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Olive mill wastewater treatment in a membrane bioreactor: Process feasibility and performances

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ABSTRACT

An experimental study of olive mill wastewater (OMW) treatment is undertaken in an external ceramic membrane bioreactor (MBR). The main objective of this work is the study of OMW treatment feasibility using an MBR with a biomass specially acclimated to phenol. The used reactor, equipped with an external ceramic microfiltration membrane gave stabilized permeate flux, around 92 L h⁻¹ m⁻², with zero suspended solid and no phenolic compounds. No fouling problems occur during all the experiments. The short backpulse method adopted allows the use of the MBR in a continuous way. The chemical oxygen demand (COD) remains quite high, and its abatement improvement can be achieved by enhancing the oxygen transfer to the mixed liquor contained in the MBR.

The combination of biological and membrane separation for the OMW treatment seems to be a serious alternative in the reduction of the environmental impact of the olive oil extraction processes effluents. The OMW treatment in a membrane bioreactor can be used as a pre-treatment stage, essentially for phenolic compounds removal before a conventional biological process.

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1. Introduction

Olive oil extraction process generates large amounts of dark liquid effluents called olive mill wastewaters (OMWs) as high as 0.5–0.8 m^3 for 1 ton of olive fruits treated. These effluents result from the mixture of "vegetation water" coming from the olives, and water added during the process. OMW is one of the most contaminated effluents. It is characterised by the variety of pollutants it contains which vary with place, age of growth, season, year, etc. The OMW is a foul smelling acidic wastewater composed of water $(83-92 \text{ wt})$, organic matter $(4-16 \text{ wt})$ and minerals $(1-2 \text{ wt})$. The organic load is so high with biological oxygen demand (BOD) up to 100 g L^{-1} and chemical oxygen demand (COD) up to 200 g L^{-1} . These values are about 300 times higher than those of a typical municipal sewage. Because of their antibacterial effects, phenolic compounds of the organic are the most problematic compounds encountered in the OMW [\[1\].](#page-5-0)

Up to date, the only used "treatment" process in Tunisia is the atmospheric evaporation in open pools or ponds. This way of treating certainly ends by reducing the OMW volume but new environmental problems arise: the evaporation residue, a black foul smelling sludge, mainly composed of high toxic organics and difficult to remove. This constitutes a problem of environmental concern, in particular in arid to semi-arid countries like Tunisia.

Alternative processes must be proposed to reduce pollutant problems with OMW. According to the literature, OMW treatment processes can be physicochemical, biological or combined processes.

Treatment of OMW by advanced oxidation processes (AOPs) like electrochemical oxidation has been increasing recently [\[2–6\]. S](#page-5-0)ome studies used solar photocatalytic pilot plants to study the OMW degradation by combined TiO₂ (very much used in the degradation of textile reactive dye) and photo-Fenton catalysis system [\[7\].](#page-5-0) However, advanced oxidation processes (electrochemical oxidation, Fenton process, ozonation, UV/H_2O_2) lead to different results; the efficiencies percentages depend strongly on the oxidation technique and the pollutant concentration. Moreover, these methods present technical and economical difficulties, which limit their field of application.

Many studies were undertaken on the biological treatment of OMW whether it is aerobic or anaerobic. Three types of organisms: white rot fungi, *Aspergillus* sp., and several different yeasts have been studied. As toxicity has been directly related to the phenolic fraction of the OMW, fungi, which produce polyaromatic hydrocarbon-degrading enzymes, including *Pleurotus* species have been the most studied to remove phenolic compounds [\[8–12\].](#page-5-0)

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Except recent studies carried out by Tsioulpas et al. [\[8\]](#page-5-0) and Aggelis et al. [\[10\], p](#page-5-0)reviously published studies typically address remediation of sterilized OMW in the presence of additional nutrients [\[12\].](#page-5-0) Aggelis et al. [\[10\]](#page-5-0) worked, moreover, on synthetic phenolic wastewater in batch bioreactor cultures and showed that high OMW dilutions should be used, and/or additional treatment should be applied for complete detoxification of the OMW. In addition, the use of mushrooms *Pleurotus* on a large scale is difficult compared to bacteria.

