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Treatment of low-strength municipal wastewater containing phenanthrene using activated sludge and biofilm process

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ABSTRACT

The main objective of this study was the comparison of activated sludge reactors with reactors containing biocarriers using a wastewater containing phenanthrene as a model compound simulating the presence of toxic substances. Five sequencing batch reactors were used. One contained a porous polyvinyl alcohol gel (PVA-gel) biocarrier and another had a high-density polyethylene biocarrier, while the other three reactors consisted of conventional activated sludge. The addition of phenanthrene at low concentration (15 μg/L in influent wastewater) did not adversely affect the removal efficiencies of chemical oxygen demand (COD) and ammonium (i.e. nitrification performance). However, with a higher addition of phenanthrene (150 μg/L in influent wastewater), a reduction in COD removal efficiency and an inhibitory effect on denitrification was observed. Generally, nutrient removal was poor, with the exception of denitrification in the reactor containing the PVA-gel. It seems that PVA-gel beads allow the formation of a stable anoxic zone in the protective core of the gel beads.

Keywords: Biological wastewater treatment; Biocarriers; Polyvinyl alcohol (PVA)-gel beads; Protistan; Nutrient removal

1. Introduction

Polycyclic aromatic hydrocarbons (PAHs) have been widely studied as environmental contaminants due to their frequent occurrence in nature and reported toxicity [1]. These pollutants are highly persistent in natural environments due to their low biodegradability and high hydrophobicity [2].

Phenanthrene is a low-molecular weight PAH which is commonly found in municipal and industrial wastewaters [3]. It is a USEPA priority pollutant and is isomeric with anthracene, which is listed as a "priority hazardous substance" under the European Water Framework Directive [4]. A recent study has shown

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that exposure to phenanthrene results in deterioration of the general health of some aquatic organisms followed by other disturbances including death [5].

PAHs may enter a wastewater treatment plant as components of domestic waste (e.g. cooking and heating oil), disposal of petroleum products from industrial sources (e.g. vehicle washing and maintenance facilities, and fuel stations), road runoff, and atmospheric deposition [6–9]. The effective operation of wastewater treatment plants plays an important role in minimizing the release of these organic pollutants into the environment.

The use of biofilm in unit processes is seen as a promising technology for treatment of wastewaters, including those containing toxic compounds. It has been applied to wastewater treatment for nitrogen removal [10], reduction of organic constituents [11], and abatement of specific toxic compounds such as phenol [12,13], Cr(VI) [14], 2,4 dichlorophenol [15], and phthalic acid esters [16].

Immobilization of biomass, which implies either attachment or entrapment of micro-organisms, has received increasing interest in the field of wastewater treatment because it offers several advantages over free, or suspended biomass such as: (1) combination of high-cell densities with high wastewater flow rates, thus allowing for effective treatment of high volumetric loadings [17]; (2) catalytic stability of biocatalysts, protection of cells from adverse environmental conditions, and tolerance against toxic compounds [18–20]; (3) reduction of excess sludge production [21]; and (4) coexistence of aerobic and anoxic metabolic activities within the same unit process [22].

Various operational parameters in wastewater treatment, such as the presence of biocarriers, have a direct effect on activated sludge microfauna. The moving bed biofilm processes promote the attached growth biological process [23] and, therefore, the dominance of sessile species is favored. The correlation of the ciliate sensitivity to a wide number of toxic substances may represent an indicator of the operating conditions of wastewater treatment plants and could provide a useful tool for the assessment of plant performance. Micro-organisms are entrapped on/in natural (polysaccharides such as alginate, carageenan, agar, etc.) or synthetic (polyvinyl alcohol (PVA), polyacrilamide, polyethylene (PE), etc.) polymeric supports. The ideal support medium must be non-toxic to the micro-organisms and have good mass transfer characteristics, stable chemical properties, and adequate structural strength [24].

