Journal of Environmental Management 125 (2013) 7–11

Contents lists available at SciVerse ScienceDirect

Journal of Environmental Management

journal homepage: www.elsevier.com/locate/jenvman

Feasibility of operating a solid–liquid bioreactor with used automobile tires as the sequestering phase for the biodegradation of inhibitory compounds

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A R T I C L E I N F O

Article history:
Received 29 January 2013
Received in revised form
21 March 2013
Accepted 28 March 2013
Available online 27 April 2013

Keywords:
Automobile tire re-use
Tire-TPPB system
Sequential bioreactor
Inhibitory compounds removal
Feasibility study

A B S T R A C T

Finding new uses for waste or discarded material is an important environmental goal; being able to use a waste material to treat another waste is an even more attractive objective, and this was the purpose of the present work. We previously showed that used automobile tires have an affinity for a toxic contaminant, dichlorophenol (DCP), absorbing and releasing it based on concentration driving forces. Here we have exploited this phenomenon by using used tires as the sequestering phase in a Two-Phase Partitioning Bioreactor (TPPB) to treat otherwise-toxic levels of DCP, far out-performing single phase operation in a sequencing batch bioreactor. A comprehensive examination of substrate loading, reactor exchange ratio, and tire fraction used, demonstrated that the tire-TPPB system could handle a 40% higher influent substrate loading and an increase of the exchange ratio value from 0.5 (prohibitive for single phase operation) to 0.7. Such improvement was obtained with a tire fraction ≤9%, comparable to that for commercial polymers previously employed in TPPBs. This study has opened the door to the identification of other waste plastics suitable for use in TPPBs for the treatment of recalcitrant organic contaminants.

1. Introduction

Low cost adsorbents have recently been utilized in wastewater treatment, for example, natural adsorbents such as wheat chaff, rice husk, sesame seed, sun flower and tea wastes have been used to remove lead (Kafia, 2011), acid-activated saw dust was used to adsorb dyes (Rahman et al., 2010), and treated agricultural wastes (char carbons, coconut husk carbons) and biosorbents (microbial biomass) were employed for arsenic removal with satisfactory results in comparison to commercial materials (Mohana and Pittman, 2007). In a recent paper Troca-Torrado et al. (2011) investigated the possibility of modifying used tire rubber via chemical and thermal pre-treatment methods to adsorb phenol, p-aminophenol, p-nitrophenol, and p-chlorophenol, as well as chromium, cadmium, mercury and lead from aqueous solutions. Although such adsorptive uptake of target solutes has the potential to remove contaminants from aqueous solutions, it does not provide an ultimate treatment method, as the moieties of interest have merely been transferred to another (solid) phase.

In this paper we propose a simpler alternative for the utilization of used tires which does not require excessive pre-treatment, and which provides final and complete destruction of organic pollutants. In our approach, used tires are utilized after simple cryogenic pre-treatment and steel and fiber separation, as the sequestering phase in a Two Phase Partitioning Bioreactor (TPPB) for the removal and biodegradation of inhibitory compounds from industrial effluents. The innovative feature of the proposed system is the combination of a TPPB, which has already been demonstrated to be effective in reducing the inhibitory effect of xenobiotic pollutants on microorganisms (Daugulis et al., 2011; Tomei et al., 2011; Prpich et al., 2008), with the use of a low cost (waste) partitioning phase whose disposal is becoming an environmental concern. By way of example, scrap tire generation in industrialized countries is approximately one passenger car tire equivalent (PTE, 9 kg) per person per year. It is estimated that 2–3 billion scrap tires are stockpiled in illegal or abandoned piles throughout the U.S., and this figure represents the cumulative scrap tire build-up of approximately ten years (Reschner, 2008). For EU member states it is likely that illegal or semi-legal scrap tire piles are the same order of magnitude (Reschner, 2008). In response to the environmental and health hazards associated with this enormous numbers of scrap tires, most industrialized countries are seeking environmentally safe disposal strategies to limit the amount of tires being stored at any given location, and to encourage the use of tire derived recycling products. In the case of scrap tires the most

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0301-4797/$ – see front matter © 2013 Elsevier Ltd. All rights reserved.
http://dx.doi.org/10.1016/j.jenvman.2013.03.047

common recycling method is to grind scrap tires into crumb rubber, while removing steel, fibers and other contaminants, and to use the crumb for low-grade re-use applications.

