On the applicability of a hybrid bioreactor operated with polymeric tubing for the biological treatment of saline wastewater

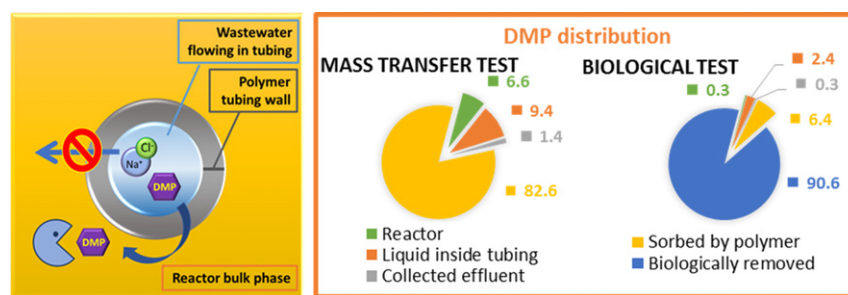M. Concetta Tomei [a,*], Domenica Mosca Angelucci [a], Valentina Stazi [a], Andrew J. Daugulis [b]

[a] Water Research Institute, C.N.R., Via Salaria km 29.300, CP 10, 00015 Monterotondo Stazione (Rome), Italy
[b] Department of Chemical Engineering, Queen's University, Kingston K7L 3N6, Ontario, Canada

HIGHLIGHTS

- A novel tubing bioreactor was applied for treating saline wastewater.
- Different polymer tubing have been tested: Hytrel G3548 gave the best performance.
- Salt separation and biodegradation of organics was achieved in a single unit.
- Organic removal efficiency of 99% was reached in the tubing stream.

GRAPHICAL ABSTRACT

ABSTRACT

Effective biological treatment of high salt content wastewater requires consideration of both salt and organic toxicity. This study treated a synthetic saline wastewater containing NaCl (100 g L^{-1}) and 2,4-dimethylphenol (1.2 g L^{-1}) with a hybrid system consisting of a biological reactor containing spiral-coiled polymeric tubing through which the mixed feed was pumped. The tubing wall was permeable to the organic contaminant, but not to the salt, which allowed transfer of the organic into the cell-containing bioreactor contents for degradation, while not exposing the cells to high salt concentrations. Different grades of DuPont Hytrel polymer were examined on the basis of organic affinity predictions and experimental partition and mass transfer tests. Hytrel G3548 tubing showed the highest permeability for 2,4-dimethylphenol while exerting an effective salt barrier, and was used to verify the feasibility of the proposed system. Very high organic removal (99% after just 5 h of treatment) and effective biodegradation of the organic fraction of the wastewater (>90% at the end of the test) were observed. Complete salt separation from the microbial culture was also achieved.

© 2017 Elsevier B.V. All rights reserved.

1. Introduction

The treatment of saline wastewater arising from a broad cross-section of industries such as agro-food operations, and textile processing poses significant challenges (Lefebvre and Moletta, 2006). This is because saline wastewater contains not only high salt (inorganic) concentrations but is also contaminated with a variety of organic compounds. Such a combination of inorganic/organic pollutants makes the design of effective treatment facilities particularly difficult as overall treatment must be able to obtain high removal efficiency of the organic components while also achieving efficient removal or recovery of the salt. This challenge is generally met by combining a number of separate treatment operations (e.g. chemical/physical for salt removal and biological for organics removal) into an integrated, multi-step treatment process, as no single treatment process for both types of contaminants is commercially available.

* Corresponding author.
E-mail address: tomei@irsa.cnr.it (M.C. Tomei).

High salt concentrations exert inhibitory/toxic effects on both aerobic and anaerobic microorganisms during biodegradation processes, and laboratory-scale research has examined a variety of mitigating strategies. One common approach is to acclimatize the microbial culture to progressively increasing salt concentrations, however acclimatization times can be quite long depending on the salt concentration employed, and microbial salt resistance can be easily lost if the salt concentration selection pressure is removed (Ketola and Hiltunen, 2014). More stable and durable process performance may also be achieved through bio-augmentation with halophilic species, i.e. microorganisms able to survive in high salinity environments, however, it is not easy to maintain dominance of such species in mixed culture systems (Cui et al., 2016). Neither strategy appears to be suited to implementation into an actual industrial wastewater treatment process.

An alternative approach to deal with saline (i.e. salt plus organics) wastewater was demonstrated by Livingston (1993a, 1993b), using extractive membrane bioreactors (EMBs). In these systems, commercially available silicone rubber (polydimethylsiloxane, or PDMS) tubing was coiled inside a stirred tank bioreactor with the salt/organic solution passing "tube-side", and an adapted microbial culture being present "bioreactor-side". Given the highly hydrophobic nature of PDMS, only the organic compound was able to permeate the tubing wall, and was degraded in the bioreactor, with the high salt concentration being retained within the tubing and thereby not being in contact with the microbial population. Brookes and Livingston (1995) provided a demonstration of concept for such a tubing reactor and a detailed mass transfer study involving partition coefficients (PCs), mass transfer coefficients and solute diffusivity. This simple, single-stage system was able to address the challenge of salt separation, thereby facilitating the biological treatment of saline wastewater in a single unit operation.

The use of polymers for the treatment of contaminants has also been demonstrated in alternative contacting configurations, namely Two-Phase Partitioning Bioreactors (TPPBs) in which small polymer pellets are utilized to absorb and release toxic organic contaminants in response to microbial metabolic demand. Although not directly applicable to the treatment of saline wastewater, such an approach has demonstrated that an extremely broad range of polymer types can be effectively exploited for treating numerous types of organic contaminants present in water, air, and soil environments (Yeom et al., 2010; Munoz et al., 2012; Tomei et al., 2015). As noted, although the PDMS silicone tubing used by Livingston (1993a, 1993b) is commercially available, PDMS is permeable only to highly hydrophobic organic solutes, which severely limits the application of this polymer/tubing type to the treatment of a limited range of organics in saline wastewater.

Given the very broad range of available polymer chemistries, a significant opportunity exists to form different types of polymers into tubing, and to use this tubing in a manner similar to that of Livingston (1993a, 1993b). DuPont (Canada) provided extruded tubing comprised of various grades of Hytrel polymers of different chemistries, and Craig and Daugulis (2014) demonstrated continuous tubing-bioreactor operation in a biosynthetic process for the production and detoxification of benzaldehyde. Although this demonstration was not focused on wastewater treatment per se, it did confirm the flexibility of application that non-PDMS polymer tubing possesses to a range of organic solutes, and provides a thermodynamic explanation for this.