In the present study, a series of bench-scale reactors were operated for the treatment of municipal wastewater containing phenanthrene. The main objectives of this study are (a) to evaluate the effectiveness of biological wastewater treatment in the presence of phenanthrene and (b) to identify changes in the activated sludge microfauna of the various reactors due to the presence of phenanthrene. The performance of the different reactors was evaluated with the removal efficiencies of organic compounds, nitrogen, and phosphorus. The emphasis of the present study was the comparison of the different systems and thus, no optimization of the reactor design was attempted.

2. Materials and methods

2.1. Reactors setup and operation

Five 1.5-L glass beakers were used as the bench-scale activated sludge reactors at room temperature. Continuous aeration was provided by three air pumps using one air diffuser in each reactor. Start up of the reactor was conducted by the addition of 150 mL of activated sludge drawn from the aeration tank of the wastewater treatment plant at the University of Patras (Greece) campus having a mixed liquor suspended solids (MLSS) content of 3,200 mg/L. Table 1 presents the operating characteristics for each reactor.

Activated sludge was taken from a municipal wastewater treatment plant and was not cultivated but was acclimatized to the new conditions. Acclimatization time may vary according to the newly introduced substances in the influent and their concentrations. It has been reported that in the case of low phenol concentrations, activated sludge microfauna could be acclimatized in 3–4 d after the introduction [25].

The reactors were initially filled with municipal wastewater, activated sludge, and, where appropriate, biocarriers up to 1.0 L total volume. Each reactor was operated in subsequent cycles of 4 d. At the end of each cycle, sludge sedimentation took place by turning off the air pumps (adapted from [14,26]). Three hundred mL of the supernatant water was withdrawn from each reactor for further analysis, and was replaced by municipal wastewater spiked with desirable phenanthrene concentrations. An acclimation period of 12 d (three cycles) was allowed for the activated sludge to adapt to the experimental conditions. After that period, phenanthrene was spiked to the wastewater influent (300 mL) of reactors R2–R5 to achieve concentrations of 15 or 150 μg/L, respectively, in the reactors (see Table 1). The reactors were operated for a total of 44 d. The experimental hydraulic residence time was calculated equal to 13.3 d. Suspended solids losses in the effluent were insignificant, thus, solids retention time was not calculated.

Table 1
Characteristics and content of each reactor at start up

Reactor	Activated sludge (mL)	Municipal wastewater (mL)	Biocarrier	C_{PHE} (μg/L)[a]
R1	150	850	–	–
R2	150	850	–	15
R3	150	850	–	150
R4	150	775	75 mL PVA	150
R5	150	700	150 mL K1	150

[a]C_{PHE}: Phenanthrene concentration in the reactors at the beginning of the 4th operation cycle.

R4 included 75 mL of PVA-gel beads (Kuraray Co., Ltd., Japan). The gel beads are 4-mm diameter spheres and are slightly heavier than water (specific gravity of 1.025 g/cm^3), hydrophilic in nature, and have a very porous structure with only 10% solids and a continuum of passages 10–20 μm in diameter tunneling throughout each bead [27,28]. R5 was filled with 150 mL of a moving-bed biocarrier (Model K1, Veolia Water, France), which consists of high-density PE rings and are slightly lighter than water (specific gravity of 0.95 g/cm^3). They have a cylindrical shape with 7 mm length and 9 mm diameter with a cross inside and fins on the outside. Biomass grows mainly on the protected area inside the cylinder, even though some biomass also grows between the fins on the outside. Treatment efficiency and activated sludge microfauna were evaluated by the systematic monitoring of effluent and activated sludge samples, respectively. A blank run was performed with a reactor containing 1 L of 150-μg/L phenanthrene solution and the same tubing as the other reactors. The aeration lasted for 4 d and phenanthrene concentration of the solution after aeration was compared to that of the initial solution. No significant difference was observed due to volatilization or sorption to the glass walls or the tubes.

2.2. Physicochemical analyses

Chemical oxygen demand (COD) was measured by the reaction digestion method (Method 8000, Hach). Ammonium nitrogen (NH_4^+–N) was measured by the Nessler Method (Method 8038, Hach), nitrate nitrogen (NO_3^-–N) by the Chromotropic Acid Method (Method 10020, Hach), and orthophosphates (PO_4^{3-}) by the ascorbic method (Method 8048, Hach). Inference of statistical significance of parameter means was determined by applying the univariate *z*-test.