A variety of anaerobic methods such as the use of granular activated carbon (GAC) [\[13\], t](#page-5-0)he upflow anaerobic sludge blanket (UASB) reactor [\[14,15\], s](#page-5-0)ilica beads (SBs) packed bioreactor [\[13\], a](#page-5-0)nd anaerobic filters [\[16\]](#page-5-0) have been also applied to diluted OMW treatment. But it seems that a major limitation of anaerobic digestion of OMW is the inhibition of methanogenic bacteria by simple phenolic compounds and polyphenols. However, the potentiality of OMWs utilisation in the fermentation process of citric acid production is studied and very promising finding is presented [\[17\].](#page-5-0)

Physicochemical processes are expensive and often do not yield full purification and no one of all the above-mentioned studies, despite the effectiveness of the obtained performances, can lead to industrial application. On the other side, the biological treatment remains the most practical and cheaper process to treat this kind of effluent. However biodegradation by conventional activated sludge systems is usually slow due to the inhibitory effect of phenols on the microbial metabolism. Several studies have concerned processes combination for the OMW treatment but no one tested the combination of biological and membrane treatment. In such context, this work is devoted to demonstrate the feasibility of OMW treatment in a membrane bioreactor (MBR).

2. Materials and methods

2.1. Experimental setup and acclimation

The experiments presented in this paper were carried out in an external ceramic membrane bioreactor (Polymem, Toulouse, France) ([Fig. 1\).](#page-2-0) The double-jacket 15 L biological reactor (C) contained 10 L of mixed liquor and was equipped with an aeration system composed of four bottom-installed oxygen nozzles (H). During all the experiments the airflow rate is maintained at a constant value of 7 L min^{-1} corresponding to a superficial gas velocity of 1.5 cm s^{−1}. The membrane module (B) was fed by means of a centrifugal pump (Salmon, Chatou, France) (E) equipped with a frequency variator. The level in the biological reactor, in fact a bubble column, was kept constant by a dosing pump (LMI Milton Toy, Acton, USA) (F) adding a diluted solution of OMW (D). Permeate was collected in a 2-L tank (A) before being sent to the drain. Analogic sensors (Keller) were installed to determine the temperature, transmembrane pressure (TMP) and pressure drops in the membrane module. Two electro-magnetic flowmeters (8732C model, Rosemount, USA) measured the flow rates of permeate and retentate. An oxygen probe (I) (Consort, C932, Belgium) was placed in the biological reactor. With an automaton (Siemens DM8, 24R), the duration and frequency of the backwashings and backpulses were programmed. All these data were transferred to a computer. The membrane was a Carbosep mineral monochannel membrane (Novasep Orelis, France) of 1.2 m length and 6/10 internal/external diameter. The mineral support is carbon with $ZrO₂$ –TiO₂ active layer and the membrane cut-off size is $0.14 \,\mu$ m. The pH range is 0–14 and the maximum TMP is 15×10^5 Pa.

In order to optimize the filtration and reduce the fouling, backpulse method, 1 s/1 min, consisting on very short backwashing of 1 s duration for each minute filtration has been adopted. During a backpulse, pressurized air (2–4 bar) is used to force a known volume of permeate to go back through the membrane to the feed side. As this technique requires high pressure resistant membranes, at the present time and for high pressures, only ceramic membranes for microfiltration and ultrafiltration seem to meet this requirement. Further information concerning the used backpulse method could be found elsewhere [\[18\]. T](#page-5-0)he biomass used in this study has been previously adapted to phenol. The total suspended solids (TSSs) of the used biomass as inoculum is 12 g L^{-1} and the volatile suspended solids (VSSs) is $10.8 g L^{-1}$. The acclimation step is of importance and makes the micro-organisms possessing the enzymatic material necessary to degrade the phenolic compounds and reveal a new population which is adapted to this toxic agent and which is able to consume it as a substrate. It has been recently [\[19\]](#page-5-0) proven that the acclimation is relatively easy to carry out by using a slightly salted medium and without inevitably being under the most favourable conditions of temperature and pH for the development of these micro-organisms, subject to respect steps of increasing phenol concentrations. Phenol being a source of carbon for bacteria, the sludge is acclimated by adding this polluting agent, whose low initial concentration is increased over time. Acclimation is completed when the bacterial growth constant becomes higher than the decay constant with a complete consumption of phenol.

