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**Improvement of the activated sludge treatment by its combination with
electro Fenton for the mineralization of sulfamethazine**

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Abstract

A combined process coupling an electro-Fenton pretreatment and a biological degradation in order to mineralize sulfamethazine (SMT) was investigated. The electro-Fenton pretreatment of SMT was first examined and the intermediates products were identified for an initial SMT amount of 0.36 mM, after 1 h electrolysis at pH 3, 18°C, 200 mA. 94.2% SMT was degraded but the level of mineralization remained low (6.5%), ensuring significant residual organic content for a subsequent biological treatment. Two possible degradation reaction pathways involving all the identified and quantified intermediates are proposed. In a second part, biological treatments with fresh activated sludge were performed to complete the mineralization of the electrolyzed solution of SMT, showing an increase of the mineralization yield with time duration of the pretreatment. For an initial SMT concentration of 0.2 mM, a ferrous ions concentration of 0.5 mM, at pH 3, 18°C and 500 mA, the mineralization yield during the biological treatment increased as follows: 61.4, 78.8 and 93.9% for 0.5, 1 and 4 h pretreatment, confirming the relevance of the proposed combined process.

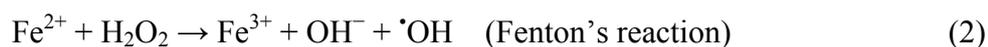
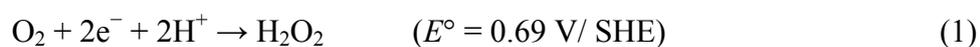
Keywords: Activated sludge; Combined process; Degradation pathway; Drug removal; Electro-Fenton; Sulfamethazine.

1 Introduction

Veterinary antibiotics are largely used for three purposes in animals: to treat sick animals, to prevent infection in animals and as growth promoters to improve feed utilization and production (Barton 2000). However, these antibiotics can reach the environment when they are excreted by animals through urine and feces and thereafter applied to soils as an organic fertilizer. Since these antibiotics are often found to be recalcitrant after excretion (Kwon-Rae et al. 2011), several researches (Burkhardt et al. 2005; Kay et al. 2005) have proved that these

compounds can spread to the groundwater and the surface water by infiltration and runoff, respectively. If these compounds are not eliminated, it can threaten the environment with the potential of adversely affecting aquatic and terrestrial organisms. Consequently and to avoid such contamination, their removal from wastewater appears to be an important current issue.

A wide variety of processes has been therefore studied in order to remove such pollution. Among destructive processes, advanced oxidation processes (AOP) have received great interest in the recent years (Brillas et al. 2009; Uslu and Balcioglu 2009; Pérez-Moya et al. 2010; Pérez-Moya et al. 2011; Saghafiniaa et al. 2011; Batista and Nogueira 2012; Garcia-Segura et al. 2012; Nasuhoglu et al. 2012; Sirés and Brillas 2012; Wei et al. 2012). These processes are based on the generation of very reactive, non-selective powerful oxidizing agents such as hydroxyl radicals $\cdot\text{OH}$ in solution (Ghoneim et al. 2011). Concerning the electro-Fenton process (Jiang and Zhang 2007; Oturan and Brillas 2007; Peralta-Hernandez et al. 2009; Muhammad et al. 2010), H_2O_2 is produced in an acidic medium from the electrochemical reduction of O_2 at the cathode (reaction 1). The generated H_2O_2 reacts with the added Fe^{2+} ions to produce hydroxyl radicals $\cdot\text{OH}$ and Fe^{3+} ions from the Fenton's reaction (reaction 2), which is favored by the catalytic action of the $\text{Fe}^{3+}/\text{Fe}^{2+}$ system, mainly from the regeneration of Fe^{2+} by the cathodic reduction of Fe^{3+} (reaction 3) (Garcia-Segura et al. 2012). Moreover, the method and the involved reactor are easy to handle and to use.



Due to a relatively high operational cost of AOP, potential advantage of the strategy of combining chemical and biological processes to treat contaminants in wastewater was

previously suggested (Scott and Ollis 1995, 1997; Oller et al. 2011). AOP constitute a pretreatment in order to increase the biodegradability of the recalcitrant compounds, leading to the formation of non toxic by-products, more readily metabolizable by microorganisms (De La Rochebrochard D'Auzay et al. 2007; Poyatos et al. 2010).

For this purpose, integrated processes including either UV/H₂O₂ or UV/O₃ (Ledakowicz et al. 2001), photo-Fenton (Farré et al. 2008), photo-Fenton UV/Fe³⁺/H₂O₂ (Dominguez et al. 2005) photocatalysis (Parra et al. 2002) or electro-Fenton (Lin and Chang 2000; Khoufi et al. 2006) followed by biological treatment have been proposed to treat wastewater containing recalcitrant compounds.

The implementation of a direct electrochemical pretreatment and a biological process has been performed in the lab for the treatment of two pesticides, phosmet and 2,4-D, leading to 97 % and 85% mineralization, respectively (Alonso salles et al. 2010; Fontmorin et al. 2012).

The feasibility of an electro-Fenton process for the pretreatment of synthetic aqueous solutions of a veterinary antibiotic, sulfamethazine, was previously verified (Mansour et al. 2012). To complete this previous work, the relevance of the combined process for sulfamethazine treatment should be assessed. For this purpose, an aerobic biological treatment was carried out using activated sludge obtained from a local wastewater treatment plant. In parallel, identification and quantification of by products has been carried out in order to propose mechanisms of electro-Fenton degradation of sulfamethazine.

Compounds structurally close to sulfamethazine have been studied using electro-Fenton as degradation process. Wang et al. (2011) have suggested a degradation mechanism for sulfamethoxazole thanks to the research of by-products by LC-MS, but no quantification nor naming have been made. Dirany et al. (2012) have followed the degradation of sulfachloropyridazine with a combination of electro-Fenton and electro-oxidation using a bore

doped diamond electrode (BDD). A mechanistic pathway was deduced from the identification of by products and the evolution of the toxicity by means of a test based on *Vibrio fischeri* has been followed. However, no biodegradation has been carried out on the electrolyzed solutions in these two studies.