The work of Craig and Daugulis (2014), on pervaporation, was successfully extended to the treatment of wastewater containing toxic compounds in two recent studies (Tomei et al., 2016; Tomei et al., 2017) focused on a demonstration of the operating principle for application to industrial wastewater treatment. The system was tested on a solution of a single target compound (4-chlorophenol) to verify effective mass transfer through the tubing walls and the removal/biodegradation efficiency of the target compound. Different loading conditions were applied, and enhanced performance of the biomass compared to a conventional bioreactor was demonstrated. In a more recent paper (Mosca Angelucci et al., 2017), the hybrid bioreactor was successfully applied to a synthetic tannery wastewater containing toxic dichromate.

The objective of the current work is to verify the applicability to another "hostile wastewater" whose biological treatment is hindered by the high salt content and biorefractory organic contaminants.

According to previous studies (Juang et al., 2009; Juang and Tseng, 2010; Tan et al., 2017) a phenolic compound, 2,4-dimethylphenol (1200 mg L^{-1}) and NaCl (100 g L^{-1}) have been employed to simulate the aromatic organic fraction and the salt content in synthetic wastewater. With this synthetic wastewater, various grades of Hytrel were examined using partition and mass transfer tests to verify effective delivery of the organic substrate from the stream flowing in the tubing, and the mass transfer barrier effect exerted by the tubing walls for the salt. As a comparison, silicone rubber (PDMS) was also assessed in terms of its suitability as tubing material for the selected organic contaminant. The study concluded with a biological test to confirm system validation.

2. Material and methods

2.1. Polymers

DuPont (Canada) kindly provided tubing samples that had been extruded from 3 different grades of Hytrel, whose properties are shown in Table 1.

In order to remove possible impurities arising from the fabrication or extrusion process, each tubing sample was pre-treated by an initial washing step (20 min, 320 rpm) with a 1:1 distilled water:methanol solution, followed by 5 subsequent washing steps (20 min, 320 rpm) with distilled water. Once washed, the tubing was air dried overnight before its use. Commercial Masterflex 96400-15 silicone tubing from VWR International (Italy) was employed for the solute sorption test on PDMS.

2.2. Wastewater

The synthetic saline wastewater was prepared by dissolving in tap water ~1200 mg L^{-1} of 2,4-dimethylphenol (DMP), (corresponding to ~3000 mg$_{COD}$ L^{-1}), and 100 g L^{-1} of NaCl. All reagents were analytical grade and were purchased from Sigma-Aldrich (USA).

2.3. C-TPPB bioreactor

The C-TPPB system, as previously described (Tomei et al., 2016), consisted of a 2 L glass vessel equipped with a magnetic stirrer and temperature control (25 °C for mass transfer tests and 28 °C for biological tests). The reactor head was equipped with a cylindrical metal grid support onto which the polymeric tubing (length 350 cm) was coiled; when assembled all of the tubing length was completely submerged. The spiral coiled tubing was connected to a micro pump (Watson-Marlow, Cellai, Italy), suitable for flow rates in the range of

Table 1
Characterization data of the Hytrel polymeric tubing (DuPont Material Data Center, 2017).

Hytrel grade	8206	G3548	4056
Density (g cm^{-3})	1.17	1.15	1.16
Tg[a] (°C)	−59	−40	−50
Tm[b] (°C)	200	157	152
Tp[c] (°C)	230	180	190
Flexural modulus (MPa)	80	25	60
Hardness (Shore D)	38	26	40
Water absorption[d] (%)	30–35	5–7	0.6–0.7
Internal diameter (mm)	5	6	7
External diameter (mm)	6	7	8

[a] Glass transition temperature.
[b] Melting temperature.
[c] Melting temperature in processing (extrusion).
[d] Immersion 24 h.

0.01–0.1 L h^{-1}, in order to continuously feed the wastewater solution into the tubing on a once-through basis. The influent flowrate was fixed at a value of ~0.018 L h^{-1} and the effluent was collected in a sealed glass flask. The applied flow rate, due to the different internal diameters of the different Hytrel tubing (see Table 1), resulted in tubing HRTs in the range of 4–7.5 h.

For the biotic test, the C-TPPB system was equipped with additional devices for monitoring and controlling the pH (set point value 7.5) and dissolved oxygen (DO) concentration (set point 3–4 mg L^{-1}). 1 M NaOH solution was dosed with a peristaltic pump for pH control while DO was controlled via an on/off strategy by varying the air supply. The bioreactor was connected to a computer through an interface for data acquisition: DO data, recorded each 15 s, allowed the estimation of the Specific Oxygen Uptake Rate (SOUR) according to a procedure previously applied in Tomei et al. (2017).

A schematic representation of the bioreactor prototype is reported in Fig. 1.

2.4. Experimental tests

2.4.1. Sorption-desorption tests

After cutting the polymer tubing into small pieces (average size of ~2–3 mm), sorption tests were conducted by contacting fixed amounts of polymers (the three Hytrel grades and PDMS) with a well-mixed (320 rpm) solution (40 mL) of ~120 mg DMP L^{-1} or NaCl (~100 g L^{-1}) in sealed flasks. Liquid samples were periodically withdrawn from the flasks and analysed for DMP and NaCl concentrations. After reaching equilibrium, the tubing pieces were separated from the solution and contacted with fresh water for the desorption test (performed with the same procedure as the sorption test and by keeping the same polymer-to-water ratio). Sorption and desorption tests were repeated at different polymer-water ratios in the range of 0.3–1.3%. In this way, two independent series of measures (5 tests for sorption and 5 for desorption) were provided for determination of the solute partition coefficients, as the "end point" equilibrium concentrations, with mass balance equations.

2.4.2. Mass transfer tests

In order to investigate the mass transfer properties of the tested Hytrel grades, abiotic experiments on the mass transfer of salt and DMP contained in the saline wastewater were performed on the C-

Fig. 1. Schematic representation of the experimental apparatus for the C-TPPB (DO: dissolved oxygen probe; pH: pH probe; T: temperature probe).

TPPB system. The tubing was initially filled with the synthetic wastewater, and the bioreactor with tap water, then the tubing was continuously fed for 24 h. Samples were periodically taken from the liquid phase inside the reactor, the tubing effluent and the liquid collected at the end of the tubing, then analysed for DMP and chloride concentrations. Sampling was performed at one hour intervals for the first 6 h, then at 22 and 24 h.