In a previous paper (Tomei et al., 2012a) we provided a preliminary validation of the use of used rubber tire crumb in a batch TPPB system, showing positive results in improving the bioremediation kinetics in treating a chloro-phenolic compound. It was demonstrated that tires can be used as the sequestering phase, being able to trigger the process of uptake-release, driven by substrate biodegradation. In this paper we present for the first time a complete and comprehensive validation of the TPPB system, operating with tires, with tests performed in a semi-continuous bioreactor at different operating conditions by varying the applied influent load, the volumetric exchange ratio (ER) and the tire fraction. The performance of the bioreactor operated with tires was also compared to the performance of a single-phase system, and one operated with a the commercial polymer (Tone™ P787) under the same operating conditions.

2. Material and methods

2.1. Chemicals and polymers

DCP (purity > 99%) was obtained form Sigma Aldrich (Italy), and all other chemicals required for the mineral medium, which were of commercial grade, were purchased from Carlo Erba (Italy). Used tires were obtained from Recovery Technologies Canada Inc. (Cambridge Ontario), in the form of rubber "crumble". Crumble is obtained by a cryogenic process in which tires are cooled to a temperature lower than −80 °C (−112 °F). Below this glass transition temperature rubber becomes brittle, and size reduction can be accomplished by crushing and grinding, after which steel and fiber separation is easily accomplished. The crumble is claimed to have an unaltered chemical composition relative to the original tires, and is currently used for rubber modified asphalt, molded rubber products and sports surfaces. In the current application the rubber crumble was in small pieces of ~4 mm and was washed repeatedly with hot water until no visible particulates or oily droplets were seen in the washwater. In this way it was possible to remove any residual contaminants accumulated during previous use, and to prevent their release during TPPB operation.

The commercial polymer Tone™ P787 (Dow Chemical Canada Inc.) a poly-caprolactone polyester (density 1.145 g cm^{-3} and melting point 60 °C, ~4 mm diameter) was selected for comparison with tires on the basis of previous positive results which showed effective DCP absorption and release (Tomei et al., 2012b). The Tone™ polymer was also pre-washed with water to remove additives and contaminants that can remain in the polymer from the manufacturing process.

2.2. Bacterial culture

The biomass utilized in the kinetic tests originated from a bacterial culture previously cultivated on phenolic compounds and progressively acclimatized to DCP. The biomass inoculum was characterized by fluorescent in situ hybridization (FISH) according to the procedure reported in Tomei et al. (2006). The probe EUB 338, specific for the domain *Bacteria*, was utilized to evaluate the bacterial presence in the consortium. Additional probes for *Alphaproteobacteria*, *Betaproteobacteria*, *Gammaproteobacteria* were also employed for a more accurate characterization. It was demonstrated that the consortium is comprised of bacterial strains mainly consisting of *Betaproteobacteria* and *Gammaproteobacteria* and in minor extent of *Alphaproteobacteria* (~10%).

The culture was grown aerobically in mineral medium on a mixture of DCP and sodium acetate, with the DCP concentration in the feed being progressively increased, and acetate progressively decreased from 30 mg/L to 0.

2.3. Bioreactor set up and operation

DCP biodegradation tests were performed in a sequencing batch reactor (SBR) consisting of a 1 L glass vessel (0.8 L working volume) operating at 25 ± 0.5 °C by means of a thermostatically controlled water jacket. Mixing was provided by magnetic stirrers and aeration was controlled on the basis of dissolved oxygen (DO) set point values in the range of 3–4 mg/L. The phase durations of the SBR work cycle were: Feed 15 min, Reaction 600 min, Waste 2 min, Settling 75 min, Draw 28 min. In the kinetic tests the aqueous DCP concentration was measured at time intervals of ~15 min during the feed and reaction phases, until no appreciable concentration decrease was observed.

A wide range of tests was performed in duplicate in the bioreactor working in single and two-phase configuration operated with tires by varying the influent concentration, the ER in the range of 0.3–0.7 and the polymer fraction in the range of 5–9%. The tests at increased influent concentration were also performed in the two-phase system operated with Tone™. An abiotic control test was also performed to exclude the possibility of DCP losses via air stripping. All kinetic tests were performed in replicates. A summary of the test plan is reported in Table 1.