2 Materials and Methods

2.1 Chemicals

Sufamethazine ($C_{11}H_{14}O_2N_4S$, 4-amino-*N*-(4,6-dimethyl-2-pyrimidinyl)benzene-sulfonamide – purity 99%) was purchased from Sigma Aldrich (Saint Quentin Fallavier, France). $FeSO_4 \cdot 7H_2O$ (purity 99%) and Na_2SO_4 (purity 99%) used as a catalyst source and inert supporting electrolyte respectively, were from Acros Organics (Thermo Fisher Scientific, Illkirch, France). Acetonitrile (purity 99.9%) (HPLC grade) was also obtained from Sigma Aldrich. The initial pH of solutions was adjusted to 3 using H_2SO_4 , the optimal pH value for electro-Fenton process (Diagne et al. 2007), using analytical grade sulphuric acid from Acros. All solutions were prepared with ultra-pure water and all the other chemicals used for analysis were purchased from Acros Organics and Sigma Aldrich.

2.2 Electrochemical reactor and Procedures

The degradation of the organic matter by the electro-Fenton process was carried out in duplicate in an undivided cylindrical glass cell equipped with two electrodes; the working volume was 1 L. The dimensions of the carbon felt piece placed on the inner wall of the cell (Le Carbone Lorraine RVG 4000 – Mersen, Paris La Défense, France), which was used as the cathode, were 260 mm×80 mm. Its specific area, measured by the BET method was $0.7 \text{ m}^2 \text{ g}^{-1}$, its thickness was 12 mm, its density was 0.088 g cm^{-3} and its carbon yield was 99.9%. The anode was a cylindrical platinum electrode (50 mm×20 mm) located in the center of the

electrochemical reactor to have a good potential distribution. Prior to electrolysis, compressed air was bubbled for 10 min through the solution at a flow rate of $450 \text{ cm}^3 \text{ min}^{-1}$ to saturate the aqueous solution.

The pH of the solutions was adjusted to 3 by sulfuric acid (H_2SO_4). A catalytic quantity of $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$ (0.1 mM) was introduced into the cell just before the beginning of the electrolysis. The electrodes were connected to a DC power supply (Metrix, model AX 322, Chauvin Arnoux Group, Paris, France) operating in galvanostatic mode to control the current intensity at a value of 200 mA. The ionic strength was maintained constant by the addition of 0.05 M Na_2SO_4 . The electrolytic solution was in circulation with the help of a peristaltic pump (flow rate of 2 L min^{-1}). The temperature was maintained at 18°C and the initial sulfamethazine concentration was 0.36 mM.

2.3 Analytical Procedures

2.3.1 LC–MS/MS method

Ultra-pressure liquid chromatography

The devices used are detailed in previous works (Gervais et al. 2008; Fontmorin et al. 2012). The analytes were separated by a Waters Acquity UPLC system (Waters, Manchester, UK) consisting of an Acquity UPLC binary solvent manager, an Acquity UPLC sample manager and an Acquity UPLC column heater equipped with a Waters Acquity UPLC BEH C18 column ($2.1 \text{ mm} \times 100 \text{ mm}$, $1.7 \mu\text{m}$ particle size) (Milford, MA, USA). Isocratic LC elution was performed with 0.1% formic acid in acetonitrile as mobile phase A and an ultrapure water 9:1 acetonitrile (v/v) mix, with added 0.1% (v/v) of formic acid as mobile phase B. Separation of the analytes on the column was performed at 0.4 mL min^{-1} flow rate.

Tandem mass spectrometry

A Quattro Premier triple-quadrupole mass spectrometer (Micromass, Manchester, UK) equipped with an electrospray ionization source was used for degradation products detection. MS/MS detection was performed in positive mode. Source conditions were capillary voltage 3 kV, source temperature 120 °C and desolvation temperature 350°C. The cone and desolvation gas flows (N₂) were 75 and 750 L h⁻¹, respectively. High-purity argon (99.99%, Air Liquid, Paris, France) was used as collision gas. The analytical device was controlled by Micromass MassLynx 4.1 software.

2.3.2 Ion chromatography

Generated carboxylic acids were identified by DIONEX DX120 ion chromatography equipped with a conductivity detector, using an anion-exchange column AS18 (4 × 250 mm) as the stationary phase, and a solution of Na₂CO₃ (9 mM) in water as the mobile phase. The flow rate was set at 1 mL min⁻¹ (Yahiat et al. 2011).

2.3.3 Chemical Oxygen Demand (COD) measurements

Chemical Oxygen Demand (COD) was measured by means of Nanocolor[®] tests CSB 40 and 160 from Macherey-Nagel (Düren, Germany). The amount of oxygen required for the oxidation of the organic and mineral matter at 148 °C for 2 h was quantified after oxidation with K₂Cr₂O₇ at acidic pH (Fourcade et al. 2012).

2.3.4 Total Organic Carbon (TOC) measurements

The solutions were filtered on Sartorius Stedim Minisart 0.40 µm GF prefilters (Goettingen, Germany). TOC was measured by means of a TOC-V_{CPH/CPN} Total Organic Analyzer Shimadzu. Organic carbon compounds were combusted and converted to CO₂, which was detected and measured by a non-dispersive infrared detector (NDIR). Reproducible TOC values were always obtained using the standard NPOC (Non Purgeable Organic Carbon) method. For each sample, each measurement was triplicated (Oller et al. 2011).

2.3.5 Biological treatment

Activated Sludge used in this study was collected from a local wastewater treatment plant in Rennes. It was washed several times by successive centrifugation, supernatant withdrawing and resuspension of pellet in water, in order to remove any residual carbon or mineral nutrient.

Culture medium was prepared in duplicate in 250 mL serum bottles containing 100 mL of non-treated SMT or a sulfamethazine solution beforehand pretreated by electro-Fenton process for one hour. Minerals were spiked in the medium as highly concentrated solutions to reach the following initial composition (mg L^{-1}): Na_2HPO_4 , 334; K_2HPO_4 , 208; KH_2PO_4 , 85; CaCl_2 , 27.4; $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$, 22.6; NH_4Cl , 2; $\text{FeCl}_3 \cdot 6\text{H}_2\text{O}$, 0.26; and the initial pH was adjusted to 7. Activated sludge was added in order to have initial concentration of 1 g L^{-1} of dry matter. Cultures were agitated at room temperature (25°C) and 5 mL samples were taken regularly, filtered through $0.45 \mu\text{m}$ -syringe filters and injected for TOC measurements.