Phenanthrene was measured in the supernatant using a cuvette mode fluorescence spectrophotometer (RF1501 spectrofluorophotometer, Shimadzu, Japan), using a quartz cuvette and excitation/emission wavelengths of 249/347. Before every sample measurement, calibration curves were constructed using phenanthrene standards prepared with wastewater effluent from reactor R1 as the solvent and not with pure DI water. This was done to account for background effects to measurements due to organic compounds found in the wastewater.

Activated sludge solids were air dried and total organic carbon was determined using the wet oxidation method with the indirect calculation of OC content through the volumetric determination of the oxidant excess in the final solution. Briefly, 0.7–1 g of dry sludge was thoroughly mixed with 5 mL of H_2SO_4/Ag_2SO_4, 10 mL of potassium dichromate ($K_2Cr_2O_7$, 0.167 M), and concentrated H_2SO_4 (20 mL). The mixture was shaken for 30 s and allowed to stand for 30 min. The sample was diluted to 200 mL with distilled water. Finally, samples were cooled and titrated with ferrous ammonium sulfate $Fe_2(NH_4)_2(SO_4)_3$ 0.5 N.

2.3. Microfauna identification and enumeration

For the analysis of protozoan community, aliquots of 200 μL were received from all bench-scale reactors. Analysis was conducted for the identification of species *in vivo* using an optical microscope (Leica DMLB, Leica, Germany) with 10×/20 magnification on the eyepiece lens and with 10×/0.25, 40×/0.75, and 100×/1.25 magnifications on the objective lens. Identification of protozoa was mainly based on their morphology and movement by comparison to morphological descriptions in Standard Methods [29].

The protistan community directly reflects any changes in bacteria community since the majority of protozoa are bacterivorous [30]. Monitoring of the protistan community is time and cost efficient and in full-scale plants is more applicable. Future research should further investigate changes in bacteria.

3. Results and discussion

The characteristics of the municipal wastewater used as influent for this study are shown in Table 2. After mixing, pH levels in the reactors were from 8.0 to 8.5.

3.1. Activated sludge reactors

The effect of phenanthrene on the activated sludge microfauna, and, consequently, on the effectiveness of the treatment process, was evaluated in reactors R1, R2, and R3 operating under activated sludge processes. For the entire operation period, the reactors demonstrated good removal efficiencies of organic matter with effluent COD levels well below the secondary effluent discharge limit of 125 mg/L (Fig. 1(a) and (b)). However, it should be noted that the prolonged addition of phenanthrene decreased COD removal efficiency after the 25th day of operation, indicating higher COD effluent values.

Following the introduction of phenanthrene, comparisons were made of the mean values of COD concentrations in the effluents of the reactors over the last three weeks of operation. From these results we can say (with 90% confidence) that the addition of phenanthrene at 15 μg/L (in R2) did not affect treatment performance; however, at 150 μg/L (in R3), the phenanthrene addition did result in a significant inhibition. Thus, further testing with the phenanthrene at 150 μg/L would allow for the assessment of any potential benefit associated with use of biocarrier-based system (below).

In reactors R3, R4, and R5, removal of organic load in terms of COD reduction was achieved effectively up to legislation limits at about the 5th day of operation. Similar pattern of removal was observed for the case of NH_4^+–N concentrations, where nitrification was more effective after the 5th day of operation.

As shown in Fig. 2(a), no significant effect in NH_4^+–N concentrations was observed by the addition of phenanthrene in any of the reactors. The nitrification process was very efficient with NH_4^+–N levels decreasing from 24 mg/L (Day 0) to 0.02 mg/L (Day 4) during the first four days. The NH_4^+–N removal efficiencies were similar between the reactors and in all cases were greater than 98%. Moreover, NH_4^+–N levels in treated effluents were much lower than the regulatory discharge limit of 2 mg/L. This is an initial, although weak, evidence that the nitrification did not appear to be affected by the addition of phenanthrene. For a stronger conclusion, a data analysis taking into account bio-kinetic equations of nitrification and denitrification is required.

