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Journal of Hazardous Materials 142 (2007) 58–67

Journal of Hazardous Materials

www.elsevier.com/locate/jhazmat

Detoxification of olive mill wastewater by electrocoagulation and sedimentation processes

Sonia Khoufi, Firas Feki, Sami Sayadi *

Laboratoire des Bio-procédés, Centre de Biotechnologie de Sfax, BP: K, 3038 Sfax, Tunisia

Received 15 March 2006; received in revised form 25 July 2006; accepted 26 July 2006
Available online 29 July 2006

Abstract

Olive mill wastewater (OMW) is characterised by its high suspended solids content (SS), high turbidity (NTU), chemical oxygen demand (COD) concentration up to 100 g l^{-1} and toxic phenolic compounds concentration up to 10 g l^{-1}. This study examined the effect of a physico-electrochemical method to detoxify olive mill wastewater prior an anaerobic biotreatment process. The proposed pre-treatment process consisted in a preliminary electrocoagulation step in which most phenolic compounds were polymerised, followed by a sedimentation step. The BOD_5/COD ratio of the electrocoagulated OMW increased from 0.33, initial value, to 0.58. Furthermore, the sedimentation step yielded the removal of 76.2%, 75% and 71% of phenolic compounds, turbidity and suspended solid, respectively, after 3 days of plain settling. The combination of electrocoagulation and sedimentation allowed a COD reduction and decoloration of about 43% and 90%, respectively. This pre-treatment decreases the inhibition of *Vibrio fisheri* luminescence by 66.4%. Continuous anaerobic biomethanization experiments conducted in parallel with raw OMW and electrocoagulated OMW before and after sedimentation at a loading rate of 6 g COD l^{-1} day^{-1}, proved that the final pre-treated OMW was bioconverted into methane at high yield while raw OMW was very toxic to anaerobic microorganisms.

Keywords: Olive mill wastewater; Electrocoagulation; Sedimentation; Polyphenols; Toxicity; Anaerobic digestion

1. Introduction

Olive oil production industry is characterised by the release of important amounts of liquid and solid by-products [olive mill wastewater (OMW) and olive husk], and by economic and organisational constraints. In the Mediterranean area, where more than 95% of the world's olives are harvested, up to 30 million tons of residues arise per year [1].

OMW contains macromolecules, such as polysaccharides, lipids, proteins and a number of monocyclic and polymeric aromatic molecules generally referred to as phenolic compounds [2]. Specifically, it was reported that the polyphenolic components of OMW are responsible for the dark color, the phytotoxic effects and the antibacterial activity [3,4]. Therefore, the practice of spreading OMW directly on agricultural soils must be accomplished with great vigilance, since it may result in more damage than benefit on soil fertility [5].

The treatment of OMW has been the object of several studies. Many studies focused on several physico-chemical and biological treatment methods that may affect the reduction of the organic load in OMW. One of the most promising OMW biological treatment technologies was the anaerobic digestion [6]. However, the effectiveness of this treatment was not always satisfactory as OMW phenolic compounds, besides contributing to inhibiting the anaerobic microflora [4,7–9] they tend to persist in the effluent of the treatment plant. In order to solve this problem, it was suggested that OMW must be pre-treated by physico-chemical or biological methods to eliminate the inhibitory substances such as polyphenols and residual lipids. Several detoxification processes were reported on the literature. Indeed, to enhance the anaerobic digestion of OMW, fungal pre-treatment was reported to reduce the amount of total phenolic compounds and associated toxicity [10–12]. Fadil et al. [13] studied the effect of microorganisms namely *Geotrichum* sp., *Aspergillus* sp. and *Candida tropicalis* on OMW biodegradation and tested their capacity in reducing both organic and phenolic

Abbreviations: BOD_5, biological oxygen demand; COD, chemical oxygen demand; EC, electrocoagulation; OMW, olive mill wastewater; TSS, total suspended solids; VSS, volatile suspended solids

* Corresponding author. Tel.: +216 74 440 452; fax: +216 74 440 452.
E-mail address: sami.sayadi@cbs.rnrt.tn (S. Sayadi).

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doi:10.1016/j.jhazmat.2006.07.053

contents. This aerobic pre-treatment was capable of reducing the COD and total phenols concentration by about 52.5–62.8% and 44.3–51.7%, respectively, for the various cultures used. As OMW also contains significant amounts of suspended solids and colloidal matter, successful treatment may require a pre-treatment stage to remove these fractions, which typically consist of pectins, proteins, oils and tannins. This can be done by the means of separation processes. Turano et al. [14] developed a two-stage centrifugation–ultrafiltration process capable of reducing the total suspended solid and concentration of COD of OMW by 80% and 90%, respectively. The use of sodium polyacrylate super-absorbent polymers to detoxify OMW has been reported in a recent study [15]. The absorbent was capable of retaining the polyphenolic fraction inside the polymeric chains, while large molecules such as proteins were concentrated in the residual liquid phase whose phytotoxicity was substantially reduced.

The interest in the electrochemical methods for the wastewater treatment, such as electrocoagulation–flocculation and electrochemical separation, is permanently increasing [16]. These are nowadays considered to be the most perspective methods for the purification and treatment of drinkable water, industrial wastewater and sewage.