2.4. Analytical methods

Volatile Suspended Solids (VSS) measured according to Standard Methods (APHA, 1998) were used to estimate the biomass concentration. DCP was analyzed spectrophotometrically via UV absorbance at 280 nm on the supernatant of samples centrifuged for 6 min at 10,000 rpm.

3. Results and discussion

In preliminary testing (Tomei et al., 2012a) of the applicability of tires as the sequestering phase for DCP biotreatment, sorption tests determined a partition coefficient (PC) of 31 and a diffusivity coefficient of $4.8 \cdot 10^{-8}$ cm^2/s for DCP in tires. In a related experiment the commercial polymer Tone™ was found to have a PC and diffusivity coefficient of 96 and $6.6 \cdot 10^{-8}$ cm^2/s respectively (Tomei et al., 2012b). A preliminary validation of the beneficial effect of using tires in two-phase systems for DCP biodegradation confirmed that biodegradation and not just removal by absorption occurs. This was demonstrated by comparing the experimental DCP concentration data with the concentration profiles obtained by

Table 1
Range of operating conditions examined.

Parameter	Single-phase	Tires	Tone™
Feed concentration (mg/L)	100–180–250	100–180–250	100–180–250
Exchange ratio	0.5	0.5	0.5
Polymer fraction (% v/v)	0	5	5
Exchange ratio	0.3–0.5	0.5–0.6–0.7	–
Feed concentration (mg/L)	200	200	
Polymer fraction (% v/v)	5	5	
Polymer fraction (% v/v)	–	5–7–9	–
Feed concentration (mg/L)		200	
Exchange ratio		0.7	

considering only sorption as the removal mechanism (Tomei et al., 2012a). From these initial positive results this study focused on an evaluation of a wide range of operational parameters specific to sequencing batch bioreactors with the objective of completing a detailed feasibility study. A variety of operating parameters was investigated by varying the influent concentration, the volumetric exchange ratio and the percent volume of the solid phase. Three series of tests were performed each one lasting at least 90 days (180 work cycles). In all the experiments evidence of "actual" DCP biodegradation was given by the oxygen consumption data (rates in the range of 1–10 mg O_2/(h gVSS)) where the lower value in the range corresponds to the endogenous respiration rate while the higher was measured for non-inhibitory kinetics (data not shown).

3.1. Varying the influent substrate concentration

Tests were performed in the SBR reactor starting at an influent concentration of 100 mg/L, which was increased to 180 and then to 250 mg/L. At the lowest DCP concentration (data not shown) there was no significant difference between the single-phase system and the two-phase bioreactors (tires or Tone™), with virtually no inhibitory effect experienced by the conventional single-phase reactor at this substrate level. In the other two cases, presented in Figs. 1 and 2, the TPPBs operating at the higher DCP concentrations showed reduced substrate inhibition arising from the polymer addition. In Fig. 1 the test replicates are also shown as an example of the reproducibility of the data. For a feed concentration of 180 mg/L a positive effect on the biodegradation rate was observed for both Tone™ and tires, and the reduction of inhibition was much more pronounced at a feed concentration of 250 mg/L. At this substrate level the single-phase system was no longer able to provide removal of the substrate while complete removal was achieved in the two-phase systems. Although better performance was observed with Tone™, the system operated with tires was also able to efficiently achieve almost complete substrate removal, something which was not possible in the single-phase system.

An additional interesting phenomenon is also seen in these figures, as has also been observed in previous investigations (Tomei et al., 2011), namely, that a residual concentration persists in the two-phase systems with higher values remaining in the case of automobile tires. Elevated residual concentrations occurring with tires could be a consequence of the heterogeneity of this material (RMA, 2012) which consists of natural rubber (14%), synthetic rubber (27%), carbon black (28%), steel and fabric (14–15%), as well as fillers, accelerators and antiozonants (16–17%).

In tires there is a significant fraction of carbon that could act as an adsorbent for compounds such as chlorophenols. It can be hypothesized that for tires, in contrast to commercial amorphous polymers, a combined mechanism of adsorption/absorption (adsorption for carbon and absorption for rubber) or a "partially reversible sorption" may be occurring which causes enhanced retention of the solute. This was also seen in earlier sorption desorption tests with tires showing a slower desorption rate in comparison to sorption (Tomei et al., 2012a).