2.4.3. Biological test

The C-TPPB with the selected Hytrel grade G3548 was employed for the biological test. The bioreactor was inoculated with a microbial consortium previously acclimated to phenolic compounds and grown in an aerobic Sequencing Batch Reactor (SBR) on DMP and a small fraction of biogenic substrate (sodium acetate) in the first month, then on DMP as the sole energy and carbon source. Influent and effluent concentrations and removal efficiency in the SBR are reported in Table S1 of the Supplementary material. The initial concentration of DMP in the bioreactor was ~0 and the biomass amount expressed as Volatile Suspended Solids (VSS) concentration was 930 mg$_{VSS}$ L^{-1}. A mineral salt medium (Williams and Unz, 1989) was added to the liquid phase of the bioreactor to provide the required nutrients and microelements to the biomass: the dosage of the mineral medium was evaluated taking into account the organic load of the feed solution on daily basis to get a C:N:P ratio of 100:5:1. The experimental procedure followed during the test was the same as described for the mass transfer test but, because of the longer duration of the test, a 2 hour sampling frequency was applied. Additional analysis, i.e. conductivity, OD$_{600}$ and COD were done on the samples from the liquid phase of the bioreactor.

Furthermore, to estimate the residual amount of DMP absorbed by the polymer tubing, at the end of the experiment, a solvent washing-extraction tests were performed on the used tubing. A solution of water:methanol (4:1) was continuously fed through the tubing until a negligible amount of DMP was detected in the effluent (≤1 mg L^{-1}). The washing solvent was collected and analysed for DMP concentration, and the measured data were employed for mass balance.

Biological test was performed in duplicate at different duration times 96 h (R1) and 36 h (R2).

2.5. Analysis

Analysis of DMP concentration on aqueous samples (both in water and water-methanol solution) were performed after centrifugation (8 min at 13,000 rpm) by using a UV/VIS spectrophotometer (PerkinElmer, Lambda 25) at λ = 280 nm.

Biomass concentration in the bioreactor was determined by measuring OD$_{600}$ using the same spectrophotometer, and by VSS concentration according to Standard Methods (APHA, 2012).

COD cell tests (MERCK-referring to EPA 410.4 method), based on potassium dichromate oxidation and spectrophotometric determination (Spectroquant Nova30), were employed for COD measurements in centrifuged (8 min at 13,000 rpm) aqueous samples.

The chloride concentration in the liquid phase of the bioreactor and in tubing effluent was measured by an ionic chromatograph (DX-100 DIONEX) according to Standard Methods (APHA, 2012).

2.6. Methods for polymer selection

Polymers are comprised of different monomers (and in many cases by co-polymers in different configurations such as block and random structures). This not only provides polymers with different physical properties (e.g. hardness) but different chemistries that would be important in interactions with solutes such as DMP. Silicone rubber is comprised substantially of PDMS (polydimethyl-siloxane) while Hytrel is a polyether-ester block copolymer containing terephthalate. This difference in chemistry explains the differences in solute affinity, and consequent different performance in the tubing system. Polymer selection
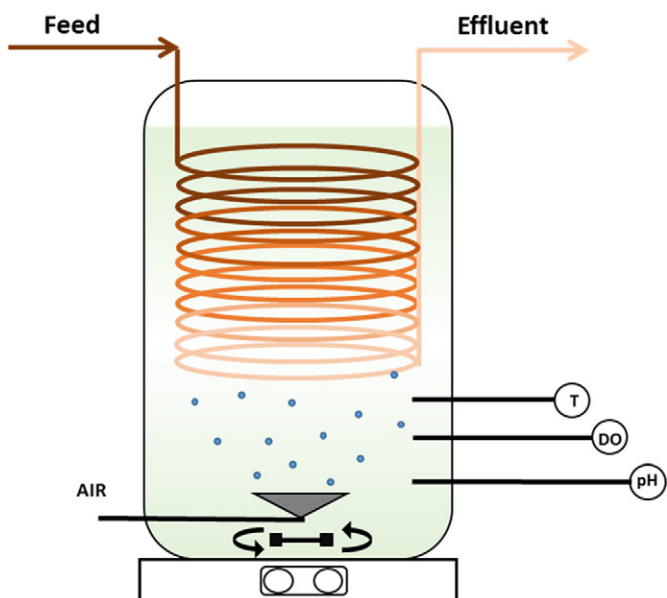
has been performed in two sequential steps: the first one was based on a theoretical estimation of polymer-solute affinity, followed by experimental mass transfer and partition tests undertaken to confirm the results under the applied operating conditions.

2.6.1. Estimation of the polymer-solute affinity

The prediction of solute-polymer affinity has recently been examined in considerable detail using first principles thermodynamic approaches. Beginning with the notion that "like attracts like", Bacon et al. (2014) examined activity-coefficient based approaches to predict solute-polymer interactions for a range of polymers, and solutes spanning a variety of types (e.g. aromatics, alcohols, ethers and esters). The simplest approach, using Hildebrand solubility parameters as a proxy for "like attracts like" used the notion that components with similar solubility parameters (which have been widely tabulated) have mutual affinity, and by evaluating the difference between solubility parameters, relative affinity can be estimated.

The second approach considered Hansen solubility parameters, including three different interactions among molecules: nonpolar (dispersion interactions), polar (dipole-dipole interactions) and hydrogen bonding interactions. The three interactions are quantified through the three related solubility parameters δ_D, δ_P and δ_H, which are related to the overall Hildebrand solubility parameter δ_{HSP} proposed for regular solutions by the following equation:

$$\delta_{HSP} = \sqrt{(\delta_D{}^2 + \delta_P{}^2 + \delta_H{}^2)} \qquad (1)$$

Closer δ_{HSP} values between a polymer and a solute indicate higher molecular similarity, thus higher affinity.

Another parameter proposed by Hansen to predict the affinity is the solubility parameter distance Ra given by:

$$Ra = \sqrt{\left[4 \cdot (\delta_{D1}-\delta_{D2})^2 + (\delta_{P1}-\delta_{P2})^2 + (\delta_{H1}-\delta_{H2})^2\right]} \qquad (2)$$

where the subscripts 1 and 2 indicate the polymer and the compound respectively. The smaller the Ra distance between a polymer and a solute, the greater the thermodynamic affinity.

Although such methods have proven to be successful in a first attempt to characterize polymer-solute interactions (Poleo and Daugulis, 2014; Dafoe and Daugulis, 2014), it is also important to recognize certain limitations such as the fact that the solubility parameter methods deal only with binary systems, and they do not consider the effect of the presence of water. Additionally, polymer properties related to solute uptake can also be affected by factors such as crystallinity, end group effects and molecular weight. This is why, after the first screening based on affinity estimation, an experimental characterization is required for a reliable evaluation of the performance of the selected polymers with the target solutes.

