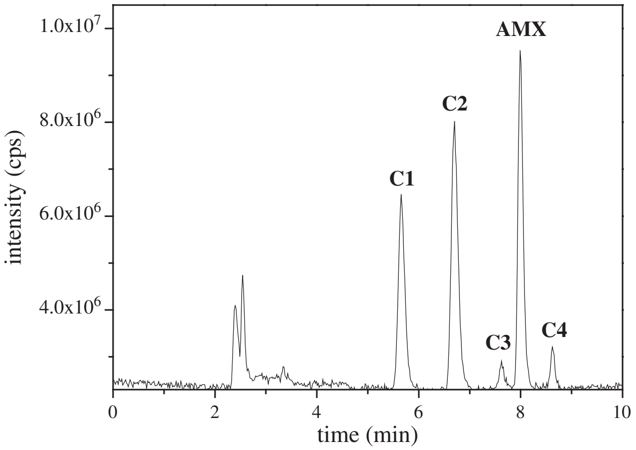

Fig. 1 – LC/TOF-MS chromatogram obtained from hydrolysis of AMX solution (10 mg L^{-1}) in water after 90 min at pH 2.5 in the dark.

it is necessary to evaluate new alternatives to prevent water contamination, considering the risks that residual pharmaceuticals can present to human health and to the environment.

The use of solar advanced oxidation processes (AOPs), such as photo-Fenton for the treatment of non-biodegradable and/or toxic compounds can be an alternative to the conventional processes. Previous works has shown the efficiency of this process on the degradation of different therapeutic classes of pharmaceuticals such as antibiotics, anti-inflammatory and analgesic drugs (Pérez-Estrada et al., 2005, 2007; Shemer et al., 2006; Bautitz and Nogueira, 2007; Trovó et al., 2008, 2009).

Some authors have reported fast and effective degradation of AMX under different conditions, such O_3, O_3/H_2O_2, H_2O_2/UV, TiO_2, Fenton and photo-Fenton (Arslan-Alaton and Dogruel, 2004; Arslan-Alaton and Caglayan, 2005; Andreozzi et al., 2005; Elmolla and Chaudhuri, 2009; Mavronikola et al., 2009; Martins et al., 2009). Arslan-Alaton and Dogruel (2004) applied a variety of advanced oxidation processes, O_3/OH^-, H_2O_2/UV, Fe^2/H_2O_2, Fe^{3+}/H_2O_2, Fe^2/H_2O_2/UV, Fe^{3+}/H_2O_2/UV, to penicillin formulation effluent, concluding that best results regarding complete removal of active substance AMX were obtained using the photo-Fenton process and alkaline ozonation. Although the literature reports the efficiency of AMX degradation by different treatments, no studies have been published on its treatment by solar photo-Fenton, including intermediates generated and evolution of toxicity.

The aim of the present work was to study the use of the photo-Fenton process for the degradation of AMX using a solar simulator. The study includes the influence of the iron species used, the identification of the intermediate products generated during the process by the use of liquid chromatography coupled to time-of-flight mass spectrometry (LC-TOF-MS) and the toxicity assessment.

2. Experimental

2.1. *Chemicals*

All the AMX solutions were prepared in distilled water. AMX was purchased from Sigma–Aldrich and used as received. Hydrogen peroxide (30% w/v) (POCH, SA), $FeSO_4.7H_2O$ and sulphuric acid (POCH, SA), NaOH and bovine liver catalase (Sigma–Aldrich) were also used as received. Potassium ferrioxalate ($K_3Fe(C_2O_4)_3 \cdot 3H_2O$), named as FeOx, was prepared and purified as described previously (Hatchard and Parker, 1956) using iron nitrate and potassium oxalate (Mallinckrodt). Ammonium metavanadate (Sigma–Aldrich) solution was prepared at a concentration of 0.060 M in 0.36 M H_2SO_4. All reagents were of analytical grade. HPLC-grade acetonitrile and methanol (Merck) and formic acid (Fluka) were used for HPLC analyses.

2.2. *Hydrolysis, photolysis and photo-Fenton experiments*

The solutions for hydrolysis and photolysis experiments were prepared by dissolving AMX in distilled water at an initial concentration of 10 mg L^{-1} (TOC 5.3 mg C L^{-1}, natural pH 6.2). Hydrolysis experiments were performed in 250 mL beakers at two different pH (2.5 and 6.2). The beakers were kept in the dark at room temperature for 330 min. The photolysis and photo-Fenton experiments were conducted in a solar simulator (Suntest CPS+ from Heraeus, Germany) equipped with a 1100 W xenon arc lamp and special filters restricting transmission of light below 290 nm. The lamp was set to minimum intensity (250 W m^{-2}), since under high intensity the intermediates generated could be quickly degraded making difficult their detection. Pyrex glass vessels (Schott Durand, Germany) provided with an internal recirculating water system were used to maintain the internal temperature at 25 ± 2 °C.

The initial AMX concentration for the photo-Fenton experiments was 50 mg L^{-1} (TOC = 26.3 mg C L^{-1}). Although this concentration is higher than that found in aqueous

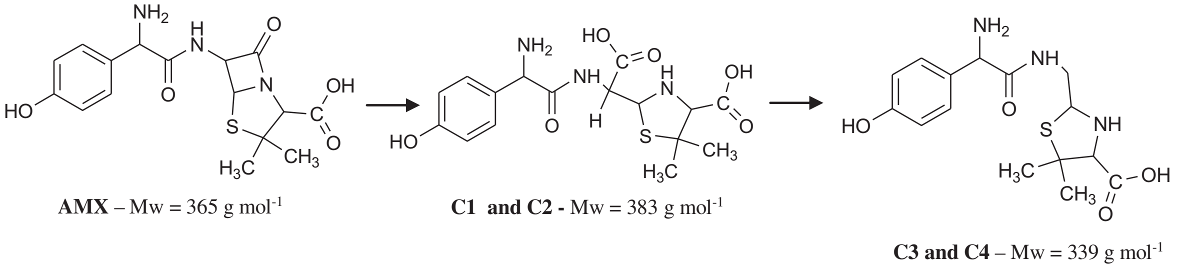

Fig. 2 – Products of AMX hydrolysis in water.