Electrocoagulation is the electrochemical production of destabilisation agents that brings about charge neutralization for pollutant removal [17]. When a direct current passes through the Fe anodes, Fe^{2+} and Fe^{3+} ions dissolve and combine with hydroxyl ions in water. They form metal hydroxyls, which are partly soluble in water under definite pH values. These metal hydroxides possess a very high ability for adsorption. Coagulated particles attract and adsorb different ions and micro-colloidal particles from the wastewater [18]. The flocks formed in the water are transported to the surface by gas bubbles (H_2O_2, H_2) produced in the electrolysis process. In electroflocculation, the pollutants are removed by the bubbles, capturing the coagulated pollutants and floating to the surface, from where they can be easily removed. This process is characterised by a reduced sludge production, no requirement for chemical use, effective and ease of operation [19]. Because of its many merits, electrocoagulation was used in many fields of industrial wastewater treatment such as textile wastewaters [20,21], landfill leachate [22,23], restaurant wastewater [24], urban wastewaters [25], chemical mechanical polishing wastewater [26] and also raw water for potable supply [27]. Furthermore, this technology was used to remove phenols [28] and surfactants [29].

Toxic OMW was also treated by an electrochemical method with Ti/Pt used as anode and Stainless Steel 304 used as cathode [30]. The results of this study strongly indicate that the total oxidation of OMW through this electrolytic method is not feasible. However, it could be used as an oxidation pre-treatment stage for the wastewater detoxification. The oxidation of phenols and polyphenols found in OMW was studied as a pre-treatment for the submission of wastewater to the traditional biological treatments [31]. Other than these studies, OMW electrocoagulation over iron and aluminium electrodes was found capable of reducing the organic load as well as decolorizing OMW [17,32] while an electro-Fenton process was studied for the treatment of OMW fraction containing various phenolics of low molecular mass [33].

This study investigated the effect of electrocoagulation process as a pre-treatment to detoxify OMW for a biological post-treatment. For that aim it studied the main chemical and physical characteristics of OMW after the electrocoagulation process and the plain settling carried out on crude and electrocoagulated wastewater. Anaerobic digestion was tested on each sample of crude and treated OMW.

2. Materials and methods

2.1. OMW characteristics

The OMW was obtained from an olive oil producing plant located in Sfax (southern Tunisia), which uses a continuous process with a three-phase decanter. The composition of the investigated OMW is reported in Table 1.

2.2. Wastewater analysis

COD was determined according to Knechtel [34]. BOD_5 was determined by the manometric method with a respirometer (BSB-Controller Model 620 T (WTW)). Soluble COD and color analysis were determined after centrifugation of samples during 20 min at 4000 rpm. The settled solids were then dried overnight at 105 °C. These represented the total suspended solids (TSS). The ash content was determined after calcination of the dry sludge at 600 °C for 2 h. The difference between TSS and the ash content was defined as volatile suspended solids (VSS). The residual volume of sludge was determined as the difference between the initial total volume of sample and the final volume of sample supernatant obtained after centrifugation during 20 min at 4000 rpm.

A spectrophotometer (UV–vis Shimadzu) was used for the photometric COD and color analysis. Absorbance at 725 nm was used to quantify phenolic compounds concentration, using the Folin–Ciocalteau method [35]. Results were expressed in terms of pyrogallol equivalent, as this was selected as a reference substance. The turbidity was measured with a turbidimeter (Turb 550IR).

Table 1
Physico-chemical characteristics of crude, EC OMW and anaerobic effluent

Parameters	Crude OMW	EC OMW	Anaerobic effluent
pH	5.2	7.2	8.12
Soluble COD ($g\,l^{-1}$)	36.9	24.5	7.5
BOD_5 ($g\,l^{-1}$)	12.5	14.5	2.25
BOD_5/COD	0.33	0.58	0.30
Monomers ($g\,l^{-1}$)	3.57	0.85	0.326
Ortho-diphenols ($g\,l^{-1}$)	2.22	1.05	0.405
Coloration at 395 nm	17.4	4.4	19
Turbidity (NTU)	2249.3	14957.0	21
TSS ($g\,l^{-1}$)	4.5	23.9	2.89
VSS ($g\,l^{-1}$)	4.0	10.5	1.75

2.3. HPLC analysis of phenolic compounds

The concentration of simple aromatic compounds was followed by HPLC (high performance liquid chromatography) using a Shimadzu 10AVP chromatograph equipped with a Shimadzu 10AVP UV detector. Separation was made by a column (Shim-pack CLC-ODS (M) 250 mm × 4.6 mm) washed with acetonitrile/water (70/30) before and after analysis. A mixture of 50% acetonitrile in 50% water was chosen as the optimal mobile phase. Data were analysed by class VP Shimadzu software. Identification was based on comparisons of the chromatographic retention time and UV absorbance spectra of compounds in OMW with those of authentic standards. *p*-Hydroxyphenylacetic acid, vanillic acid, caffeic acid, *p*-coumaric acid, ferulic acid, tyrosol, and vanillin were obtained from Sigma–Aldrich (Chemie GmbH, Steinheim, Germany). Hydroxytyrosol, the major compound of OMW was prepared in our laboratory after chromatographic purification of ethyl acetate extract from OMW using a silica gel (liChroprep RP; 25–40 μm) column.

A Progel TSK-G 2000-SW Supelco column (300 mm × 7.8 mm) was used with the same Shimadzu 10AVP apparatus described above to analyse the molecular mass distribution of the polyphenolic compounds (complex phenolic compounds) present in both crude and treated OMW. The elution was carried out using a phosphate buffer pH 6.8 and 0.6 ml min^{-1} flow rate. The wave length of the detector was adjusted to 280 nm.