2.6.2. Experimental characterization

The resistance-in-series model has been applied and demonstrated suitable in previous studies on EMBs (Livingston et al., 1998; Manconi and Lens, 2009) or pervaporation systems (Overington et al., 2009) to evaluate the mass transfer coefficient in tubular membrane systems. The proposed bioreactor for saline wastewater has a similar configuration, thus the same calculation procedure can be applied. According to the model, the overall mass transfer coefficient (K_O) is expressed as the sum of the three different contributions: the two resistances in the liquid film inside ($1/k_t$) and outside ($1/k_r$) the tubing, and the diffusion resistance across the tubing wall:

$$\frac{1}{K_O} = \frac{1}{k_t} + \frac{r_i \; \ln\left({r_e}/{r_i}\right)}{D \cdot PC} + \frac{r_i}{r_e k_r} \qquad (3)$$

where PC is the partition coefficient, D the diffusion coefficient, and r_e and r_i are the external and internal tubing radius, respectively. Assuming that the liquid film resistances on both sides of the tubing are negligible, the resulting expression for K_O is:

$$\frac{1}{K_O} = \frac{r_i \; \ln\left({r_e}/{r_i}\right)}{D \cdot PC} \qquad (4)$$

The tubing permeability P is defined (Han et al., 2002) as:

$$P = D \cdot PC \qquad (5)$$

The two parameters PC and K_O can be determined by independent experimental tests i.e. partition and mass transfer tests, respectively, as it has been done in the present study.

The PC was evaluated by data fitting of sorption and desorption tests performed with different polymer-to-water ratios from the following mass balance equation:

$$PC = \frac{[(C_0/C)-1]}{[M_{pol}/(\rho \cdot V_w)]} \qquad (6)$$

where C_0 and C are the initial and final liquid concentration of the solute, respectively, M_{pol} is the mass of polymer, ρ is the density and V_w the liquid solution volume.

According to Freitas Dos Santos and Livingston (1995), the overall mass transfer coefficient, has been determined as:

$$K_O = \frac{F_T \; \ln\left(\frac{C_{T,in}-C_R}{C_{T,out}-C_R}\right)}{2\pi r_i L} \qquad (7)$$

where F_T is the tubing flowrate, $C_{T,in}$ and $C_{T,out}$ are the influent and effluent tubing concentrations, respectively, C_R is the concentration in the reactor and L is the tubing length. Eq. (7) is applicable for tubing in plug flow regime and by assuming that the bulk fluid in the bioreactor is perfectly mixed, as it was verified in our case.

Once determined PC and K_O, knowing the tubing dimensions, the diffusivity and permeability were evaluated by Eqs. (4) and (5).

3. Results and discussion

3.1. Polymer selection

3.1.1. Polymer-solute affinity

Solubility parameters have been employed for a first prediction of the polymer-solute affinity for DMP by the various Hytrel grades and PDMS to have a qualitative evaluation of their suitability for this C-TPPB application. The Hildebrand solubility parameter δ_{HSP} and the Ra distance have been calculated and are shown in Table 2.

Table 2
Solubility parameters and Ra distance for DMP-Hytrel and DMP-PDMS (δ and Ra units are MPa$^{1/2}$). Hildebrand solubility parameter δ_{HSP} and Ra distance are reported in bold.

Compound/polymer	δ_D	δ_P	δ_H	δ_{HSP}	Ra	Reference
2,4 xylenol (DMP)	18.6	5.1	9.5	**21.5**		Hansen (2007)
Hytrel 8206	16.46	5.93	4.40	**18.1**	**6.7**	Poleo and Daugulis (2014)
Hytrel G3548	16.43	5.01	4.03	**17.7**	**7.0**	Poleo and Daugulis (2014)
Hytrel 4056	16.78	6.71	3.04	**18.3**	**7.6**	HSPiP software v3.1 (2017)
PDMS	12.3	0.4	0.3	**12.3**	**16.3**	Hansen (2007)
				13.4–15.1[a]		Mark (1999)

[a] Range of values depending on molecular weight and temperature.

Both metrics of affinity i.e. more similar δ_{HSP} values and lower Ra distances between DMP and tubing components give a first indication of a higher affinity of Hytrel with the target solute, in comparison to PDMS.

3.1.2. Sorption-desorption tests

The higher affinity of Hytrel for the target compound was also confirmed by the experimental sorption test results demonstrating that for PDMS no appreciable DMP sorption was detected for a 24 hour interval time. The unsuitability of PDMS (silicone rubber) tubing for solutes possessing some hydrophilicity has also been noted for the case of phenol in previous studies (Brookes and Livingston, 1995; Fam and Daugulis, 2012), which demonstrated that PDMS has virtually no affinity for phenol. In contrast, Hytrel 8206 has been shown to possess high affinity for such compounds, giving a relatively high PC for phenol of 43 as reported by Fam and Daugulis (2012).

Fig. 2 shows the sorption (Fig. 2a) and desorption (Fig. 2b) results for the three tested Hytrel grades in terms of DMP concentration profiles vs. time. Sorption and desorption rates are important in that their characteristic times should not be the limiting step for the biological processes. All the tested polymers demonstrated equilibrium in about 5 h, which is of the same time scale as biological processes.

In the sorption tests performed for NaCl, no transfer into the polymer was observed for all the three Hytrel grade tested, as demonstrated by the negligible deviation of the chloride concentration ≤0.7% (in respect to the initial value) detected during the entire test (24 hour contact time). For this reason, the subsequent desorption test was not performed with the tubing pieces used for sorption test with NaCl.

PCs for DMP were evaluated according to the procedure for data analysis reported in Tomei et al. (2009) by linear fitting of the partition test data. Experimental data and mass balance for PC calculation are reported in the Supplementary material (par. S1 and Fig. S1). Evaluated PC
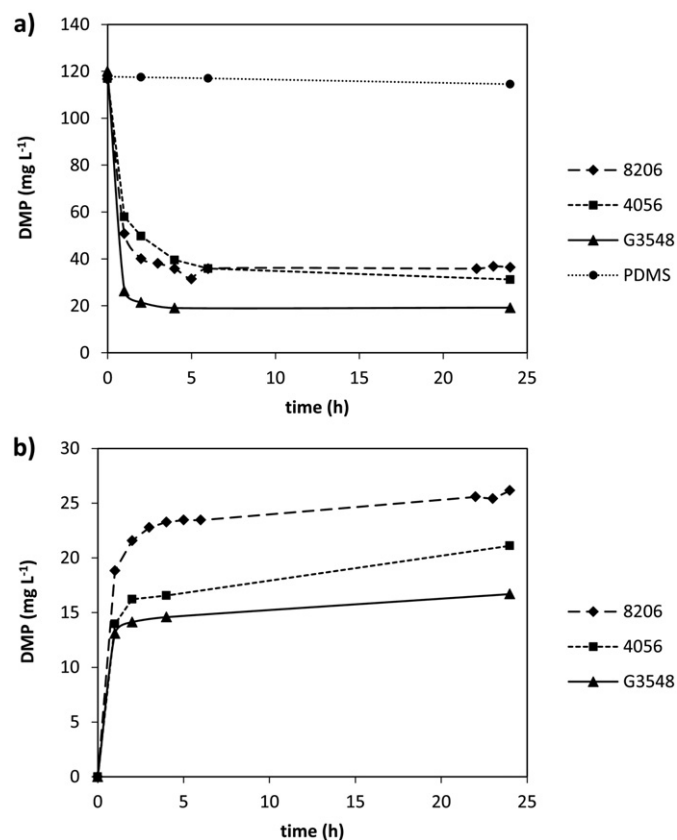
data are reported in Table 3. Good agreement is observed for the two PC values determined independently in the sorption and desorption tests.