3 Results and Discussion

3.1 Pretreatment of SMT by the electro-Fenton process

Electrolysis of 0.36 mM (100 mg L^{-1}) SMT showed a rapid degradation until 94.2% of elimination after 1 h pretreatment at pH 3, 18°C , 200 mA, 0.1 M of ferrous ions and 50 mM of Na_2SO_4 . (Fig.1). A low level of mineralization was concomitantly observed (6.5% – Fig.1), as well as a rather low level of oxidation (23.6% from an initial amount of $146 \text{ mg L}^{-1} \text{ O}_2$), which suggested the formation of organic intermediate products, as confirmed by the examination of the LC–MS/MS spectra (not shown), showing that the disappearance of SMT was accompanied by the formation of several intermediates products (Tables 1 and 2).

It can be noticed that in view of a readily analysis of the results, the ratios of the considered parameter amount at a given time t to its initial amount are displayed in Fig.1.

3.2. Identification of the intermediates products

Concomitantly to the degradation of 0.36 mM SMT concentration, in order to elucidate the mechanism of the electro-Fenton oxidation, identification of the main reaction intermediates have been conducted. This identification was carried out by LC-MS/MS analysis by comparing their retention time and their molar mass with those of commercially available standard compounds (Table 1). Generated carboxylic acids were identified by ion chromatography and their retention times were compared with standard compounds (Table 2). The time-course changes of intermediate compounds formed showed that these aromatic compounds are formed simultaneously with the degradation of the target compound (Fig.2). The major product which was formed at the beginning of the electrolysis with a high concentration (4.22 μM) was 2-amino-4,6-dimethylpyrimidine (from the N-S bond cleavage), which reached its maximum concentration after 30 minutes and then began a slow degradation.

The simultaneous apparition of 2-hydroxy-4,6-dimethyl pyrimidine and sulfanilamide boded well for the breaking of the azo bond (C-N). It was found that the concentration of 2-hydroxy-4,6-dimethyl pyrimidine reached a maximum value lower than 0.05 μM after 40 minutes of electrolysis and then followed a slow degradation kinetic. However, the rapid decrease of the concentration of sulfanilamide by the 10th minute of treatment suggested that it was an intermediate product which was rapidly transformed. Furthermore, the shape of the curve corresponding to sulfanilic acid showed an opposite trend to that of sulfanilamide. These opposite trends between these two compounds can be assumed to the fact that the sulfanilamide was oxidized during the process in sulfanilic acid, and the latter caused the rapid formation of 4-aminophenol whose changes showed an increase parallel to that of the acid. On the other hand, the 4-((4,6-dimethyl-2-pyrimidinyl)-amino) phenol appeared during the first minutes of treatment, reaching a maximum concentration of 2.35 μM after 50 minutes of electrolysis. It was formed by oxidation of the N-(4-aminophenyl)-4,6-dimethyl-2-

pyrimidiamine whose changes during the degradation of SMT have not been followed, because no standard of this compound was commercially available.

The comparison of Fig.1 and Fig.2 showed an almost total SMT disappearance (94.2 %) after 60 min of electrolysis under the experimental conditions used, while the total degradation of the intermediate compounds required a longer time, as shown from an electrolysis carried out on a longer time (4 h – not shown).

3.3. Degradation pathway of SMT

Based on the identified degradation products (Tables 1 and 2), two plausible degradation pathways for the electro-Fenton degradation of SMT have been proposed (Fig.3). The intermediates products identified suggest that the degradation process can be initiated by two possible paths due to the oxidative attack of $\cdot\text{OH}$: a cleavage of the C–N bond leading to the formation of sulfanilamide (C_1) and 2-hydroxy-4,6-diméthylpyrimidine (C_2) (path A), and a cleavage of the N–S bond leading to the formation of Sulfanilic acid (C_3) and 2-Amino-4,6-diméthylpyrimidine (C_4) (path B). The 4-Aminophénol (C_5) can be formed from the desulfonation and the hydroxylation of C_3 . Subsequently, these by-products can be oxidized and hence undergo ring cleavage reactions leading to the formation of short chain aliphatic acids such as glyoxylic, succinic and oxalic acids which were the last by-products before mineralization (Fig.3a), as shown from an experiment carried out on a longer time (4 h – not shown).

Another degradation pathway for sulfamethazine can be proposed on the basis of the intermediate products identified (Fig.3b). At first, the N-(4-aminophenyl)-4,6-dimethyl-2-pyrimidinamine (C_7) may appear as the product of a SO_2 extrusion, a phenomenon frequently exhibited by sulfonamides (Neafsey et al. 2010). Then, the 4-((4,6-dimethyl-2-pyrimidinyl)-amino)-phenol (C_6) can be formed by the hydroxylation of (C_7). The hydroxyl radical

attacked the C–N bond in (C₇) producing the 4-Aminophenol (C₅) and the 2-Amino-4,6-dimethylpyrimidine (C₄). On the other hand, a cleavage of the C–N bond of (C₆) leading to the formation of 4-Aminophenol (C₅) and 2-hydroxy-4,6-dimethylpyrimidine(C₂) may occur. The (C₂) can be also produced by hydroxylation of C₄.

The oxidation of these aromatic derivatives during electro-Fenton process led to the formation of short chain aliphatic acids such as glyoxylic, succinic and oxalic acids which are the last by-products before mineralization.

Dirany et al. (2012) have studied the oxidation by hydroxyl radicals of sulfachloropyridazine, a sulfonamide antibiotic with a structure similar to those of sulfamethazine. They have suggested different mechanisms of degradation and some are in agreement with the initial step proposed in this work for SMT, namely either the release of the sulfate group (mechanism B) or the cleavage of the S-N bond (mechanism A).

On the one hand, a partial mineralization, as experimentally observed (6.5% – Fig.1), ensure significant residual organic content for a subsequent biological treatment, and on the other hand the ring opening may lead to the presence of short chain aliphatic acids (Fig.3). From this, an improvement of the biodegradability can be expected (see 3.4).

3.4 Biodegradation of the electrolyzed solution of SMT

A first biological treatment was tested on non-treated SMT and a SMT solution pretreated in the above conditions (see 2.2), and used for the identification of SMT by products (Fig.4).