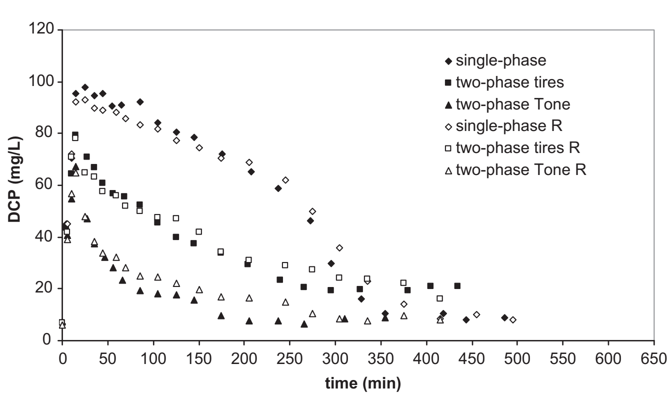

Fig. 1. SBR and TPPB SBR tests (two replicates) operated with tires and Tone™ 5%. DCP feed concentration = 180 mg/L. R = replicate.

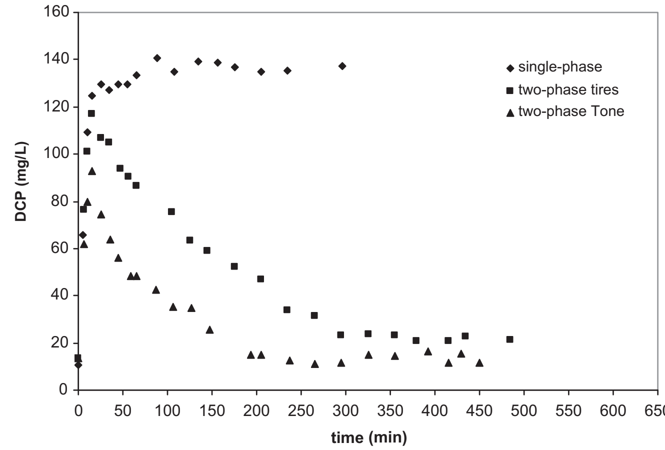

Fig. 2. SBR and TPPB SBR tests operated with tires and Tone™ 5%. DCP feed concentration = 250 mg/L.

3.2. Varying the volumetric exchange ratio

The volumetric exchange ratio, ER, is an operational parameter specific to sequencing batch reactors, and is equal to the ratio Va/V_T where Va is the volume of untreated wastewater added per cycle and V_T the total working volume of the SBR. The ER provides flexibility in system operation as it can be used to vary both hydraulic and substrate loads to the system. The upper ER limit is determined by the substrate removal rate and by the settleability characteristics of the biomass, which is retained in the residual volume of the bioreactor at the end of each work cycle to act as inoculum for the next cycle. For suspended biomass systems maximum ER values compatible with biomass settleability characteristics, are generally in the range of 0.7–0.8 and the upper value is applicable to easily settleable granular biomass systems (Wang et al., 2006).

The first series of experiments was carried out in the bioreactor operated in single-phase mode starting with an ER of 0.3, which was increased in one step to 0.5, at an influent concentration of 200 mg/L. Concentration profiles of DCP in the

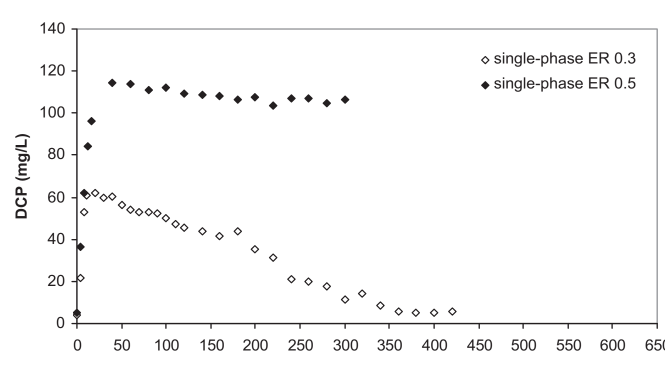

Fig. 3. Single-phase SBR operation with a DCP feed concentration of 200 mg/L.