2.4. Electrocoagulation procedure

The experimental set-up is illustrated in Fig. 1. In the electrochemical process, the material of electrodes is essential for reactions. Iron electrodes are used in electrocoagulation, because they are non-toxic, cheap and easy to produce. A semi-pilot scale reactor (36 l), divided in four compartments, was used for electrocoagulation with a special cover supporting a series of iron electrodes. Each compartment contains four plates; where two electrodes were connected as anodes and two as cathodes. The electrodes, having each an immersed area of 8 dm^2 (length 20 cm × height 20 cm × 2 faces), were fixed on the cover and mounted vertically in the cell at a distance of 2 cm from each other. There was a 7 cm distance between the bottom of the electrodes and that of the cell, which allowed easy stirring of the effluent. A direct current (20 A under 40 V) was imposed by a stabilized power supply during 2 h.

2.5. Plain settling

The two samples of crude and electrocoagulated OMW were directly introduced in the settling column. The diagram of the settling column is shown in Fig. 1. This column is a cylinder made of PVC tubing with an internal diameter of 20 cm and a height of 2 m. Sampling ports were uniformly spaced along the length of the column at a height of 25 (L1), 50 (L2), 75 (L3), 100 (L4), 125 (L5) and 150 cm (L6) from the bottom of the column. Samples were collected from each sampling point at 1, 2, 3, 4, 5, 6 and 7 days and analysed to determine the mean characteristics of the supernatants at different times.

2.6. Bioluminescence toxicity test

The microtoxicity test consists of the inhibition of the bioluminescence of *Vibrio fischeri* LCK480 using the (Dr. Lange GmbH, Düsseldorf, Germany) LUMIStox system and according to ISO 11348-2 [36]. Percentage inhibition of the bioluminescence was achieved by mixing 0.5 ml of OMW samples and 0.5 ml luminescent bacterial suspension. After a 15 min exposure at 15 °C, the decrease in light emission was measured. The toxicity of the OMW is expressed as the percent of the inhibition of bioluminescence relative to a non-contaminated reference. A positive control (7.5% NaCl) was included for each test.

2.7. Anaerobic digestion and biogas analysis

Three anaerobic filters (AF) made of a glass column having a working volume of 3 l were used in this study. The inner tubes were enclosed in a jacket through which hot water was circu-

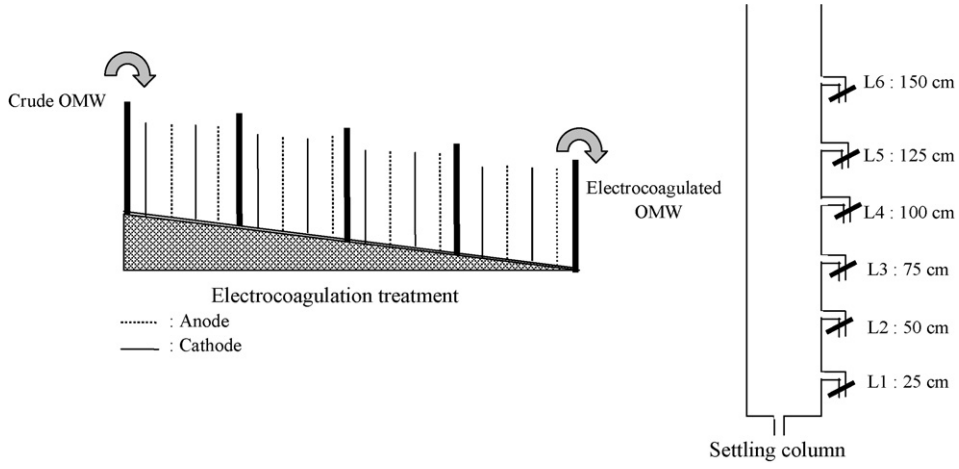

Fig. 1. Electrocoagulation reactor and the settling column used in the laboratory experiments.

lated to maintain the temperature of the filter at 37 °C. These anaerobic filters were packed with polyurethane foam cubes 2 cm × 2 cm × 1 cm (Filtren T45, from Recticel, Wetteren, Belgium) and inoculated with an 8-year-old digester operated with pre-treated OMW. These digesters were run during 2 months with diluted OMW. After this acclimatization period, each bioreactor was supplied with one of the three samples of OMW at broadly the same COD. In all cases, urea was added to the medium to maintain the ratio COD:N = 50. The pH of influent was adjusted to 7.4 with adding $NaHCO_3$. The three bioreactors were then fed in parallel under the same conditions with the crude OMW, EC OMW or S3 at a constant loading rate of 6 g COD l^{-1} day^{-1}. The influent was fed in six times into the reactor using a pump connected to a programmer.

Gas flow rates were measured by a Biogas Counter. Gas samples were taken with a syringe from the tank of biogas and analysed by a gas chromatograph (11M Delsi Instruments) equipped with a stainless steel OD column (2 m in length, Support: Haysep Q) and with a thermal conductibility detector.

3. Results and discussion

3.1. Electrocoagulation of OMW

The electrocoagulation step was performed on raw OMW and without adjustment of pH. The effect of the electrolysis treatment on OMW quality was characterised using mainly pH, COD, BOD_5, color intensity, turbidity, TSS and phenol content measurement as indicators for water quality change. The characteristics of the crude and the electrocoagulated OMW are illustrated in Table 1. As preliminary remarks, it should be noted that, when comparing the two samples, a wide characteristics variation was made. As shown in Table 1, the crude OMW was characterised by a low pH value of 5.2, which limits its biological treatment. However, this pH increased to 7.2 during electrolysis treatment, which may be attributed to the smaller production of H^+ than OH^- as was explained earlier [30] and the reduction in phenol concentration. Indeed, phenols are acids in liquids, and their removal from a solution reduces its acidity.