In all three reactors, the levels of NO_3^-–N increased until the 15th day of operation, leveling off at approximately the concentration of total nitrogen in the influent (Fig. 2(b) and (c)) due to the nitrification response noted above. Though nitrification of the applied NH_4^+–N had been nearly completed by the 5th day (Fig. 2(a)), the level of total nitrogen in the reactors (Fig. 2(c)) dropped during the first 5 d and then increased to approximately the original applied level in the following 5 to 10 d. The temporary apparent loss of NH_4^+–N and NO_3^-–N during the initial 10 d of the study would most certainly have been due to the appearance of transitory nitrite (NO_2^-–N), which was not quantified in this study. Following this initial period, with nearly complete nitrification in all reactors, nitrogen levels consisted mostly of NO_3^-–N (Fig. 2(c)).

Following the addition of phenanthrene to R2 and R3 on Day 15, a gradually decreasing trend in effluent NO_3^-–N levels was observed, which became more pronounced for R1 and R2, containing the addition of no or very little (15 μg/L in the influent) phenanthrene, respectively. Over the last 20 d of operation, NO_3^-–N levels in R1 and R2 were reduced to approximately 15 mg/L, indicating effectiveness of the nitrification process reaching up to 50% of the total influent NH_4^+–N concentration. Conversely, for R3, with a phenanthrene addition of 150 μg/L in the influent, the level of NO_3^-–N was reduced by only approximately 5 mg/L over the same period. However, the observed losses of NO_3^-–N for all three reactors were generally poor in these reactors probably due to the lack of anoxic zones or anoxic operation periods [31]. The denitrification process is generally ineffective when dissolved-oxygen levels exceed 1.0 mg/L [32]. Generally, the addition of phenanthrene seems to decrease the denitrification in R2 and R3 compared to R1. Furthermore, given that all three reactors operate under the same aeration conditions, it is considered that the extremely weak denitrification response in R3 (Fig. 2(b)) may be attributed to the inability of denitrifying bacteria to

Table 2
Physicochemical characteristics of raw wastewater used as influent

Characteristic	Raw wastewater
Chemical oxygen demand (COD)	250 mg/L
NH_4^+–N	24 mg/L
NO_3^-–N	5.0 mg/L
NH_4^+–N and NO_3^-–N	29 mg/L
PO_4^{3-}	13 mg/L
pH	8.1

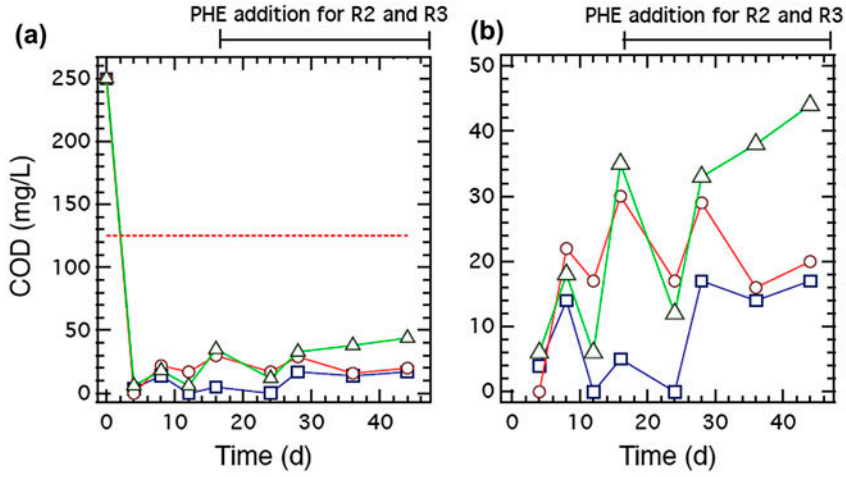

Fig. 1. COD concentrations in R1 (square), R2 (circle), and R3 (triangle) as a function of operation time. Dotted line in Fig. 1(a) is the disposal limit. (a) Influent concentration at $t = 0$ days and effluent concentrations, (b) effluent concentrations.