In these conditions, the mineralization yield of the pretreated solution remained limited (6.5%), and hence an important part of the initial organic content remained available for microbial growth. An activated sludge culture of such solution was then carried out for 17 days (Fig.4). The biological treatment was carried out in duplicate and both are displayed in the figure.

As expected and in agreement with the low value previously obtained for the BOD5 on COD ratio (0.17) (Mansour et al. 2012), the absence of biodegradability of SMT was confirmed, since TOC values remained nearly constant during the 20 days of culture. In the case of treated SMT solution, almost constant TOC values were observed until nearly 10 days of culture; while an almost total mineralization was then observed (97.3 %), since TOC decreased until negligible values after about 13 days of culture.

It can be assumed that at the beginning of culture, microorganisms were not able to assimilate the by-products generated during the degradation of SMT by the electro-Fenton process. A clear decrease was observed from 10 days old culture, indicating activated sludge acclimation to the degradation products resulting from the electrolysis pretreatment. It was in agreement with previous results recorded during the treatment of a pesticide, phosmet, by means of a combined process coupling an electrochemical pretreatment with a biological treatment (Alonso salles et al. 2010). This trend has been also highlighted by Pulgarin et al. (1995) on the photochemical and biochemical degradation of 4-nitrophenol.

The microorganism culture has been carried out with fresh activated sludge highlighting therefore the need to an acclimation step. However, in case of a continuous supply of an electrolyzed solution on an industrial scale, the activated sludge could be consequently acclimated, leading to a decrease of the time needed for biological mineralization.

In parallel, biodegradation tests have been carried out with electrolyzed solutions considering previous operating conditions (Mansour et al. 2012). The corresponding conditions were 0.2 mM initial SMT concentration, 18°C, 500 mA current intensity, and ferrous ions and sodium sulfate concentrations of 0.5 and 50 mM respectively. In these conditions, 99.1% degradation yield of SMT was obtained after 30 min electrolysis.

Since previous results highlighted that the biodegradability based on the BOD₅/COD ratio increased with the pretreatment duration, activated sludge cultures were then carried out for 17-18 days on SMT solutions pretreated in these conditions during various electrolysis times, namely 0.5, 1 and 4 h (Fig.5) (Mansour et al. 2012). For 0.5 h, the electrolyzed solution was not estimated as biodegradable with a ratio < 0.4 (limit of biodegradability); the objective was to conclude on a possible biodegradation despite this unfavorable trend. For 1 h electrolysis, the BOD₅/COD ratio was 0.5; the pretreated solution was therefore supposed to be biodegradable and the objective was to confirm this result. Then, a long time duration (4 h) was tested since a higher biodegradability was expected if compared with 60 min of electrolysis. The objective was to check if mineralization was higher or faster.

The following mineralization yields were observed at the end of the pretreatments, 1.5, 12.5 and 51.9 % for 0.5, 1 and 4 h electrolysis, respectively.

The production of nitrate and ammonium ions has been followed during the electrolysis in these operating conditions (data not shown). Even if ammonium ions were produced from the beginning of the electrolysis, namely before 3 h of electrolysis, the production of nitrate ions resulting from the oxidation of ammonium ions remained negligible. After 0.5 and 1 h of pretreatment, only 1.8 and 3.5% of the initial nitrogen was found in solution suggesting a first attack of only the NH₂ group of SMT. The amount of nitrogen in solution reached 10% after 4 h; while mass balance for nitrogen reached stoichiometric yield only at the end of electrolysis after 10 h. The increasing amount of mineral nitrogen-based ions in solution suggested the pyrimidine ring cleavage leading to short chain aliphatic acids, accounting on the one hand for biodegradability improvement, since they are more biodegradable, and on the other hand for the higher mineralization yield achieved after 4 h of electrolysis if compared to 0.5 or 1 h duration.

The corresponding biological treatments were carried out in duplicate and both are displayed in Fig.5. Similarly to the above biological treatment, almost constant TOC values were observed until nearly 10 days of culture, confirming the need for activated sludge acclimation to the degradation products. A clear decrease was then observed from 10 days of culture and as expected mineralization yields increased with the pretreatment time from 61.4 (Fig.5a) and 78.8 % (Fig.5b) for 0.5 and 1 h electrolysis until an almost total mineralization (93.9 % – Fig.5c) for 4 h electrolysis.

When only SMT was degraded (0.5 h pretreatment), biodegradation was effective but some by-products remained recalcitrant to a biological assimilation. The amounts of recalcitrant by products decreased for a longer duration of the electrolysis. Indeed, after 1 h of pretreatment, the solution was more biodegraded, since nearly 80% of by products were assimilated, and for a longer electrolysis time (4 h), the mineralization was not faster but almost total.

The energy consumption per unit TOC mass (EC_{TOC}) of the electro-Fenton pretreatment was calculated from equation 4 for 0.5, 1 and 4h of electrolysis (Ruiz et al. 2011).

$$EC_{TOC}(kWh.g^{-1}TOC) = \frac{E_{cell}It}{V_s \Delta(TOC)_{exp}} \quad (4)$$

Where E_{cell} is the average potential difference of the cell (V), I the applied current (A), t the duration of electrolysis (h), V_s the solution volume (L) and $\Delta(TOC)_{exp}$ the experimental TOC decay ($mg.L^{-1}$)

If the energetic cost is high ($2.5 kWh.g^{-1}TOC$) at the beginning of electrolysis (0.5h), it has been decreased to reach 0.44 and 0.4 $kWh.g^{-1}TOC$ for 1 and 4 h respectively, in agreement with the findings of Ruiz et al. (2011). This decrease was the consequence of a better mineralization of the solution compared to the sole degradation of SMT.

4 Conclusions

The electro-Fenton degradation of SMT lead to low levels of oxidation and mineralization, close to 24 and 6.5% respectively after 1 h of electrolysis with an initial SMT concentration of 0.36 mM, a ferrous ions concentration of 0.1 mM, at pH 3, 18°C and 200 mA.