Biodegradability is measured according to the ratio between the biochemical oxygen demand (BOD_5) and COD, whose value must be less or equal to 0.5. After electrocoagulation, the soluble COD of OMW drops to approximately 33.6% of the initial value. This result points out the ability of the electrolysis process to eliminate a lot of soluble compounds present in OMW. BOD_5 values increased from 12.5 to 14.5 g l^{-1} before and after treatment, respectively. Thus, BOD_5/COD ratio was 0.33 and 0.58, respectively. It appears therefore that a significant proportion of the non-biodegradable matter present in OMW was removed by electrocoagulation. Chromatography analysis confirmed the removal of most low molecular mass phenolics. Besides, the concentration of ortho-diphenols, monitored by Folin–Ciocalteau method, was significantly reduced during the electrocoagulation process. Removal efficiency was about 91.6% for total monomers and 47.5% for ortho-diphenols. The reduction in wastewater phenolic content could be attributed to the polymerisation occurrence of simple aromatic compounds and to the physical and/or chemical adsorption of phenols to solid particles in the remaining sludge as demonstrated in a previous study [33].

Crude OMW was highly colored due to its high content of polyaromatic compounds. In the beginning of the electrolysis treatment, the color intensity of the effluent increased (data not shown). This could be explained by the oxidative polymerisation of phenols and tannins originally present in the sample, which resulted in dark colored organic compounds [32]. However, color intensity decreased to 74.7% of the initial color at the end of the treatment. On the other hand, electrocoagulated OMW (EC OMW) had a relatively higher concentration of suspended solids (23.9 g l^{-1}) than crude OMW (4.5 g l^{-1}) (Table 1). Presumably, the formation of these suspended particles was caused by the electrocoagulation process. The polymers were precipitated with iron, which was continuously dissolved into the wastewater from the cast iron anodes as governed by the Faraday's law [37].

EC OMW had a turbidity value of 14,957 NTU, which was four times higher than the initial value. It should be noted that turbidity depends on the amount of TSS and the presence of gas bubbles produced during the electrolysis process. Thus, an appropriate technique should be adopted for TSS and turbidity removal after electrolysis treatment.

In this study it is suggested that a simple sedimentation process would be able to improve the OMW quality for the biological post-treatment mainly in terms of TSS and turbidity.

3.2. Sedimentation process and mean characteristics of supernatants

The process of electrocoagulation as practised in water treatment can be divided in three separated and sequential steps: coagulant formation, particle destabilisation and interparticle collision. This can explain the existence of high amounts of solid particles in the wastewater during the treatment. After electrocoagulation, the sedimentation step seems necessary to separate the suspended solids from the wastewater. For this reason, EC pre-treated OMW was transferred in the settling column for 7 days. The behaviour of both crude and EC OMW inside the settling column was observed through the analysis of samples collected at different levels.

3.2.1. Removal of suspended solids

Experimentation showed that small suspended particles appeared in wastewater and increased in number and size as the electrolysis treatment continued. Crude OMW contained a smaller quantity of suspended solids (4.5 g l^{-1}) compared to EC OMW (23.9 g l^{-1}). Hence, two different settling processes depending on initial TSS were applied.

Fig. 2 shows TSS evolution across time in each sampling point for EC (a) and crude OMW (b). During the first settling day of EC OMW, mixed solids reflocculated at the top of the column (L6). This flocculated phase was followed by a downward movement of particles, which concentrated towards the bottom of the column. TSS concentration also continued to decrease up to the third settling day in levels L4 and L5 reaching a value inferior to 1 g l^{-1} (Fig. 2a). Results showed that TSS was concentrated

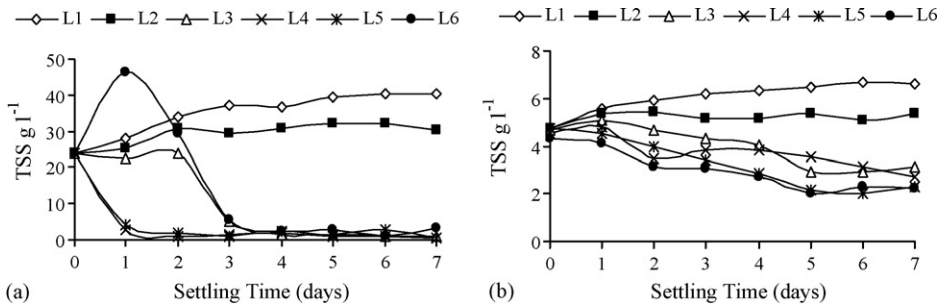

Fig. 2. Total suspended solids evolution during the settling processes of EC (a) and crude (b) OMW.

in levels L1 and L2 and reached at 3 days of settling time 37 and 29.6 g l^{-1}, respectively. It can be noted that coagulated matter settled in function of time, producing a dark precipitate at the bottom of the decanter. The rapid accumulation of TSS at the bottom confirmed that the electrocoagulation process resulted in larger aggregates, thus promoting sedimentation efficiency. For all samples collected from the top of the column (L3, L4, L5), 97.4% of TSS was removed after 6 days of settling time. The choice of the OMW electrocoagulation step appeared to be advantageous for sedimentation efficiency.