bioreactor during the feed and reaction periods for the two operating conditions tested in single-phase operation are shown in Fig. 3. The single-phase system was able to efficiently remove the feed load at ER = 0.3, while a marked inhibitory effect by DCP is observed at an ER of 0.5, with essentially no substrate removal. The bioreactor was then switched to operate in two-phase mode by adding tire pieces at a ratio of 5% of the total working volume. A notable improvement in performance was observed with practically complete DCP removal from the liquid phase (Fig. 4). The removal process was completed in 300 min, which is half of the reaction phase duration (600 min) provided in the SBR work cycle. This suggests that at these operating conditions, the system could potentially treat additional substrate. Additional substrate loading was then applied by increasing the ER to 0.6 and 0.7, and Fig. 4 shows that the two-phase system was able to efficiently remove DCP at an influent load more than double that of the single-phase system (0.3 vs 0.7). This enhanced performance is attributed to the more favorable reaction environment arising from the presence of tires which reduces the inhibitory effect of DCP, thereby conferring a beneficial effect on cell activity. Similar behavior was also observed using commercial polymers and similar substrates (Tomei et al., 2010).

The DCP mass balance over a typical work cycle for the three ER values provides an estimate of the amount of DCP degraded in the single and two-phase SBRs. The substrate fraction retained in the solid phase was calculated from the partition coefficient, by assuming equilibrium conditions at the end of the work cycle. The results shown in Fig. 5 confirm high levels of performance by the TPPB at an ER of 0.5 (98% biodegradation), and negligible DCP retention in the tires. At higher ER values of 0.6 and 0.7 TPPB performance was still impressively high (>85% biodegradation). These findings show that with an appropriate choice of SBR operating conditions substrate release from tires can be enhanced to give not only an efficient substrate removal from the liquid phase, but also biodegradation of the xenobiotic compounds similar to that previously observed for commercial polymers as reported in Tomei et al. (2012b).

3.3. Varying the tire ratio

In these experiments the tire fraction was increased from 5 to 7 and 9% at an ER = 0.7 and an influent DCP concentration of 200 mg/L (Fig. 6). Increasing the tire ratio had a clear positive effect on the reaction time: assuming that reaction time corresponds to 95% DCP removal, a reduction of 37% and 62% for tire fractions of 7 and 9%, respectively, was seen in comparison to the 5% fraction. These results demonstrate the extra flexibility of TPPB systems, namely, that an additional degree of freedom is available (varying the tire ratio) relative to single-phase systems to enhance biotreatment performance. Such additional flexibility could be exploited, for example, by varying the amount of polymer in the TPPB in anticipation of changed substrate loadings, or by merely ensuring that adequate sequestering phase is present to cover expected substrate surge dynamics (Hagesteijn and Daugulis, 2012). Furthermore, the significant reduction of the reaction time obtained by increasing the solid fraction could also result in reducing the number of work cycles per day.

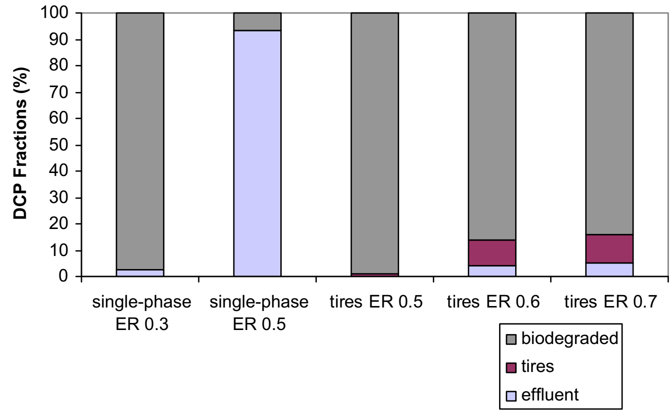

Fig. 5. DCP mass balance for single and two-phase systems operated at different ER values.

3.4. Comparison with other biological systems

A comparison of our proposed technological approach with other biological systems is reported in Table 2, and the data show a strong dependence of the process rates and efficiencies on the influent substrate concentration levels. In previous studies higher removal efficiencies were obtained only for influent concentration of about 100 mg/L, and a conspicuous decrease was observed for higher concentration values. TPPB bioreactors operated with commercial polymers were shown to be effective at DCP concentrations up to 320 mg/L where removal efficiencies ≥90% were still achieved. It is worth noting, however, the excellent performance of the TPPB operated with tires as the partitioning phase at influent concentration of 200 mg/L; almost complete DCP removal is achieved with specific removal rates in the upper range of values reported for much more complex systems.