The oxidation process produce organic intermediate products and their identification showed that the main ones were sulfanilamide, 2-hydroxy-4,6-diméthylpyrimidine and sulfanilic acid, as well as carboxylic acids such as succinic, glyoxylic and oxalic as aliphatic by-products. Their biodegradability appeared higher than that of the target compound, since a total mineralization of the electrolyzed solution was obtained after about 14 days of biological treatment. The relevance of the coupling of an electro-Fenton pretreatment with a biological treatment for the removal of an antibiotic, sulfamethazine, was also highlighted for other electrolysis conditions. Indeed, for an initial SMT concentration of 0.2 mM, a ferrous ions concentration of 0.5 mM, at pH 3, 18°C and 500 mA, the mineralization yield during the biological treatment increased to 93.9% when the pretreatment time was increased to 4 h. The time duration of the pretreatment entails therefore a better biodegradability which results in a higher mineralization of the electrolyzed solution.

References

- Alonso salles, N., Fourcade, F., Geneste, F., Floner, D. and Amrane, A., 2010. Relevance of an electrochemical process prior to a biological treatment for the removal of an organophosphorous pesticide, phosmet. *Journal of Hazardous Materials* 181, 617-623.
- Barton, MD, 2000. Antibiotic use in animal feed and its impact on human health. *Nutrition Research Reviews* 13, 279-299.
- Batista, A.P.S. and Nogueira, R.F.P., 2012. Parameters affecting sulfonamide photo-fenton degradation - iron complexation and substituent group. *Journal of Photochemistry and Photobiology A* 232, 8-13.
- Brillas, E., Sirés, I. and Oturan, M.A., 2009. Electro-Fenton process and related electrochemical technologies based on Fenton's reaction chemistry. *Chemical Reviews* 109, 6570-6631.
- Burkhardt, M., Stamm, C., Waul, C., Singer, H. and Müller, S., 2005. Surface runoff and transport of sulfonamide antibiotics and tracers on manured grassland. *Journal of Environmental Quality* 34, 1363-1371.
- De La Rochebrochard D'Auzay, S., Brosillon, S., Fourcade, F. and Amrane, A., 2007. Integrated process for degradation of amitrole in wastewaters: photocatalysis/biodegradation. *International Journal of Chemical Reactor Engineering* 5, A51.
- Diagne, M., Oturan, N. and Oturan, M.A., 2007. Removal of methyl parathion from water by electrochemically generated Fenton's reagent. *Chemosphere* 66, 841-848.
- Dirany, A., Sirés, I., Oturan, N., Özcan, A. and Oturan, M.A., 2012. Electrochemical treatment of the antibiotic sulfachloropyridazine: kinetics, reaction pathways and toxicity evolution. *Environmental Science and Technology* 46, 4074-4082.

- Dominguez, J.R., Beltran, J. and Rodriguez, O., 2005. Vis and UV photocatalytic detoxification methods (using TiO_2 , $\text{TiO}_2/\text{H}_2\text{O}_2$, $\text{TiO}_2/\text{S}_2\text{O}_8^{2-}$, O_3 , H_2O_2 , $\text{S}_2\text{O}_8^{2-}$, $\text{Fe}^{3+}/\text{H}_2\text{O}_2$, and $\text{Fe}^{3+}/\text{H}_2\text{O}_2/\text{C}_2\text{O}_4^{2-}$) for dyes treatment. *Catalysis Today* 101, 389-395.
- Farré, M.J., Maldonado, M.I., Gernjak, W., Oller, I., Malato, M. and Domenèch, X., 2008. Coupled solar photo-Fenton and biological treatment for the degradation of diuron and linuron herbicides at pilot scale. *Chemosphere* 72, 622-629.
- Fontmorin, J.M., Huguet, S., Fourcade, F., Geneste, F., Floner, D. and Amrane, A., 2012. Electrochemical oxidation of 2,4-dichlorophenoxyacetic acid: analysis of by-products and improvement of the biodegradability. *Chemical Engineering Journal* 195-196, 208-217.
- Fourcade, F., Yahiat, S., Elandaloussi, K., Brosillon, S. and Amrane, A., 2012. Relevance of photocatalysis prior to biological treatment of organic compounds. *Chemical Engineering & Technology* 35, 238-246.
- Garcia-Segura, S., Garrido, J.A., Rodriguez, R.M., Cabot, P.L., Centellas, F., Arias, C. and Brillas, E., 2012. Mineralization of flumequine in acidic medium by electro-Fenton and photoelectro-Fenton processes. *Water Research* 46, 2067-2076.
- Gervais, G., Brosillon, S., Laplanche, A. and Helen, C., 2008. Ultra-pressure liquid chromatography-electrospray tandem mass spectrometry for multiresidue determination of pesticides in water. *Journal of Chromatography A*. 1202, 163-172.
- Ghoneim, M.M., El-Desoky, H.S. and Zidan, N.M., 2011. Electro-Fenton oxidation of Sunset Yellow FCF azo-dye in aqueous solutions. *Desalination* 274, 22-30.
- Jiang, C.C. and Zhang, J.F., 2007. Progress and prospect in electro-Fenton process for wastewater treatment. *Journal of Zhejiang University Science A* 8, 1118-1125.
- Kay, P., Blackwell, P.A. and Boxall, A.B., 2005. Transport of veterinary antibiotics in overland flow following the application of slurry to arable land. *Chemosphere* 59, 951-959.

- Khoufi, S., Aloui, F. and Sayadi, S., 2006. Treatment of olive oil mill wastewater by combined process electro-Fenton reaction and anaerobic digestion. *Water Research* 40, 2007-2016.
- Kwon-Rae, K., Gary, O., Soon-Ik, K., Deaog-Bae, L. and Yong, S.O., 2011. Occurrence and environmental fate of veterinary antibiotics in the terrestrial environment. *Water Air and Soil Pollution* 214, 163-174.
- Ledakowicz, S., Solecka, M. and Zylla, R., 2001. Biodegradation, decolourisation and detoxification of textile wastewater enhanced by advanced Oxidation Processes. *Journal of Biotechnology* 89, 175-184.
- Lin, S.H. and Chang, C.C., 2000. Treatment of landfill leachate by combined electro-Fenton oxidation and sequencing batch reactor method. *Water Research* 34, 4243-4249.
- Mansour, D., Fourcade, F., Bellakhal, N., Dachraoui, M., Hauchard, D and Amrane, A., 2012. Biodegradability improvement of sulfamethazine solutions by means of electro-Fenton Process. *Water Air and Soil Pollution* 223, 2023-2034.
- Muhammad, U., Hamidi, A.A. and Mohd, S.Y., 2010. Trends in use of Fenton, electro-Fenton and photo-Fenton for the treatment of landfill leachate. *Waste Management* 30, 2113-2121.
- Nasuhoglu, D., Rodayan, A., Berk, A. and Yargeau, V., 2012. Removal of the antibiotic levofloxacin (LEVO) in water by ozonation and TiO₂ photocatalysis. *Chemical Engineering Journal* 189-190, 41-48.
- Neafsey, K, Zeng, X. and Lemley, A.T., 2010. Degradation of sulfonamides in aqueous solution by membrane anodic Fenton treatment. *Journal of Agricultural and Food Chemistry* 58, 1068-1076.
- Oller, I., Malato, S. and Sánchez-Pérez, J.A., 2011. Combination of Advanced Oxidation Processes and biological treatments for wastewater decontamination-A review. *Science of the Total Environment* 409, 4141-4166.