Comparing the results shown in Fig. 2a and b, the sedimentation of EC OMW seems to be much more effective than the crude OMW in TSS removal. As can also be seen from Fig. 2b, the TSS of crude OMW did not change along the height of the column and during the settling time. After 7 days of settling time, the accumulation of TSS was observed at levels L1 and L2 and reached 6 and 5 g l^{-1}, respectively, where TSS have decreased at levels L5 and L6 reaching 2 g l^{-1}. This indicated that only a small part of TSS can be removed during the plain settling process. This may be explained by the fact that crude OMW particles were in physical contact with each other, thus reducing the settling rate. The maximum removal of TSS was only about 51% of the initial wastewater content in level L5, even after 7 days of settling. This finding may imply that a large part of TSS in crude OMW was unsettlable suspended matter, which presents a major difficulty in the treatment and handling of OMW. It can be concluded, that electrocoagulation leads to good solid matter removal efficiency and, therefore, it is required before the sedimentation of OMW.

3.2.2. Removal of turbidity

Fig. 3 illustrates turbidity evolution during the settling process of EC (a) and raw (b) OMW. These results were similar to those of TSS. The turbidity of EC OMW was bigger than that of crude OMW (14,957 NTU and 2249.3 NTU, respectively). Turbidity distribution along the settling column height (Fig. 3a) showed that turbidity increased in levels L1 and L2 and decreased in levels L3, L4 and L5 up to the stable value of 1000 NTU after 3 days. However, increasing the settling time above 3 days did not improve the turbidity removal significantly. For the top of column (L6), turbidity increased up to 36,840 NTU after 1 day of settling time but afterward, turbidity decreased to 1200 NTU after 3 days, with a removal efficiency of 95.7%. According to these results, sedimentation can be considered essential for high turbidity removal. However, a longer settling time exceeding 3 days would be not beneficial for the process.

For crude OMW, turbidity was lower than 3500 NTU but there was no improvement in clarification after the settling time. Turbidity remained constant at the different levels of the column even after 7 days of settling time. For instance, at level L4, turbidity removal efficiency of 95% can be reached after only 3 days in the case of EC OMW, while 5.4% efficiency was achieved in the case of crude OMW even after 7 days settling. Hence, it can be concluded that compared to raw OMW, electrocoagulation led to a significant improvement in the turbidity removal. Consequently, electrolysis followed by the sedimentation process before a post biological treatment would result in high TSS and turbidity removals.

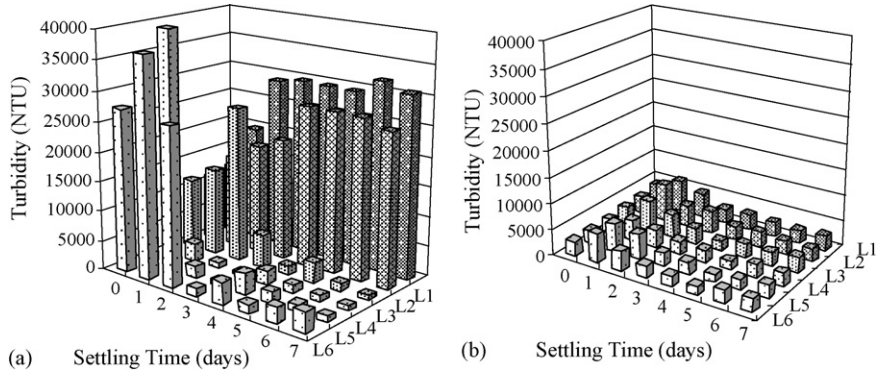

Fig. 3. Turbidity evolution during the settling processes of EC (a) and crude (b) OMW.

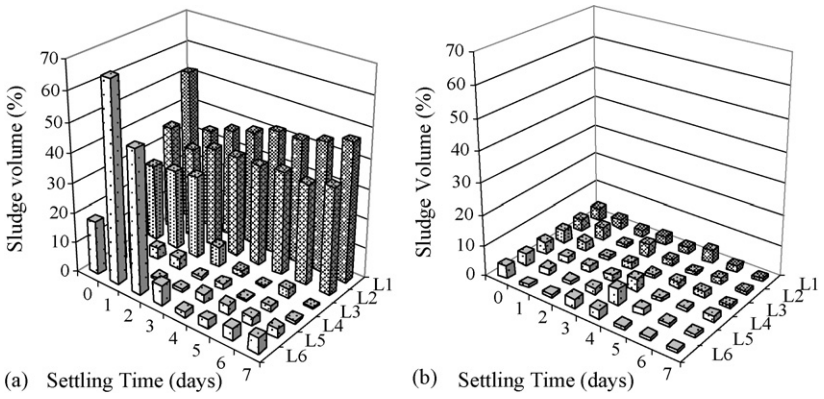

Fig. 4. Sludge volume (%) evolution during the settling processes of EC (a) and crude (b) OMW.