2.2. Analytical techniques

The biomass concentration in the mixed liquor and more especially the mixed liquor suspended solids (MLSSs) were determined by centrifugation (Sigma 265, SIGMA, Germany) of 33 mL of sludge at 4000 rpm for 10 min. The residue was placed in a crucible in a stove (105 \degree C) for 24 h. The concentration of volatile suspended solids was determined by placing the dry matter in an oven at 550 \circ C for 2 h. The difference between the weight of the dry matter previously obtained and that of the matter heated at 550 ◦C give the quantity of mixed liquor volatile suspended solids (MLVSSs). The rheological behaviour of the used biomass is determined using an "Advanced Rheometer AR 550". The COD corresponding to the amount of oxygen necessary for a chemical oxidation of the organic matter contained in the solution was determined using a hot sulphuric solution of potassium dichromate ($K_2Cr_2O_7$) with silver sulphate (Ag_2SO_4) being used as a catalyst, for 2h at 148 ◦C (COD tube (Merck, Darmstad, Germany; COD reactor,

Fig. 1. Membrane bioreactor used for OMW treatment experiments.

Bioblock Scientific)). The addition of $HgSO₄$ prevents the oxidation of inorganic matter. COD was then measured by spectrophotometry (Aqualate, Thermosopectronic) at two different wave lengths (605 and 325 nm) depending on the sample concentration. Gas chromatography (GC) was used to analyze the phenolic compounds (Chrompack, CP 9001, Middelburg, the Netherlands, Capillary Col. SGE 928126). The carrier gas used was helium and the detector was a flame ionization detector (FID). The GC technique has been used in this work for immediate quantification of phenolic compounds presence. The detection time was about 25 min. The injections are done directly after sample extraction from the permeate sampling gauge and the feeding solution. The sampling is filtering a 0.45 μ m pore size filter. Phenol and polyphenolic compounds (2,6-xylenol, *o*-cresol, *o*-ethylphenol, 2,5-xylenol, *p*cresol, 2,4-xylenol, *m*-cresol, 2-*iso*-propylphenol, 2,3-xylenol and 3,5-xylenol + *p*-ethylphenol) have retention time with the used GC column between 7 and 14 min. Using the BORWIN chromatography software (version 1.21.60) we treat all the obtained spectra. This treatment consist on the automatic baseline correction, automatic smoothness, manual peaks indexation according to polyphenol retention time and then surface area calculation which is also made using the BORWIN software. This procedure allowed us for polyphenolic compounds other than phenol, to compare between the peaks areas before and after MBR treatment which are proportional to compound concentration in the studied media. For phenol, standards are used to get a calibration curve which has a linear fit for concentration between the 0.0001 and 1 g L^{-1} and between 0.1 and $10 g L^{-1}$ with a correlation factor of 0.9999.

The temperature programme was: detector temperature 240 ◦C, injector temperature 230 ◦C, oven initial temperature 105 ◦C, oven final temperature 190 °C, oven rise 10 °C min⁻¹, initial time 2 min and final time 15 min.

3. Results and discussion

The batch biodegradation kinetic study carried out with a 40 times diluted solutions of raw OMW, whose main characteristics are presented in Table 1, shows that the used biomass, acclimated to phenol only, behaves normally in spite of the heterogeneity of the used substrate. The cell growth occurs concurrently with the oxidation of organic compounds. Because biomass material is mostly organic, the increase in biomass can be measured by the estimation of the MLSS or the MLVSS. The increase of