High PC values were obtained for all the three Hytrel polymers, which confirmed their affinity for DMP. Partition data for the granular Hytrel 8206 and G3548 with the same phenolic compound were determined in a previous study (Tomei et al., 2011): 201 and 543 were respectively the PC values estimated in that case, i.e. quite close to the values found in this study. This finding confirms that the extrusion processing of the polymeric materials did not influence the partition properties of the polymers, as already observed for 4-chlorophenol (Tomei et al., 2016) and for benzaldehyde (Craig and Daugulis, 2014).

3.1.3. Mass transfer tests

Mass transfer tests were undertaken to verify both the effective mass transfer of DMP and the barrier effect for the salt with the same apparatus to be employed in the biological test. At this step of the experimental programme, we determined that the Hytrel 4056 tubing, for this tubing wall thickness, was not suitable to be coiled on the support due to kinks in the tubing making it difficult to ensure the continuous constant internal flow. This finding may be justified with the high values of flexural modulus and hardness of Hytrel 4056, as shown in Table 1. Although Hytrel 8206 shows similar values of these both parameters, this latter polymer was easier to manipulate, probably due to the smaller inner and outer diameter with respect to 4056. For this reason, the polymer selection continued with the two other Hytrel grades.

The results of the mass transfer tests for the two polymers are reported in Figs. 3 and 4 for DMP and chlorides, respectively, and are plots of the concentration in the tubing effluent and in the liquid phase of the reactor. There were no differences for the two polymers regarding the transfer of DMP, while a different behaviour is observed for chloride. No appreciable chloride transfer was observed for Hytrel G3548, while Hytrel 8206 showed an increase of the chloride concentration in the reactor and a corresponding decrease of the concentration in the tubing effluent.

A possible explanation of this finding may be the high water content of Hytrel 8206 (see Table 1), which facilitates the transfer of chloride from the tubing walls to the external liquid phase. Even if the concentration in the reactor after 24 h is about two orders of magnitude lower than the concentration in the tubing, the chloride accumulation during the continuous operation could be detrimental for the biomass. To avoid this negative effect, periodic settling and extraction of the supernatant from the bioreactor is required and this would imply the interruption of the continuous operation.

3.1.4. Selection criteria and results

According to Han et al. (2002) parameters influencing the selection of the most suitable polymer grade for the case under examination are summarized in Table 4.

PC and K_O have been evaluated by correlating independent experiment data with Eqs. (6) and (7), respectively, then diffusivity and permeability coefficients have been estimated. The salt transfer has been expressed as percent ratio between the salt amount in the reactor at the end of the experiment and the fed amount.

As regards D, there are no data available for DMP, although a comparison with diffusivity data reported for phenol (Pittman et al., 2015) and 2,4-dichlorophenol (Tomei et al., 2012), for different Hytrel polymer beads, showed that the values calculated in this study are of the same order of magnitude of the literature data estimated for conventional solid-liquid TPPBs.

Fig. 2. DMP concentration profiles in sorption (a) and desorption (b) tests. DMP initial concentration: ~120 mg L^{-1}; polymer was in small pieces (average size ~2–3 mm). Polymer-to-water ratio = 1.3%.

Table 3
PC values for the three tested Hytrel grades in sorption and desorption tests.

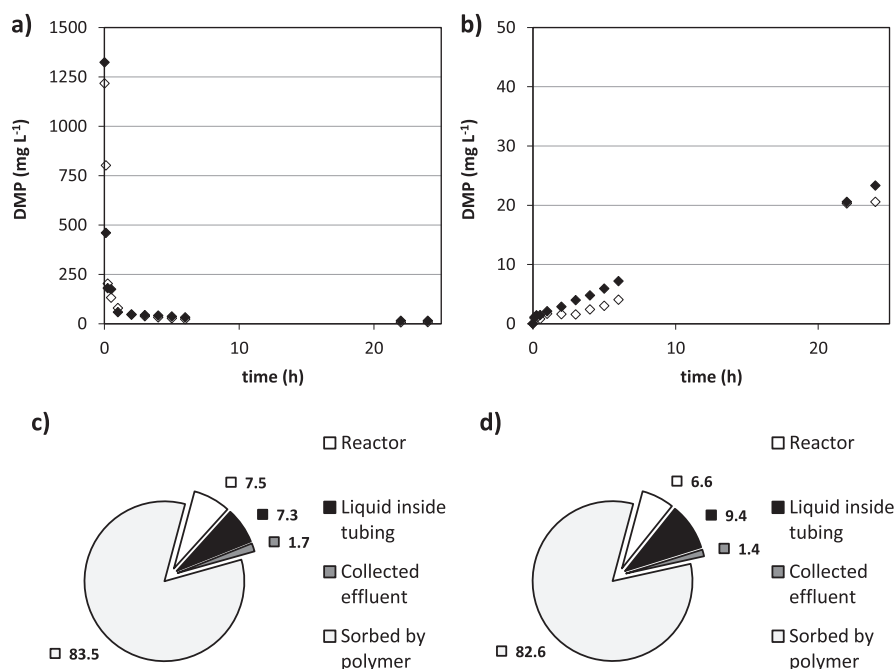| Test	Hytrel 8206	Hytrel G3548	Hytrel 4056
Sorption	179	423	225
Desorption	168	406	256

Fig. 3. DMP concentration profiles in the tubing effluent (a) and in the liquid phase in the reactor (b) during mass transfer tests: filled symbols for Hytrel 8206 and empty symbols for G3548. Distribution (as percentage with respect to the fed amount) of DMP at the end of the test with Hytrel 8206 (c) and Hytrel G3548 (d). DMP initial concentration: 1323.8 and 1217.3 mg L^{-1} for 8206 and G3548, respectively.

The two screened Hytrels show values of the same order for K_O, D and P, so the higher PC value and the absence of salt transfer through the tubing walls motivated the selection of Hytrel G3548 for the biological experiment in C-TPPB. Furthermore, as already pointed out, additional reasons to select Hytrel G3548 are the lowest values of hardness and flexural modulus (see Table 1), both of them positively affecting the suitability of coiling the tubing around the support inside the bioreactor.