3.2.3. Removal of sludge

The residual volume of sludge across time at various heights of the settling column is presented in Fig. 4a and b. In the case of EC OMW, the amount of sludge decreased gradually during the settling time at the top of the column. However, in levels L3 and L6 (75 and 150 cm from the bottom, respectively), sludge volume increased slightly at the beginning of the settling process, then it decreased gradually at the bottom of the column (L1 and L2) where the sludge is compacting. This could be due to the phenomenon of flocculation of particles at level L6 and to the accumulation of solids in level L3 originating from the top levels. After 1–2 days, sludge percentage began to decrease. Then it reached 2% after 4 days of settling. The produced sludge constituted about 28% (v/v) of the total wastewater volume. A rapid elimination of sludge in the middle of the column was observed only at L4 and L5. After a 7-day settling period, the sludge volume represented 1% of mixed effluent volume at L4 and L5. After 7 days of settling, sludge at the bottom of the column constituted 47% and 36% of the original sample volume, respectively, at L1 and L2. As for crude OMW, the average of the initial volume of sludge was 4%. The percentage of the sludge volume remained stable during the settling time at the different levels of the column. It can be deduced that plain settling was not an effective treatment for raw OMW due to its high stable colloids content and electrocoagulation pre-treatment destabilized these colloids, which will settle rapidly. It would be better to treat OMW with electrocoagulation before the settling process.

3.2.4. Removal of phenolic compounds and toxicity

The results of this study showed the remarkable positive effect of electrocoagulation followed by sedimentation on phenolic compounds removal (Table 1). C18-HPLC analysis showed that 76.2% of monoaromatic compounds were removed in the EC OMW. The removal of monomer compounds was probably due to the polymerisation phenomenon. Samples of untreated and treated OMW were examined by size exclusion-high pressure liquid chromatography (SE-HPLC) in order to clarify the possible changes of OMW phenolic distribution (Fig. 5). Untreated OMW showed a wide molecular mass range of phenolic compounds (Fig. 5a). The molecular mass of these polyphenolics varied from the simple phenolics to the polymers of molecular mass, which exceed 60 kDa. Two groups of peaks were observed:

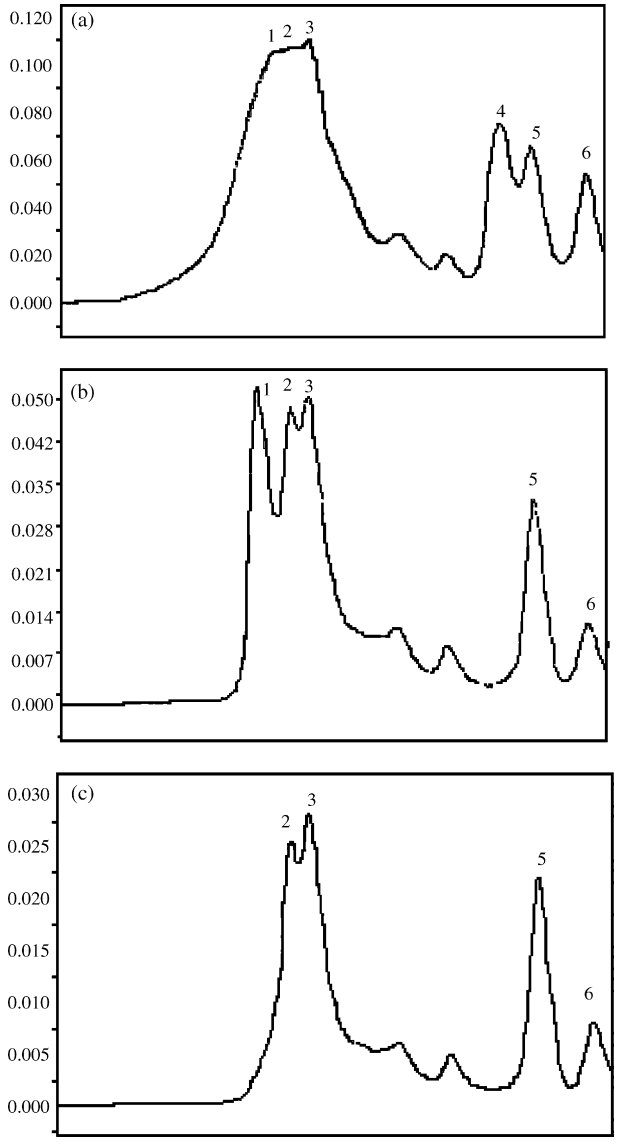

Fig. 5. Molecular mass distribution of phenolics from crude OMW (a), EC OMW (b) and pre-treated OMW sampled from levels L4 and L5 after 3 days of settling time (c).

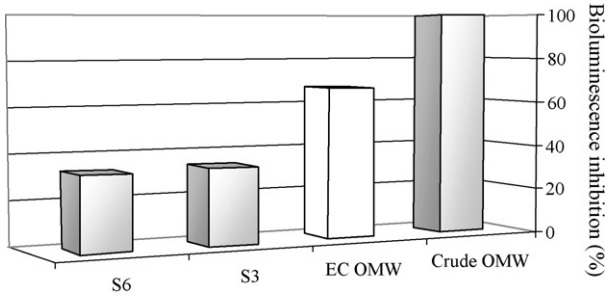

Fig. 6. Percentages of inhibition of *Vibrio fisheri* luminescence after 15 min of exposure with crude OMW, EC OMW and the supernatants S3 and S6 from the settling process vs. time 3 and 6 days from level L4, respectively.

the first group corresponded to high molecular mass phenolics (peaks 1, 2 and 3) and the second group corresponded to medium and low molecular-mass phenolic compounds (peaks 4, 5 and 6). Chromatograms of crude and EC OMW (Fig. 5a and b) show an area reduction of all peaks, particularly in those corresponding to high molecular mass aromatic compounds. Moreover, peak 4 corresponding to the medium molecular mass disappeared. The disappearance of the fraction of intermediate molecular mass compounds can be explained by the polymerisation of these aromatic compounds. As for the chromatogram of pre-treated OMW (S3) sampled from levels L4 and L5 after 3 days of settling time (Fig. 5c), a further reduction in peak areas and a disappearance of peak 1 corresponding to polyphenols of high molecular mass was noted. This may be due to the rapid sedimentation of highly polymerised polyphenolic compounds. These results confirmed the relative predominant role of electrocoagulation in the polymerisation of phenolic compounds. Whereas, sedimentation improves the elimination of polymerised fraction composed of high molecular mass polyphenolics.