Table 1 Characteristics of the used OMW

pH	5.21
COD $(g_{O_2} L^{-1})$	117.6
SS (gL^{-1})	7.65
$N-NO_3$ ⁻ (mg L ⁻¹)	41
$N-NH_4$ ⁺ (mg L ⁻¹)	32
Total salinity (gL ⁻¹)	18.46
Total phenols (gL^{-1})	6.32
Volatile fatty acids (gL^{-1})	9.78

the bacterial mass, estimated by the MLSS evolution (Fig. 2) is 0.175 g L⁻¹ h⁻¹. The COD removal as measured from batch exper-iments ([Fig. 3\)](#page-3-0) is 0.347 $g_{CD} L⁻¹ h⁻¹$. The ratio of the amount of biomass produced to the amount of substrate consumed is defined as the biomass yield. The yield is based on easily measurable parameter reflecting the overall organic compound consumption, in our case the COD. Thus, the calculated biomass yield is $0.51\,g_{MLSS}\,g_{\rm COD}^{-1}$ removed. The acclimated biomass used here has been exposed within batch experiments to different phenol concentrations, and the kinetic constants of the Haldane equation were determined: the specific growth rate ($\mu_{\rm m}$ = 0.438 h⁻¹), the half saturation constant $(K_s = 29.54 \text{ mg L}^{-1})$ and the substrate inhibition constant (K_i = 72.45 mg L⁻¹). Haldane kinetic parameters applied to the obtained data of biomass growth of the batch experiments made with the OMW solution shows good agreement. The MLVSS

Fig. 2. Mixed liquor suspended solid variation during batch experiment.

Fig. 4. Typical example of respirometric measurements with the OUR variation.

represents 87–92% of the MLSS for all the measured samples during the set of made experiments.

A typical oxygen uptake rate (OUR) test is presented in Fig. 4. During the gassing-off period, the dissolved oxygen concentration measured in the bulk of themixed liquor decreases rapidly, showing high biomass oxygen consumption. The switch to the gassing-on, re-establish the oxygen level at its initial value (2.5 ppm). Many information can be deduced from such test. The first one is the K_1a , the volumetric mass transfer coefficient. From the re-aeration part of the test, and supposing a completely stirred behaviour of the mixed liquor within the MBR, the K_1a is estimated to 0.27 min⁻¹. The relatively high value of K_la in a media as concentrated as the one used within the MBR, 12 g L^{-1} , is due to the excess of airflow, 7 L min−1, which maintain a constant dissolved oxygen value of 2.5 mg L⁻¹. The oxygen mass balance in the reactor leads to the following expression: OUR = $K_1a(C^* - C) - (dC/dt)$ [\[20\], w](#page-6-0)hich evaluates of the rate of consumed oxygen from the OUR test. The main information we tried to search for from this kind of test is the ability of the micro-organisms to follow up their activity even when the aeration is cut-off (during the gassing-off period) and their ability to consume all the available dissolved oxygen even at very low concentration. The MLSS, COD analysis and OUR measurements are carried out in both batch kinetic study and continuous MBR feeding. The mean bacterial mass increase during the continuous MBR feeding operation is 1.7 kg m⁻³ day⁻¹.

The strategy adopted in this work has consisted to feed continuously the MBR by diluted OMW solutions which have been concentrated gradually. The operating conditions of the MBR are summarized in Table 2. This has allowed the realisation of a set of four experiments before blocking completely the biomass activ-

Fig. 5. Total phenolic compounds variation in the MBR mixed liquor.

ity by the accumulation of phenotoxic compounds in the reactor. This accumulation is due to the microfiltration step, which gives a retentate with no suspended solid nor phenolic compounds.

The phenol compounds analysis made during this work concerns the permeate and the feeding solution. No analysis of the adsorbed phenolic compounds on the extra/intra cell's part is made in this work. The difference of the phenol compounds between the feeding solution and the permeate is the accumulated phenols in the bioreactor. The daily excess sludge extracted from the reactor, 3.6 L day⁻¹, is not enough to remove the accumulated phenol compounds whose concentration increase during all the set of made experiments as it can be seen from Fig. 5 which is simply the phenols mass balance expression with the assumption that no bioconversion occurs during all the process. The final phenols concentration, at the end of the fourth experiment, reaches 5.41 g L⁻¹ within the mixed liquor of the MBR. At this concentration phenols become toxic for the used micro-organism. The MBR excess biomass extracted containing adsorbed phenols are subject to the conventional sludge treatment (thickening—stabilization, dewatering and thermal or agricultural valorisation).