3.2. Biological test

Hytrel G3548 was employed in the biological test performed continuously under the same operating conditions of the mass transfer test, allowing a direct comparison of the results and the evaluation of the biodegradation of the organic fraction in the saline wastewater. Fig. 5 shows the concentration profiles for the two replicates of the DMP in the bioreactor (a) and in tubing effluent (b). Very good reproducibility of the results is observed for the two replicates. Corresponding profiles detected in the mass transfer tests are reported for comparison. Times of

the order of 24–48 h are sufficient to achieve a stable response of the bioreactors with removal efficiency of 99% in the tubing stream. With reference to R1 test, the DMP concentration in the tubing effluent was 1.6 ± 0.4 mg L^{-1} for the time interval of 48–100 h. The DMP concentration in the bioreactor, in the same time interval, was 3.1 ± 0.7 mg L^{-1}, that is about one order of magnitude lower than the value (20.45 ± 0.15 mg L^{-1}) measured in the abiotic tests, and this is a first demonstration of DMP biodegradation. Additional evidence of DMP biodegradation is given by the low COD values detected in the bioreactor in the period 6–100 h equal to 15.2 ± 10 mg L^{-1}, which are consistent with the residual DMP concentration values.

The typical feature of TPPBs i.e. the transfer of substrate from the partitioning phase (in this case the polymeric tubing) driven by the metabolic process is highlighted by the two concentration profiles in the tubing effluent in Fig. 5b where a consistent enhancement of DMP transfer is observed.

To confirm the barrier effect of the tubing for NaCl, in parallel to DMP monitoring, the chloride concentration was followed in the tubing and in the bioreactor. Data of R1 test are reported in Fig. 6.
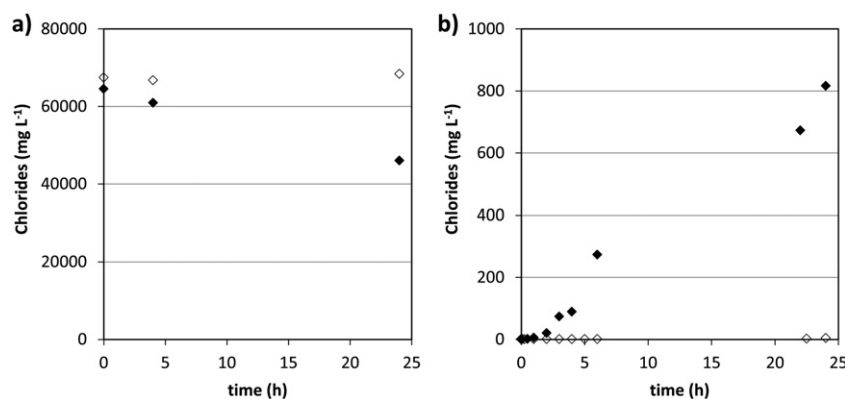

Fig. 4. Chloride concentration profiles in the tubing effluent (a) and in the liquid phase of the reactor (b) during mass transfer tests: filled symbols for Hytrel 8206 and empty symbols for G3548. Chloride initial concentration: 64.5 and 67.4 g L^{-1} for 8206 and G354, respectively.

Table 4
Polymer properties considered in the Hytrel grade selection (n.d. = not determined).

Parameter	Hytrel 8206	Hytrel G3548	Hytrel 4056
PC[a]	173.8	414.8	240.5
K_o (m s^{-1})	$4.3 \cdot 10^{-7}$	$5.3 \cdot 10^{-7}$	n.d.
D (cm^2 s^{-1})	$1.1 \cdot 10^{-8}$	$0.6 \cdot 10^{-8}$	n.d.
P (m^2 s^{-1})	$2.0 \cdot 10^{-10}$	$2.5 \cdot 10^{-10}$	n.d.
Salt transfer (%)	+5%	–	n.d.

[a] Calculated as average data of sorption and desorption tests.

No significant variation was observed in the tubing effluent during the entire test (Fig. 6b).

Also in the bioreactor the variation was minimal (from 3 to 6 mg L^{-1}) and due to the addition of the mineral medium (Fig. 6a). Parallel measurements of conductivity, whose trend is mainly due to chloride variation in the bioreactor, were also performed and data shown in Fig. 6a.

Biomass evolution and oxygen consumption during the biological test R1 are shown in Fig. 7 reporting the optical density (OD$_{600}$) and SOUR vs time. The consistent biomass increase (~80%) highlights the DMP biodegradation. SOUR values are consistently higher than the values measured for the endogenous respiration in the SBR fed with DMP, i.e. 2.1 ± 0.2 mg$_{O2}$ g$_{VSS}^{-1}$ h^{-1} (as reported by dashed line in Fig. 7), estimated as average value from data of 7 working cycles in the last period of biomass maintenance in SBR before biological test.

Fig. 6. Chloride concentration and conductivity in the bioreactor (a) and chloride concentration in the tubing and the collected effluent (b) during biological test R1. The arrows in (a) indicate the addition of mineral salt medium in the bioreactor.