As shown in Fig. 6, untreated OMW exercised 100% inhibition on *V. fischeri*. It was reduced to 66.4% in electrocoagulated OMW, which contained 1.05 g l^{-1} of ortho-diphenolics. After 3 and 6 days of settling time bioluminescence toxicity has been decreased in the range of 40%. The percentages of inhibition of *V. fisheri* luminescence showed that compared to the 66.4% inhibition of EC OMW, sedimentation step decreased the relative toxicity by a range of 50%. Moreover, the results showed that high polyphenolic compounds (monitored by SE-HPLC analysis) as well as residual monoaromatic compounds (monitored by C18-HPLC analysis) were eliminated after sedimentation (Fig. 5).

The electrochemical method applied to OMW resulted in the removal of a large amount of recalcitrant polyphenolic compounds and consequently a decrease in toxicity [33]. Hence, considering the overall results, the combination of electrocoagulation with a sedimentation step was efficient for the detoxification of OMW.

Table 2 lists the mean characteristics of supernatants from the settling process during settling time (1, 3 and 6 days) from level L4. Final pH reached 6.26 after 3 days of settling time, allowing the effluent to be treated by conventional biological processes without pH adjustment. After 3 days of settling time, the reduction in total solids and turbidity was more than 95%, while COD removal was more than 22% (Table 2). The results of the experiments showed that sedimentation could efficiently remove TSS and turbidity from the EC OMW. The removal of color from crude OMW was experimentally investigated by electrocoagulation treatment; a removal efficiency of 74.7% was obtained. However, Table 2 showed an increase in the coloration measured as the absorbance at 395 nm from 1 to 1.7 and to 7.1 after 3 and 6 days of settling time, respectively. This was probably due to a further polymerisation of phenolic compounds caused by the presence of metal hydroxides. It is known that iron upon oxidation in an electrolytic system produces iron hydroxide. The $Fe(OH)_n$ formed remains in the aqueous fraction as a gelatinous suspension, which can remove the pollutants such as phenolic compounds from wastewater either by complexation or by electrostatic attraction, followed by coagulation [18]. In the surface complexation mode, the pollutant acts as a ligand to chemically bind hydrous iron [18].

Good quality of treated OMW can be obtained from level L4 and likewise from L3 after 3 days of settling time. The further OMW settlement (over 3 days) lets to low gain in TSS and turbidity removal and so it will be economically ineffective. The obtained pre-treated OMW may be either treated by means of other physico-chemical techniques (ultrafiltration, reverse osmosis, advanced oxidation, etc.), or by biological purification (aerobic or anaerobic digestion) that reduced wastewater polluting load.

3.3. Anaerobic digestion of crude OMW, EC OMW and S3 in continuous anaerobic filter reactors

Fixed-film bioreactors are capable of attaining higher biomass concentrations. These processes offer the advantage of high-load systems, requiring less volume and space compared to conventional reactors to achieve the same performance in treat-

Table 2
Mean characteristics of the supernatants from the settling process vs. time 1, 3 and 6 days from level L4

Parameters	EC OMW	1 day		3 days		6 days	
		Value	Removal (%)	Value	Removal (%)	Value	Removal (%)
pH	7.2	6.55	–	6.26	–	5.70	–
Soluble COD (g l^{-1})	25.3	20.7	18	19.6	22.52	20.1	20.55
Coloration at 395 nm	23.9	1.0	95.8	1.8	92.5	7.1	70.3
Turbidity (NTU)	13280	2980	77.5	520	96	772	94.2
TSS (g l^{-1})	21.73	2.7	87.6	1.41	93.5	0.97	95.5

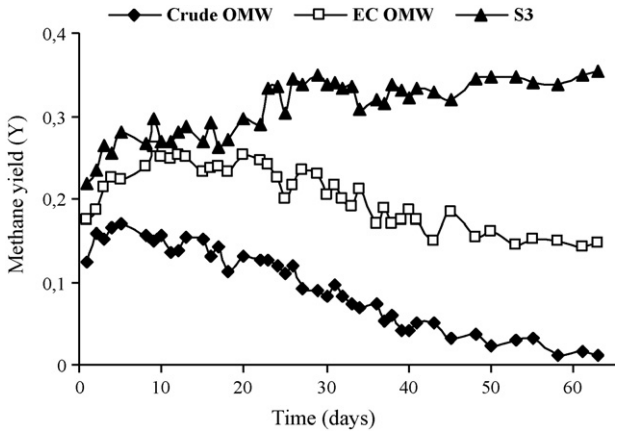

Fig. 7. Methane yield (liter of methane g COD^{-1} introduced) vs. time of the methanization of crude OMW, EC OMW and S3 in continuous anaerobic filters operated at a loading rate of 6 g COD l^{-1} of reactor day^{-1}.

ment. These kinds of reactor also deserve increased attention because of its ability to maintain bacterial populations having low growth rates. In addition, these bioreactors remain stable under transient conditions such as fluctuation in effluent composition, flow and operating conditions and even in the presence of toxic compounds [38]. Because of these features, anaerobic filters were used for the anaerobic biotreatment of crude OMW, EC OMW and S3. Performance of these reactors was characterised in terms of COD removal, pH, volatile fatty acids concentration and methane yield (Y). The methane yield is expressed as litre of CH_4 produced per gram of COD introduced. This value will reflect the effective biotransformation of organics into methane. The optimum theoretical yield is 0.35 l CH_4 produced per gram of COD transformed [39].