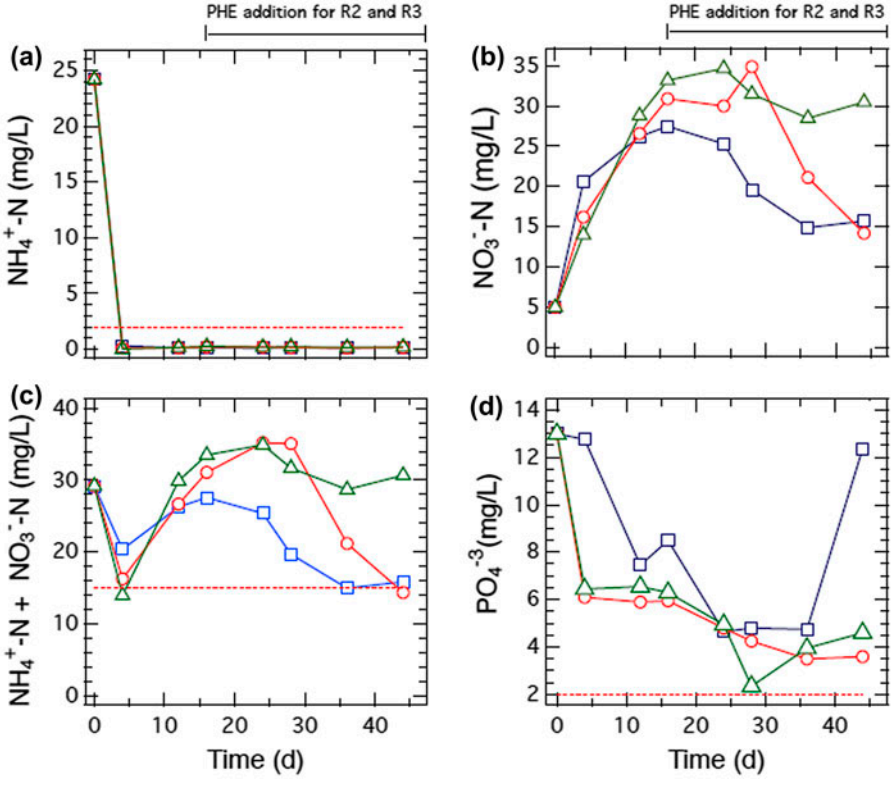

Fig. 2. (a) Ammonium–Nitrogen (NH_4^+–N), (b) Nitrate–Nitrogen (NO_3^-–N), (c) NH_4^+–N and NO_3^-–N, and (d) Orthophosphate (PO_4^{3-}) concentrations in R1 (square), R2 (circle), and R3 (triangle) as a function of operation time. Dotted lines are the respective disposal limits.

metabolize the NO_3^-–N under the toxic effects of phenanthrene in the influent (150 μg/L).

Effluent phosphates concentration as a function of operation time as shown in Fig. 2(d) was generally decreasing due to uptake of this essential nutrient for cell growth. Over the final 10 d of operation, R2 and R3 had almost the same levels of PO_4^{3-} (ca. 4 mg/L) in the effluents. However, the results of R1, which did

not contain phenanthrene, did not follow the same trend and reached the initial concentrations (12 mg/L) on the 44th day of operation. Thus, it appears the removal of phosphates was enhanced by the addition of phenanthrene. This can either be attributed to the increased metabolic needs of the activated sludge micro-organisms to produce ATP in order to cope with the toxic stress, or to the increased production of extracellular polymeric substances that protect the cells from phenanthrene. In all reactors, the effluent phosphates did not comply with the discharge limits.