From this value an endogenous decay coefficient (b) equal to 0.04 ± 0.005 d^{-1} has been estimated. Furthermore, we observed a slight decrease in the second part of the test corresponding to the increase of the biomass concentration. This trend can be explained considering that for a greater biomass amount in the bioreactor, a lower specific oxygen consumption is required for degrading the same substrate load. Moreover, from the tubing washing at the end of the test, the DMP retained in the polymer was estimated as ~6.4% of the fed amount, thus demonstrating a minor contribution of absorption to DMP removal.
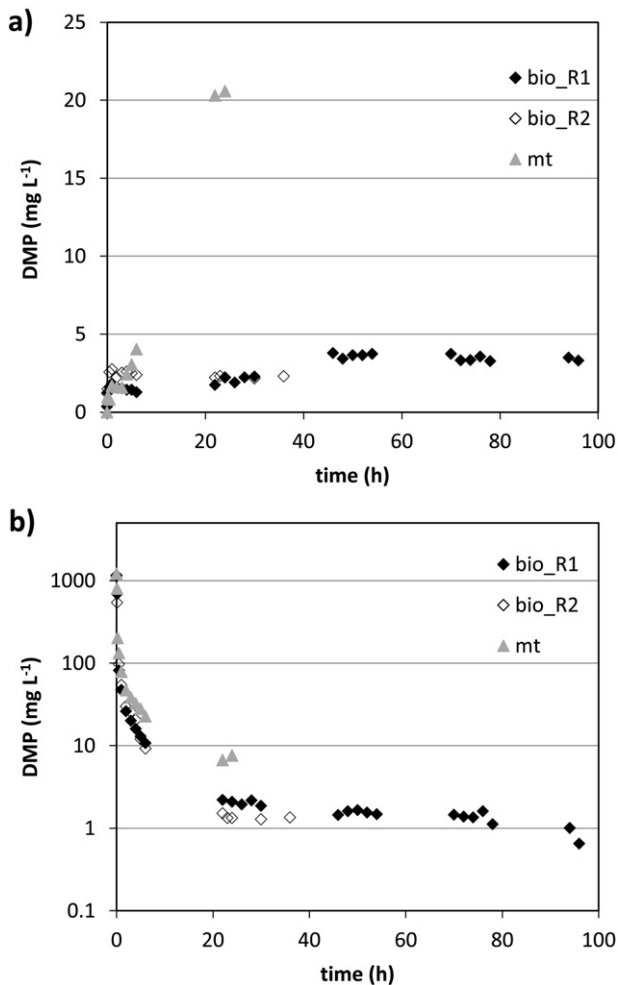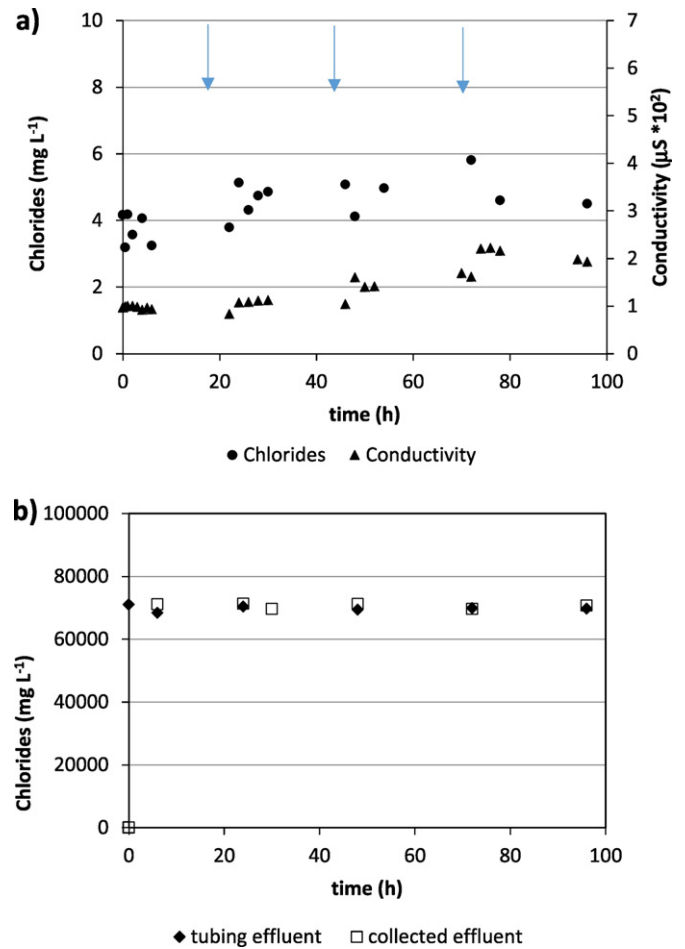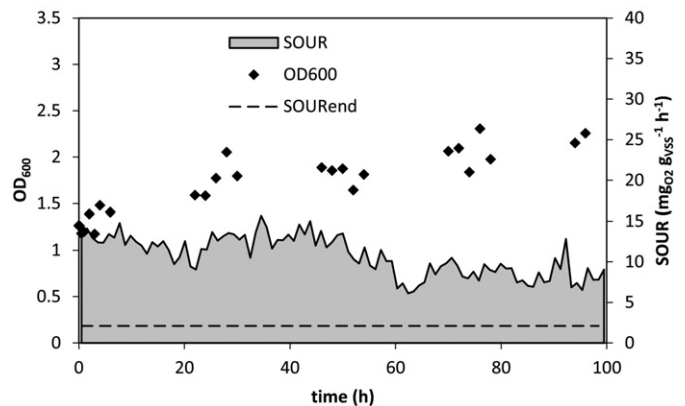

Fig. 5. DMP concentrations vs. time in the bioreactor (a) and tubing effluent (b) during mass transfer (mt) and biological (bio_R1 and bio_R2) tests performed with Hytrel G3548. DMP initial concentration: 1217.3 and 1161.3 mg L^{-1} for bio_R1 and bio_R2 tests, respectively, and 1144.2 mg L^{-1} in mass transfer.

Fig. 7. Biomass and oxygen consumption evolution in the bioreactor: OD$_{600}$ and SOUR vs. time during the biological test R1. Dashed line indicates the endogenous SOUR (SOURend) calculated for SBR.

The DMP and biomass mass balance referring to the test duration time (Δt) gives an observed yield coefficient (Y_{obs}) of 0.35 (on COD basis, i.e. $COD_X \, COD_{DMP}^{-1}$) which is in the range of values commonly detected in activated sludge plants. Details on the calculation procedure are reported in the Supplementary material (par. S2).

4. Conclusions

In this study, the feasibility of a hybrid bioreactor extending the TPPB concept to include the use of rationally-selected polymer tubing is demonstrated for treating highly saline wastewater. Tubing allows for the spontaneous separation of a high salt concentration from a biodegradable organic, resulting in selective transport of the organic to a cell-containing phase, while sequestering the salt.

This configuration gives highly effective biological removal (i.e. >99% after 5 h of treatment) of a saline-organic waste solution that would not otherwise be degraded without the use of the polymer tubing. Actual biodegradation and not only sorption in the tubing is confirmed by the measurement of the DMP amount retained in the tubing (≤7% of the fed amount), in addition to the COD balance and the oxygen consumption.

A first principles thermodynamic approach that considers solubility parameters was effective in explaining why Hytrel G3548, rather than PDMS/silicone rubber, is suitable for DMP, and suggests that this approach can be used as first selection step of polymers to be formed into tubing for application to other specific organic contaminants.

Additional investigations are underway to verify the system response in more critical operating conditions such as in the presence of high organic loads and highly saline wastewater.

Acknowledgements

Appendix A. Supplementary data

Supplementary data to this article can be found online at http://dx.doi.org/10.1016/j.scitotenv.2017.05.042.

References

APHA (American Public Health Association), 2012. Standards Methods for the Examination of Water and Wastewater. 22nd ed. (Washington, DC).

Bacon, S.L., Parent, J.S., Daugulis, A.J., 2014. A framework to predict and experimentally evaluate polymer-solute thermodynamic affinity for two-phase partitioning bioreactor (TPPB) applications. J. Chem. Technol. Biotechnol. 89, 948–956.

Brookes, P.R., Livingston, A.G., 1995. Aqueous-aqueous extraction of organic pollutants through tubular silicone rubber membranes. J. Membr. Sci. 104, 119–137.

Craig, T., Daugulis, A.J., 2014. Strategies for improved bioproduction of benzaldehyde by *Pichia pastoris* and the use of Hytrel as tubing material for integrated product removal by in situ pervaporation. Biochem. Eng. J. 82, 97–104.