Within those four experiments which last 48 h, we successively used OMW solutions of 1500, 2700, 5300 and 1500 ppm COD. No pre-treatment of the OMW is adopted and the dilution rate of the reactor, based on 1 h feed flowrate duration, is 4.75. The chemical oxygen demand abatement efficiency is respectively 81%, 58%, 37% and 78%.When the biomass was in contact with a strong concentration of COD (5300 ppm) it could eliminate only 37%. However, the biomass maintained a percentage of COD removal of approximately 80% when OMW solutions of 1500 ppm COD was used again. Thus biomass resisted well to a strong load in COD.

These results are compared with literature [\(Table 3\),](#page-4-0) for example, treatment by *Pleurotus* spp. strains showed that more than 70% of the initial phenolic compounds could be removed. At the same time the remaining phenolic and/or some of the oxidation products in the treated OMW were more toxic than the

MBR operating parameters

nd: no data.

original phenolic compounds [\[8\]. C](#page-5-0)omplete degradation of phenols was achieved with electrochemical oxidation of diluted OMW but this was accompanied by relatively low COD removal that never exceeded 40%[\[5\]. I](#page-5-0)n the same way, biotreatment by fungal laccase is selective as suggested by the rather low COD reductions (about 5%). Then enzyme acts on phenolic substrates by removing one electron from the hydroxyl group [\[21\].](#page-6-0)

Therefore, compared with literature, MBR gave interesting results in the COD and phenol removal. For all the performed experiments, the MLSS is kept at a constant value (12 g L^{-1}) in the MBR. The excess biomass is extracted from the MBR to keep the bioreactive media at a constant value. The amount of extracted quantity necessary to have a constant value of 12 g L^{-1} has been estimated from the batch experiments and confirmed by MLSS sampling. The flowrate of the extracted mixed liquor used here is $0.15 L h^{-1}$ and allows the maintaining of a constant value of MLSS in the MBR.

The relatively high biomass concentration in the MBR represents a real limitation factor of the oxygen transfer [\[22\].](#page-6-0) Several authors have showed the biomass influence on the reduction of the aeration capacity of biological systems [\[23–26\]. T](#page-6-0)he gas/liquid transfer is highly dependent on the biomass concentration. An increase in sludge solid concentration induces an increase in apparent viscosity of the suspension that can strongly affect the oxygen transfer in the liquid phase, leading to insufficient aeration rates of the micro-organisms and thus to low purification yields [\[22\]. F](#page-6-0)or biomass concentrations common in a MBR, the aeration capacity of the bioreactor decreases by approximately a factor 4 compared with clean water. However, because of the complex rheological behaviour of biological suspensions, the rheological characterisation of wastewater sludge, in relation to its processing in aeration bioreactors of different geometries [\[27,28\]](#page-6-0) or MBR [\[29,30\], h](#page-6-0)as only been recently studied in the literature. Structural units of biological sludge (clusters, aggregates, flocs, etc.) described by Quemada [\[31\]](#page-6-0) have a size that depends on the applied shear through the hydrodynamic stress acting on the structural units. However, in the case of our experiments, we can consider that the suspension structure is similar whatever the experiment since the injection of air bubbles and MLSS are similar from one experiment to another the only parameter which varies is the COD concentration. The shear stress applied to the suspension (τ in Pa) is related to the rate of strain (in s−1) in stationary laminar flows (Fig. 6). Wastewater treatment plant sludges are known as viscoplastic fluids and yield stress models allow to represent their behaviour from very small stresses (before flow occurs) to stresses exceeding the yield value, for which the sludge begins to flow in a shear-thinning manner (Herschel–Bulkley (HB) model). Our sludges are well represented by this HB model. The rheological measurements are done for all the set of experiments. The results are exactly the same since those measurements depend essentially on the MLSS in MBR. This parameter is kept during all the experiments at a constant value (12 g L^{-1}). The variation during the experiments of the biomass quantity do not affect the rheological behaviour of the bioreactive media.