3.2. Activated sludge reactors supplied with biocarriers

To compare the effectiveness of biocarrier-based processes with traditional activated sludge, biological treatment reactors utilizing activated sludge (R3), PVA-gel beads (R4), and K1 (R5) were evaluated. For all three reactors, the same influent wastewater containing phenanthrene was used.

All reactors demonstrated good removal efficiencies of organic matter with COD levels of the effluents reduced by over 80% within the first four days of operation (Fig. 3(a) and (b)). Generally, the time courses of COD effluent values were similar for all three reactors with the exception that R4, containing the PVA-gel biocarrier, demonstrated higher removal efficiencies.

Following the introduction of phenanthrene, comparisons were made of the mean values of COD concentrations in the effluents of the reactors over the last three weeks of operation (days 24–44). From these results we can say with 72% confidence that the use of the PVA-gel biocarrier (R4) does appear to protect the biomass from inhibition associated with the addition of phenanthrene (at 150 μg/L); however, even with 50% confidence, no such inference can be made by the use of the K1 biocarrier (R5) (i.e. results of R3 and R5 were not significantly different).

As shown in Fig. 4(a), no significant differences in ammonium concentrations were observed for the three reactors. The nitrification process was very efficient with NH_4^+–N levels decreasing from 24 to 0.02 mg/L during the first four days and remaining well below the discharge limit of 2 mg/L for the duration of testing in all cases.

The levels of nitrate (Fig. 4(b)) trended upward until the 12th day of operation in all reactors. However, from that point, denitrification appeared to occur in R4, under the presence of PVA-gel biocarrier, demonstrating a pronounced reduction in NO_3^-–N levels, which decreased significantly until leveling off at approximately 5 mg/L by the 36th day of operation. This occurrence of denitrification only in the reactor with the PVA-gel biocarrier would indicate that the porous PVA-gel medium provides a superior niche for formation of a stable anoxic zone in the protective core of the gel beads, which has been suggested by others [33,34].

Due to nearly complete nitrification responses in all reactors, nitrogen levels (Fig. 4(c)) followed closely those of NO_3^-–N (Fig. 4(b)). In addition, only R4 containing the PVA-gel biocarrier achieved reduction of nitrogen to levels well below the 15 mg/L discharge limit, apparently due to the benefit of denitrification as discussed above. Finally, as shown in Fig. 4(d),

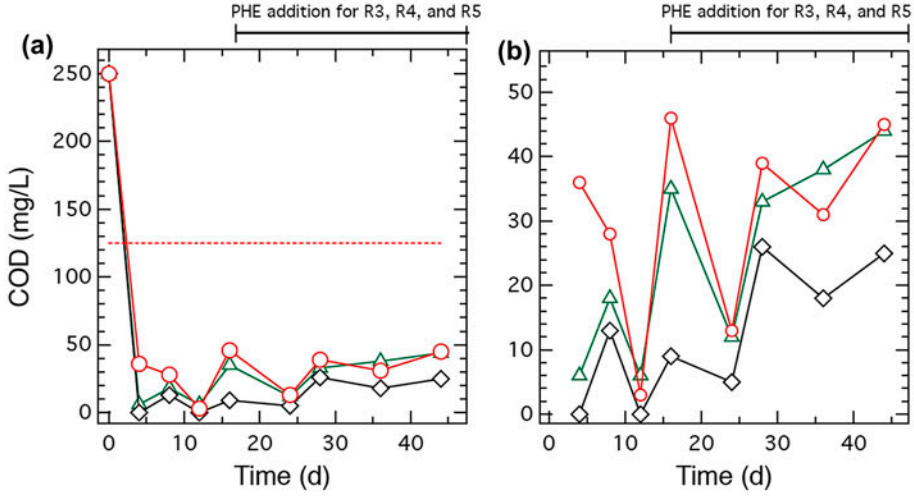

Fig. 3. COD concentrations in R3 (triangle), R4 (diamond), and R5 (circle) as a function of operation time. Dotted line in Fig. 3(a) is the disposal limit. (a) Influent concentration at t = 0 days and effluent concentrations, (b) effluent concentrations.