Cui, Y.W., Zhang, H.Y., Ding, J.R., Peng, Y.Z., 2016. The effects of salinity on nitrification using halophilic nitrifiers in a Sequencing Batch Reactor treating hypersaline wastewater. Sci. Rep. 6, 24825.

Dafoe, J.T., Daugulis, A.J., 2014. In-situ product removal in fermentation systems: improved process performance and rational extractant selection. Biotechnol. Lett. 36, 443–460.

DuPont, Material Data Center, 2017. https://dupont.materialdatacenter.com/profiler/B0Ejv/ accessed 17.03.17.

Fam, H., Daugulis, A.J., 2012. Substrate mass transport in two-phase partitioning bioreactors employing liquid and solid non-aqueous phases. Bioprocess Biosyst. Eng. 35, 1367–1374.

Freitas Dos Santos, L.M., Livingston, A.G., 1995. Novel membrane bioreactor for detoxification of VOC wastewaters: biodegradation of 1,2-dichloroethane. Water Res. 29, 179–194.

Han, S., Puech, L., Law, R.W., Steinke, J.H.G., Livingston, A.G., 2002. Selection of elastomeric membranes for the separation of organic compounds in acidic media. J. Membr. Sci. 199, 1–11.

Hansen, C.M., 2007. Hansen Solubility Parameters: A User's Handbook. second ed. CRC Press, Taylor & Francis Group, Boca Raton, FL.

HSPiP, Hansen Solubility Parameters in Practice, 2017. https://www.hansen-solubility.com last accessed 13.03.17.

Juang, R.S., Tseng, K.J., 2010. Experimental investigation of bio-removal of toxic organic pollutants from highly saline solutions in a triphasic system. J. Hazard. Mater. 178, 706–712.

Juang, R.S., Huang, W.C., Hsu, Y.H., 2009. Treatment of phenol in synthetic saline wastewater by solvent extraction and two-phase membrane biodegradation. J. Hazard. Mater. 164, 46–52.

Ketola, T., Hiltunen, T., 2014. Rapid evolutionary adaptation to elevated salt concentrations in pathogenic freshwater bacteria *Serratia marcescens*. Ecol. Evol. 20 (4), 3901–3908.

Lefebvre, O., Moletta, R., 2006. Treatment of organic pollution in industrial saline wastewater: a literature review. Water Res. 40, 3671–3682.

Livingston, A.G., 1993a. A novel membrane bioreactor for detoxifying industrial wastewater: I. Biodegradation of phenol in a synthetically concocted wastewater. Biotechnol. Bioeng. 41, 915–926.

Livingston, A.G., 1993b. A novel membrane bioreactor for detoxifying industrial wastewater: II. Biodegradation of 3-chloronitrobenzene in an industrially produced wastewater. Biotechnol. Bioeng. 41, 927–936.

Livingston, A.G., Arcangeli, J.P., Boam, A.T., Zhang, S.F., Marangon, M., Santos, L.F.D., 1998. Extractive membrane bioreactors for detoxification of chemical industry wastes: process development. J. Membr. Sci. 151, 29–44.

Manconi, I., Lens, P.N.L., 2009. Removal of H_2S and volatile organic sulfur compounds by silicone membrane extraction. J. Chem. Technol. Biotechnol. 84, 69–77.

Mark, J.E., 1999. The Polymer Data Handbook. first ed. Oxford University Press (1999).

Mosca Angelucci, D., Stazi, V., Daugulis, A.J., Tomei, M.C., 2017. Treatment of synthetic tannery wastewater in a continuous two-phase partitioning bioreactor: biodegradation of the organic fraction and chromium separation. J. Clean. Prod. 152, 321–329.

Munoz, R., Daugulis, A.J., Hernandez, M., Quijano, G., 2012. Recent advances in two-phase partitioning bioreactors for the treatment of volatile organic compounds. Biotechnol. Adv. 30, 1707–1720.

Overington, A.R., Wong, M., Harrison, J.A., Ferreira, L.B., 2009. Estimation of mass transfer rates through hydrophobic pervaporation membranes. Sep. Sci. Technol. 44, 787–816.

Pittman, M.J., Bodley, M.W., Daugulis, A.J., 2015. Mass transfer considerations in solid-liquid two-phase partitioning bioreactors: a polymer selection guide. J. Chem. Technol. Biotechnol. 90, 1391–1399.

Poleo, E.E., Daugulis, A.J., 2014. A comparison of three first principles' methods for predicting solute-polymer affinity, and the simultaneous biodegradation of phenol and butyl acetate in a two-phase partitioning bioreactor. J. Chem. Technol. Biotechnol. 89, 88–96.

Tan, S., Chen, X., Cui, C., Hou, Y., Li, W., You, H., 2017. Biodegradation of saline phenolic wastewater in a biological contact oxidation reactor with immobilized cells of *Oceanimonas sp.* Biotechnol. Lett. 39, 91–96.

Tomei, M.C., Annesini, M.C., Prpich, G.P., Daugulis, A.J., 2009. Biodegradation of 4-nitrophenol in a two-phase system operating with polymers as the partitioning phase. Environ. Sci. Technol. 43, 7105–7110.

Tomei, M.C., Rita, S., Mosca Angelucci, D., Annesini, M.C., Daugulis, A.J., 2011. Treatment of substituted phenol mixtures in single phase and two-phase solid-liquid partitioning bioreactors. J. Hazard. Mater. 191, 190–195.

Tomei, M.C., Annesini, M.C., Daugulis, A.J., 2012. 2,4-Dichlorophenol removal in a solid-liquid two phase partitioning bioreactor (TPPB): kinetics of absorption, desorption and biodegradation. New Biotechnol. 30 (1), 44–50.

Tomei, M.C., Angelucci, D.M., Ademollo, N., Daugulis, A.J., 2015. Rapid and effective decontamination of chlorophenol-contaminated soil by sorption into commercial polymers: concept demonstration and process modeling. J. Environ. Manag. 150, 81–91.

Tomei, M.C., Mosca Angelucci, D., Daugulis, A.J., 2016. Towards a continuous two-phase partitioning bioreactor for xenobiotic removal. J. Hazard. Mater. 317, 403–415.

Tomei, M.C., Mosca Angelucci, D., Daugulis, A.J., 2017. A novel continuous two-phase partitioning bioreactor operated with polymeric tubing: performance validation for enhanced biological removal of toxic substrates. J. Environ. Manag. 187, 265–272.

Williams, T.M., Unz, R.F., 1989. The nutrition of *Thiothrix*, type 021N, *Beggiatoa* and *Leucothrix* strains. Water Res. 23, 15–22.

Yeom, S.H., Daugulis, A.J., Lee, S.H., 2010. Bioremediation of phenol-contaminated water and soil using magnetic polymer beads. Process Biochem. 45, 1582–1586.