Fig. 7 shows the methane yield of crude OMW, EC OMW and S3 separately at a loading rate of 6 g COD l^{-1} of reactor day^{-1} corresponding to hydraulic retention time of 4 days. It is noted from this figure that the anaerobic fermentation of S3 produced the highest volume of methane compared to crude and to EC OMW. High values of methane yields (0.32–0.34 l CH_4 g^{-1} COD introduced) were obtained during the fermentation of S3. The high methane yield also gives evidence that the organic matter was biologically degraded in the reactor. However, during the fermentation of crude OMW, the methane yield dropped dramatically from an average value of 0.15–0.05 l CH_4 g^{-1} COD introduced. As about the EC OMW fermentation, an increase of the methane yield from 0.18 to 0.25 l CH_4 g^{-1} COD introduced was observed up to the 25th day after which it decreased progressively to reach a stable value around 0.15 l CH_4 g^{-1} COD introduced. However, methane production cannot be used to detect problems of anaerobic digesters because it does not reflect the state of imbalance in the reactor [40]. Volatile fatty acids (VFA) have long been recognized as the most important intermediates in the anaerobic process and have been proposed as a control parameter [40,41]. Therefore, changes in VFA concentration can be in response to variation in temperature, organic loading rates or the presence of toxicants. The dynamics of VFA production and pH measurements at different levels of each reactor were determined (data not shown). The performance of the biomethanization process of S3 was noted by the stability of pH at optimum values (7.2) and the low production of VFA at different levels of reactor (data not shown). However, the anaerobic digestion of EC OMW showed a gradual increase of the total VFA concentration in parallel with a pH decrease from 7.2 to 6.2. Inhibition of raw OMW fermentation was accompanied by a total acidification of the reactor (pH lower than 5.6) and a high accumulation of the VFA which exceeded 8 g l^{-1} (data not shown).

Knowing that S3 is exclusive of low and high molecular mass phenolics, it can be concluded that the residual polyphenolic organics identified in S3 (Fig. 5c) could be degraded by the adapted anaerobic consortium at this loading rate. These results are in good agreement with the results obtained by Sayadi et al. [42]. It has been reported that low and high molecular mass phenolics are toxic for anaerobic bacteria at a loading rate of 2.75 g COD l^{-1} day^{-1} corresponding to a residence time of 14.5 days.

The additional sedimentation treatment removes significantly more polyphenolic compounds and possibly other toxic pollutants that have toxic effects on the biomass activity. The increase in methane yield by about 60% more than EC OMW and 90% more than the diluted raw OMW, indicate that electrolysis treatment and sedimentation step contribute significantly to the removal of polyphenolic compounds (Fig. 5) and therefore led to the enhancement of the biological activity as can be seen in Fig. 7.

The biologically treated effluent S3 was characterised by the determination of common parameters (pH, COD, BOD_5, coloration, TSS, concentration of total monomers). The main results are plotted in Table 1. The COD and coloration of the anaerobic effluent remained high. Table 1 showed also that the residual COD was poorly biodegradable (BOD_5 = 0.30 g l^{-1}). C18-HPLC analysis showed that monoaromatics were removed after anaerobic digestion. The total concentration of monoaromatics was 326 mg l^{-1} in the biotreated OMW.

Knowing that untreated OMW causes inhibition of methanization at a loading rate of 2–4 g COD l^{-1} day^{-1} [43,44], it can be concluded that pre-treatment of OMW with EC-sedimentation resulted in decreasing the toxic effect of this wastewater on anaerobic digestion.

4. Conclusion

Olive mill wastewater was treated by electrocoagulation process followed by a sedimentation step. The performance of OMW sedimentation and the removal efficiencies in terms of TSS, turbidity polyphenolic compounds and biotoxicity were investigated. Experimental results revealed the following informations:

- After electrocoagulation pH increased to 7.2, soluble COD was reduced by 33.6%, coloration was reduced to 75% and total simple monomers were reduced to 76.2%.
- Electrocoagulation treatment makes good solid matter and turbidity removal efficiency, 71% and 75%, respectively.

- The electrocoagulation–sedimentation method applied to OMW resulted in the removal of a large amount of recalcitrant polyphenolic compounds as well as a decrease of the microtoxicity.
- The comparison of the biodegradation of crude OMW, EC OMW and S3 in continuous anaerobic digestion showed that the toxicity of crude OMW appeared rapidly as the methane yield decreased drastically.
- The removal of the phenolic compounds and possibly other toxic materials that inhibit the growth of anaerobic microorganism using EC and sedimentation contributes significantly in increasing the efficiency of anaerobic digestion.

Acknowledgements

This research was supported by Contrats Programmes (MRSTDS, Tunisia) and EU contract N° ICA3-CT-2002-00034.

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