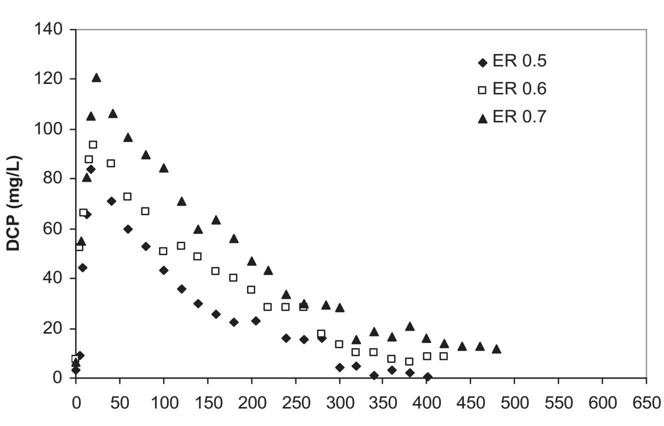

Fig. 4. TPPB SBR operation with tires (5%) and DCP feed concentration of 200 mg/L.

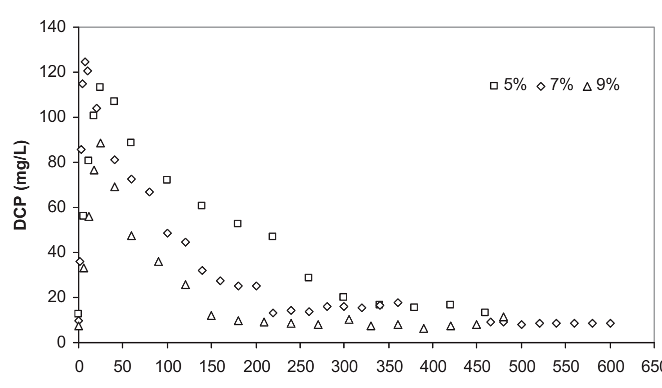

Fig. 6. TPPB SBR tests performed at different tire volumetric ratios. DCP feed concentration = 200 mg/L; ER = 0.7.

Table 2
Comparison of DCP removal efficiencies and removal rates obtained in different biological systems.

System	Removal rate	Removal efficiency (%)	Influent concentration (mg/L)	Reference
Pre-ozonation and semi-continuous bioreactor	–	70	100	Contreras et al., 2003
Batch-pure culture of *Pseudomonas putida*	–	≈100	30	Kargi and Eker, 2004
		22	300	
Air lift honeycombe-like ceramic reactor				Quan et al., 2003
Batch	0.59–3.13 (mg/L h)	≈100	17–50.8	
Continuous	–	93–99	15–102	
Acclimated mixed cultures	8.84 (mg/gVSS h)	≈100	75	Sahinkaya and Dilek, 2007
	7.11 (mg/gVSS h)	≈100	100	
	–	21	108.8	
	39.15 (mg/gVSS h)	≈100	75	
	33.18 (mg/gVSS h)	≈100	100	
Rotating biological contactors	–	92.2	120	Swaminathan and Ramanujam, 1999
		99.5	253	
Activated sludge bioreactor added of surfactant	–	99.7–99.8	30–100	Uysal and Turkman, 2005
		32.9	150	
SBR with granular biomass	39.6 (mg/gVSS h)	94	105	Wang et al., 2007
	25 (mg/gVSS h)	40	250	
	23 (mg/gVSS h)	33	300	
SBR-TPPB with Tone™ (5%) ER = 0.5	15.67 (mg/L h) 6.30 (mg/gVSS h)	91	320	Tomei et al., 2012b
SBR-TPPB with Tone™ (5%) ER = 0.5	28.06 (mg/L h) 10.67 (mg/gVSS h)	91.8	250	This study
SBR-TPPB with tires (5%) ER = 0.5	16.45 (mg/L h) 6.60 (mg/gVSS h)	83.4	250	This study
SBR-TPPB with tires (5%) ER = 0.5	17.77 (mg/L h) 14.81 (mg/gVSS h)	99	200	This study

4. Conclusions

This study confirmed the effectiveness of used automobile tires as the partitioning phase in TPPBs with performance comparable to that obtained with commercial polymers for the treatment of DCP. The two-phase system showed superior performance with respect to the single-phase bioreactor, being able to handle more than double the influent DCP load (ER from 0.3 to 0.7). By increasing the ER to 0.7 biodegradation efficiencies ≥85% were still observed. Performance can also be improved by increasing the tire fraction, with a reduction of the reaction times of 62% for an increased tire fraction from 5 to 9%.

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