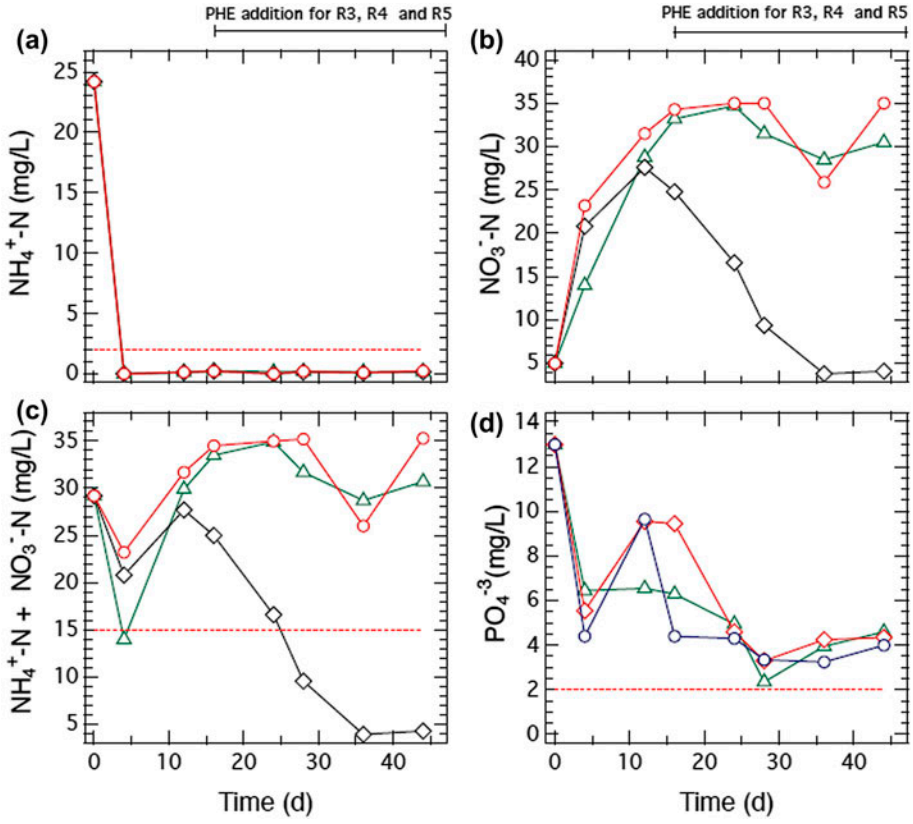

Fig. 4. (a) Ammonium–Nitrogen (NH_4^+–N), (b) Nitrate–Nitrogen (NO_3^-–N), (c) NH_4^+–N and NO_3^-–N, and (d) Orthophosphate (PO_4^{3-}) concentrations in R3 (triangle), R4 (diamond), and R5(circle) as a function of operation time. Dotted lines are the respective disposal limits.

there were no significant differences in the phosphate removal efficiencies between the different reactors, suggesting that the use of biocarriers offers no benefit with respect to this parameter. Typical reactor values such as sludge retention time and sludge age were not calculated since the research was oriented to the monitoring of activated sludge microfauna.

3.3. The fate of phenanthrene

Phenanthrene concentrations were below the detection limit by the end of the first 4-d cycle for R2, which had the lowest (15 μg/L) applied loading. The removal efficiencies for the other reactors, though, with the higher loading in the influent, increased over the first 28 d of operation as shown in Fig. 5. This response may have reflected the need for the biomass to acclimate to this relatively difficult to degrade hydrocarbon. Overall, all reactors had similar removal efficiencies leveling off at approximately 50% by the final two weeks of the testing period. The removal efficiencies for the two biocarrier systems were very similar, achieving efficiencies approximately 10% over that of the suspended growth system by the end of the study. Further testing would be needed to see if further improvements could be achieved over long-term operation, where the retention of substrate-specific micro-organisms by biocarrier-based methods would potentially be a benefit, as observed by others [33].