Oturan, M.A. and Brillas, E., 2007. Electrochemical advanced oxidation processes (EAOPs) for environmental applications. *Portugaliae Electrochimica Acta* 25, 1-18.

Parra, S., Malato, M. and Pulgarin, C., 2002. New integrated photocatalytic-biological flow system using supported TiO₂ and fixed bacteria for the mineralization of isoproturon. *Applied Catalysis B: Environment* 36, 131-144.

Peralta-Hernandez, J.M., Martinez- Huitle, C.A., Guzman-Mar, J.L. and Hernandez-Ramirez, A., 2009. Recent advances in the application of electro-Fenton and photoelectro-Fenton for removal of synthetic dyes in wastewater treatment. *Journal of Environmental Engineering and Management* 19, 257-265.

Pérez-Moya, M., Graells, M., Castells, G., Amigo, J., Ortega, E., Buhigas, G., Pérez, L.M. and Mansilla, H.D., 2010. Characterization of the degradation performance of the sulfamethazine antibiotic by-photo-Fenton process. *Water Research* 44, 2533-2540.

Pérez-Moya, M., Mansilla, H.D. and Graells, M., 2011. A practical parametrical characterization of the Fenton and the photo-Fenton sulfamethazine treatment using semi-empirical modeling. *Journal of Chemical Technology and Biotechnology* 86, 826-831.

Poyatos, J.M., Munio, M.M., Almecija, M.C., Torres, J.C., Hontorio, E. and Osorio, F., 2010. Advanced oxidation processes for wastewater treatment: state of the art. *Water Air and Soil Pollution* 205, 187-204.

Pulgarin, C., Peringer, P., Albers, P. and Kiwi, J., 1995. Effect of fe-ZSM-5 zeolite on the photochemical and biochemical degradation of 4-nitrophenol. *Journal of Molecular Catalysis A: Chemistry* 95, 61-74.

Ruiz, E.J., Arias, C., Brillas, E., Hernández-Ramírez, A. and Peralta-Hernández, J.M., 2011. Mineralization of Acid Yellow 36 azo dye by electro-Fenton and solar photoelectro-Fenton processes with a boron-doped diamond anode. *Chemosphere* 82, 495-501.

Saghafiniaa, M.S., Emadiana, S.M. and Vossoughia, M., 2011. Performances evaluation of photo-Fenton process and sonolysis for the treatment of penicillin G formulation effluent. *Procedia Environmental Science* 8, 202-208.

Scott, J.P. and Ollis, D.F., 1995. Integration of chemical and biological processes for water treatment: review and recommendations. *Environmental Progress* 14, 88-103.

Scott, J.P. and Ollis, D.F., 1997. Integration of chemical and biological oxidation processes for water treatment: II. recent illustrations and experiences. *Journal of Advanced Oxidation Technologies* 2, 374-381.

Sirés, I. and Brillas, E. , 2012. Remediation of water pollution caused by pharmaceutical residues based on electrochemical separation and degradation technologies. A review. *Environment International* 102, 212-229.

Uslu, M.O. and Balcioglu, I.A., 2009. Simultaneous removal of oxytetracycline and sulfamethazine antibacterials from animal waste by chemical oxidation processes. *Journal of Agricultural and Food Chemistry* 57, 11284-11291.

Wang, A., Li, Y.Y. and Ledezma Estrada, A., 2011. Mineralization of antibiotic sulfamethoxazole by photoelectro-Fenton treatment using activated acarbon fiber cathode and under UVA irradiation. *Applied Catalysis B: Environment* 102, 378-386.

Wei, L., Venkateswarlu, N., Qixing, Z. and Gregory, V.K., 2012. Effects of Fenton treatment on the properties of effluent organic matter and their relationships with the degradation of pharmaceuticals and personal care products. *Water Research* 46, 403-412.

Yahiat, S., Fourcade, F., Brosillon, S. and Amrane, A., 2011. Removal of antibiotics by integrated process coupling photocatalysis and biological treatment - Case of tetracycline and tylosin. *International Biodeterioration & Biodegradation* 65, 997-1003.

Table 1. Intermediates products generated during the electrolytic degradation of sulfamethazine

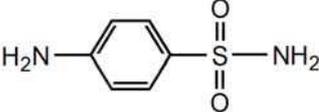
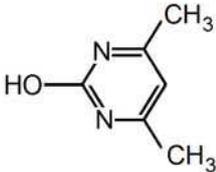
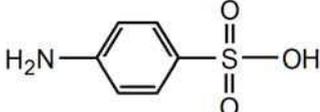
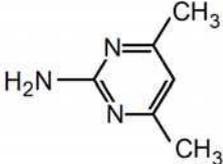
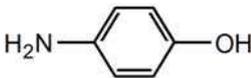
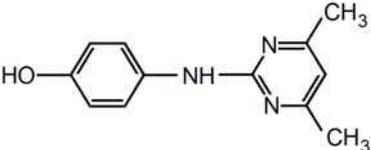
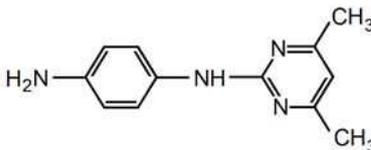
Compound	Name	Chemical structure	Molar mass (g mol ⁻¹)	Retention time (min)
C ₁	sulfanilamide		172.33	0.82
C ₂	2-hydroxy-4,6-dimethylpyrimidine		124.14	0.66
C ₃	Sulfanilic acid		173.21	0.63
C ₄	2-Amino-4,6-dimethylpyrimidine		123.12	0.75
C ₅	4-Aminophenol		109.13	0.63
C ₆	4-((4,6-dimethyl-2-pyrimidinyl)-amino)phenol		215.25	1.98
C ₇	N-(4-aminophenyl)-4,6-dimethyl-2-pyrimidinamine		214.27	1.19