In MBRs, as in all aerobic wastewater processes, both the biomass characteristics and the design of the aeration system affect the oxygen transfer. Biomass is a heterogeneous mixture of particles, micro-organisms, colloids, organic polymers and cations, of widely varying shapes, sizes and densities. The oxygen transfer limitation being one of the biomass activity declines, the global MBR efficiency improvement depends on its enhancement.

In another hand, the most problematic substances met in the OMW are the phenolic compounds especially when biological processes are used. The phenolic compounds present in the feeding substrate are considerably reduced [\(Fig. 7\).](#page-5-0) The used GC column allows the resolution of 10 phenolic compounds, other than phenol, with a retention time between 7 and 14 min. The chromatographic analysis of both the feeding solution and the permeate shows that nearly all the phenolic compounds have been totally removed. Based on peak surface calculation, the phenol is removed with an efficiency of 92%. The same chromatograms are obtained for all the undertaken experiments showing the high rate biomass phenolic compound biosorption which may be the main phenols abatement mechanism. The phenolic compounds concentration in permeate was decreased with respect to OMW fed as result of both adsorption onto cells surface and bioconversion. The phenolic compound adsorption on both active and dead biomass has been widely studied [\[32–35\].](#page-6-0) It is reported that phenolic compounds are mostly retained on cell surfaces than biodegraded. Since the MBR biomass concentration is no longer controlled by secondary clarifier solids loading limitation, the ability of MBR to operate at high MLSS concentration permits a high amount of phenolic compound retention on biomass surface. No specific measurements in this work are done to evaluate the phenolic compound bioconversion. Nevertheless slow rate phenolic compounds biological conversion has been reported [\[36\].](#page-6-0) The membrane filtration step provides an effluent with high quality in terms of suspended solid, the phenolic compounds adsorbed on suspended solid remains within the retentate phase and the obtained permeate is phenols free.

Fig. 6. Rheological behaviour of the mixed liquor.

Fig. 7. Typical chromatogram of OMW feed (up) and permeate (bottom) solutions.

Fig. 8 shows typical record of the permeate flowrate. The fluctuations observed during OMW treatment phase are due to the backpulse washing step. Those fluctuations are also observed for all the measured pressures (pressure drop in the membrane module and pressure in both sides of the membrane) and retentate flowrate and translate the backwash effect on process progress. The MBR has been adjusted to treat nearly $1.5 L h^{-1}$ of OMW with a membrane surface of only $0.0226 \,\mathrm{m}^2$. In order to avoid the ceramic membrane fouling, short backpulse duration of 1 s for each minute filtration (1 s/1 min) is found to be suitable to get stabilized process functioning and stabilized permeate flowrate. The mean permeate flowrate value is 2.1 L h⁻¹. The TMP is kept at a constant value of 1–1.5 bar. Higher TMP causes serious fouling problems especially when the biomass concentration in the retentate is more than 12 g L^{-1} .

Fig. 8. Permeate typical flowrate records.

4. Conclusion

Diluted solutions of OMW have been treated in a membrane bioreactor. The used reactor, equipped with an external ceramic ultrafiltration membrane gave stabilized permeate flux, $92 L h^{-1}$ m⁻², with zero suspended solid and no phenolic compounds. No fouling problems occur during all the made experiments. The backpulse method adopted allows the MBR use in a continuous way. COD remains quite high, from 285 to 3339 mg_{O2} L⁻¹ depending on the used dilution factor, and its abatement improvement may be accomplished by the enhancement of the oxygen transfer to the mixed liquor contained in the MBR. Further studies concerning the mass transfer in high concentrated mixed liquor treating an OMW solution, and the practical operating conditions for best oxygen transfer rate are planned and will enable better COD removal performances. OMW treatment in a membrane bioreactor is possible and can be used as a pre-treatment stage, essentially for phenolic compounds removal before a conventional biological process.

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