Table 2. Carboxylic acids produced during the electrolytic degradation of sulfamethazine

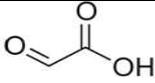
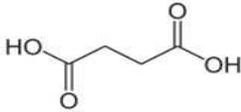
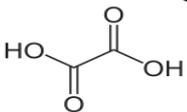
Carboxylic acid	Name	Chemical structure	Molar mass (g mol ⁻¹)	Retention time (min)
C ₈	Glyoxylic acid		74.03	4.54
C ₉	Succinic acid		118.08	14.69
C ₁₀	oxalic acid		90.03	5.42

Figure captions

Fig.1. Time-courses of normalized sulfamethazine concentration ($[\text{SMT}]/[\text{SMT}]_0$) (\blacklozenge) and mineralization, Total Organic Carbon values, ($[\text{TOC}]/[\text{TOC}]_0$) (\blacktriangle) during an electro-Fenton treatment of SMT. $[\text{SMT}]_0 = 0.36$ mM, $[\text{Fe}^{2+}] = 0.1$ mM, $[\text{Na}_2\text{SO}_4] = 50$ mM, $\text{pH} = 3$, $T = 18^\circ\text{C}$, $I = 200$ mA, $V = 1$ L.

Fig.2. Time-course changes of aromatic derivatives of SMT during the electro-Fenton process. $[\text{SMT}]_0 = 0.36$ mM, $[\text{Fe}^{2+}] = 0.1$ mM, $[\text{Na}_2\text{SO}_4] = 50$ mM, $\text{pH} = 3$, $T = 18^\circ\text{C}$, $I = 200$ mA, $V = 1$ L

Fig.3. Pathways proposed for the degradation of SMT by the electro-Fenton process.

Fig.4. Mineralization of non-pretreated SMT (\times) and an electrolyzed solution of SMT (\bullet and Δ) during activated sludge culture. Electro-Fenton pretreatment conditions: $t = 1$ h, $[\text{SMT}]_0 = 0.36$ mM, $[\text{Fe}^{2+}] = 0.1$ mM, $[\text{Na}_2\text{SO}_4] = 50$ mM, $\text{pH} = 3$, $T = 18^\circ\text{C}$, $I = 200$ mA, $V = 1$ L.

Fig.5. Mineralization during activated sludge cultures of pretreated solutions of SMT electrolyzed during 0.5 h (Fig.5a), 1 h (Fig.5b) and 4 h (Fig.5c). Electro-Fenton pretreatment conditions: $[\text{SMT}]_0 = 0.2$ mM, $[\text{Fe}^{2+}] = 0.5$ mM, $[\text{Na}_2\text{SO}_4] = 50$ mM, $\text{pH} = 3$, $T = 18^\circ\text{C}$, $I = 500$ mA, $V = 1$ L. Duplicate experiments (\bullet , Δ).

Figure 1

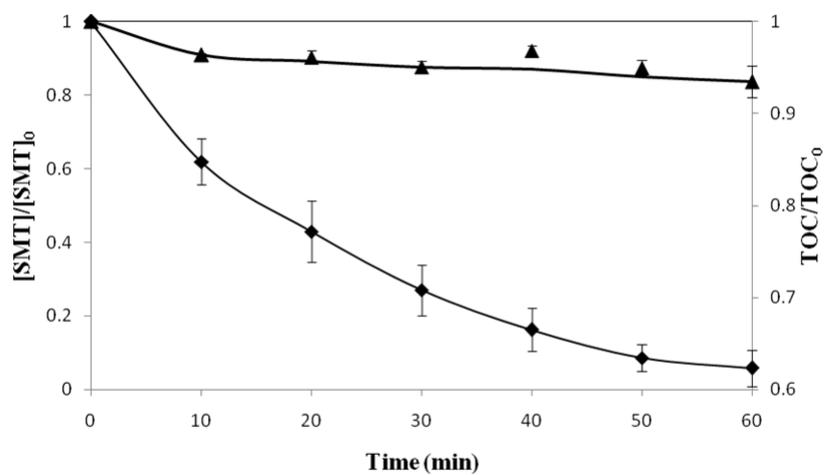


Figure 2

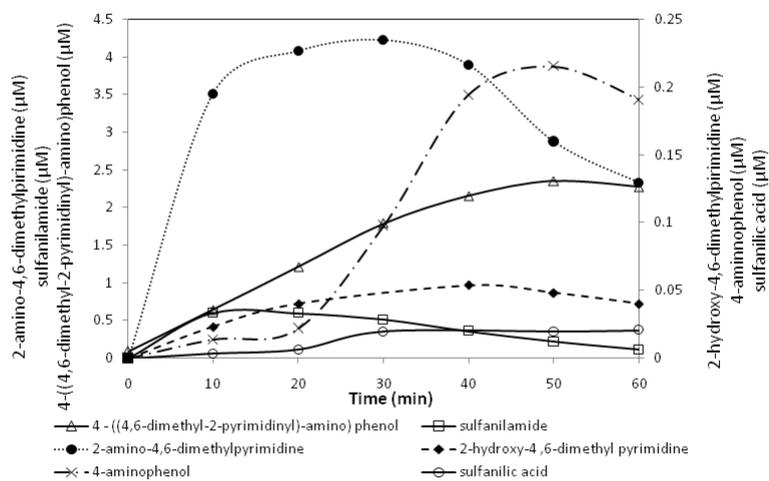


Figure 3a

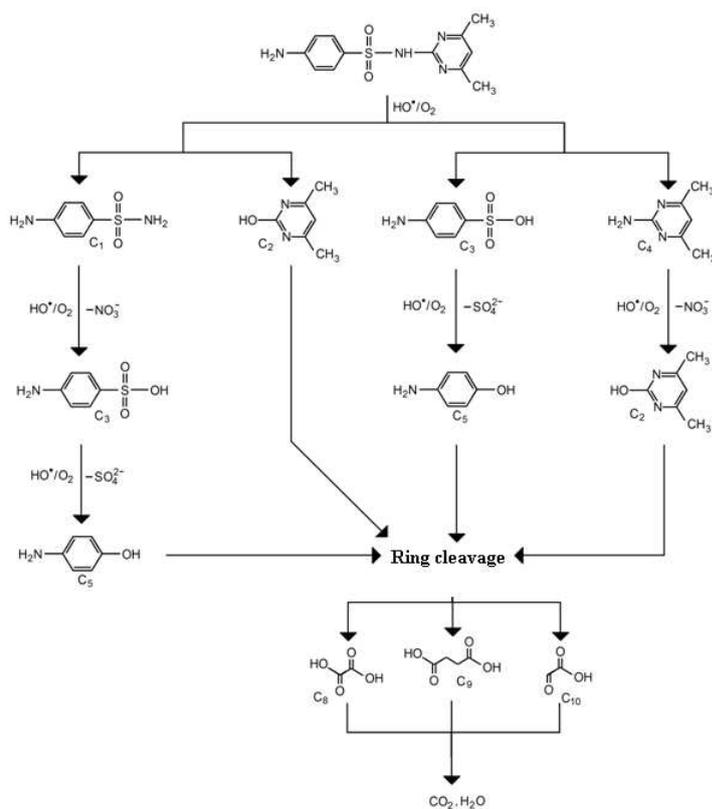


Figure 3b

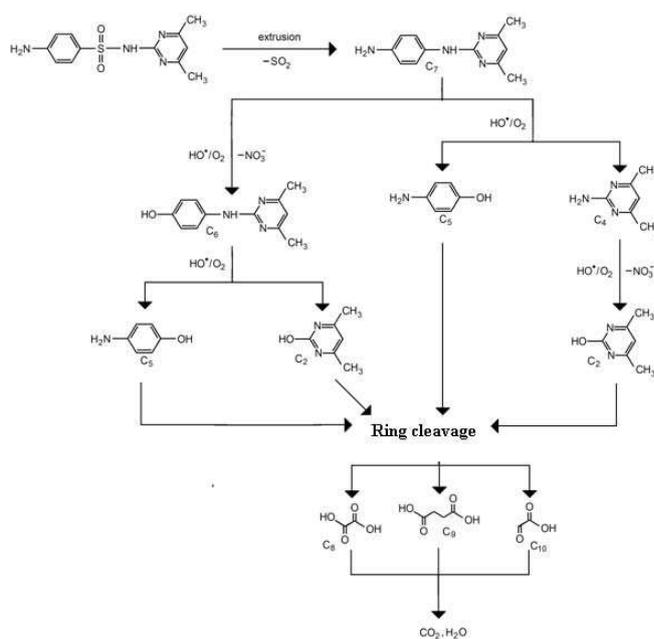


Figure 4

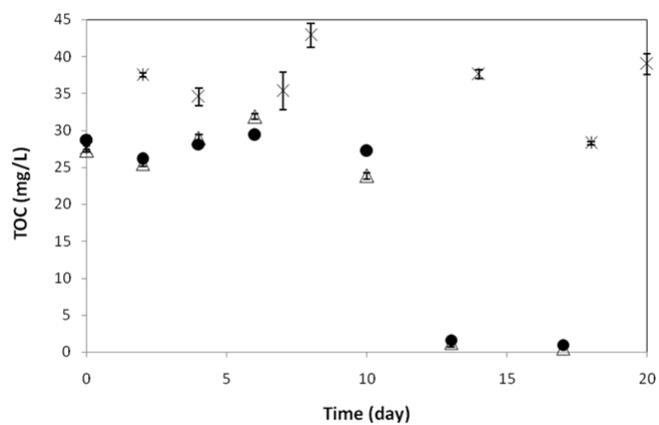


Figure 5a

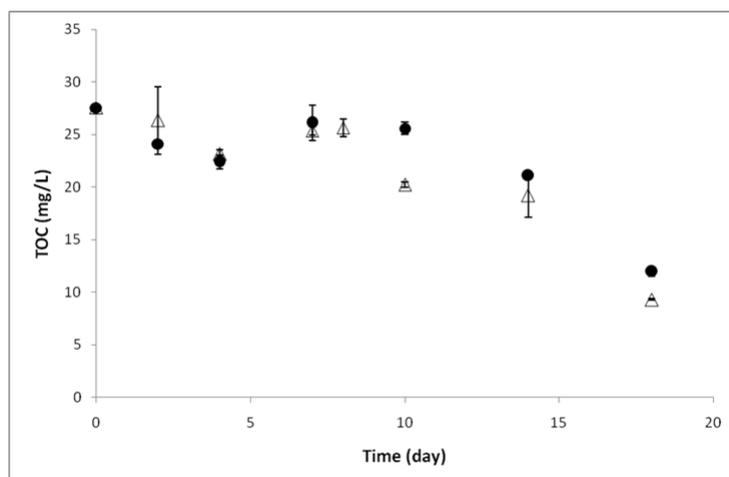


Figure 5b

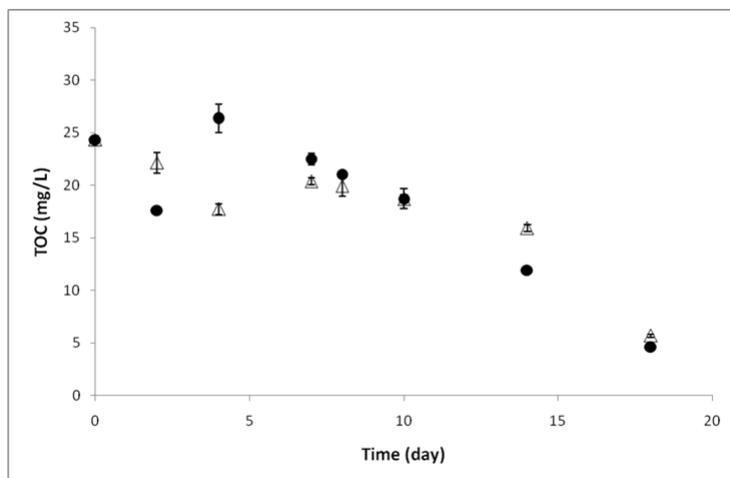


Figure 5c