Activated sludge solids were measured to contain a 25 ± 5% organic carbon. The MLSS concentration of the reactors was 190 ± 22 mg/L. If one calculates the sorption coefficient based on the organic carbon content, phenanthrene organic carbon distribution coefficient (K_{oc}), and the solid-to-liquid ratio and if the activated sludge solids are considered passive organic carbon solids, they could uptake at equilibrium from 28 to 37% of the initial phenanthrene added. However, it is not known how much time is needed to reach equilibrium. A loading rate study to see what would happen at various biomass and substrate (both the COD and PHE) concentrations would be interesting for a future study.

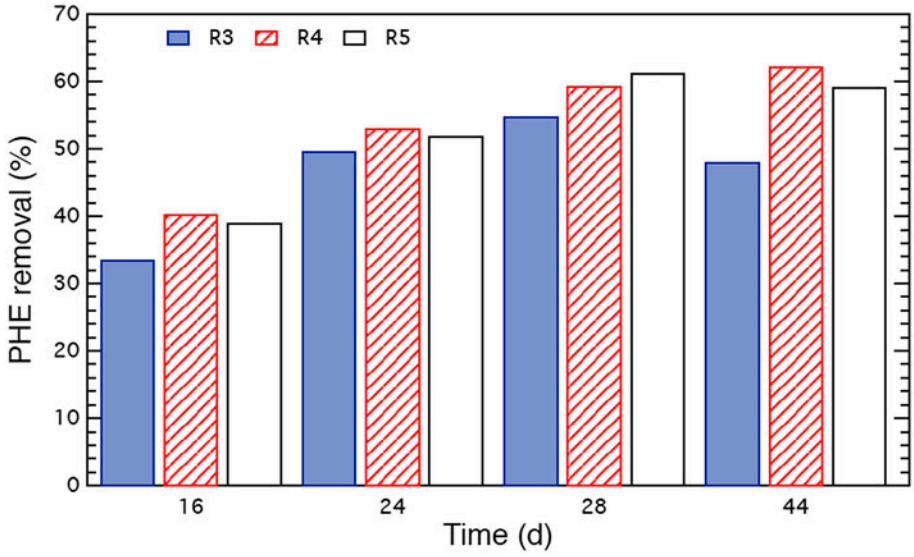

Fig. 5. Phenanthrene removal as a function of operation time. Phenanthrene was introduced to all three systems at a concentration of 150 μg/L every 4 d starting on the 12th day.

A recent study on the uptake of phenanthrene by PVA and PE suggests that these polymers uptake only 10% of the initial phenanthrene added for a contact time of 4 d and 70–80% at equilibrium requiring a contact time from 30 to 100 d [35]. These numbers change for different solid-to-liquid ratios and for different polymer widths and are valid only for the first addition of phenanthrene. Then, the uptake rate is expected to be lower since sorption would be closer to equilibrium or saturation.

Based on these findings, sorption onto the activated sludge solids and onto the biocarriers could be important during the first 4-d cycle after the addition of phenanthrene. However, there is still a small percentage that can be attributed to degradation. In addition, if phenanthrene was not being degraded by the biomass, then eventually sorption would equilibrate and the concentration would start rising, but there is no indication of that. The long-term accumulation of phenanthrene into the organisms at high concentrations could lead to population degradation, but more advance techniques are required to be able to distinguish the percentage accumulated from that that is degraded.

3.4. Changes in the activated sludge microfauna

In order to investigate possible effects of phenanthrene on the activated sludge protistan microfauna, the protistan species in the activated sludge of each reactor were identified in the beginning and at the end of the operation period (Table 3). Reactor R1 served as a control reactor in order to identify the changes that occurred under the presence of biocarriers and/or phenanthrene.

In general, low activated sludge microfauna diversity was observed in the initial activated sludge

Table 3
Identified species in the sludge at the beginning (inoculum) and end of the operation period in each reactor (R1–R5)

Micro-organism species	Inoculum	R1	R2	R3	R4	R5
Protozoa						
Vorticella sp.	√	√	√		√	√
Chilodonella sp.	√	√				
Euplotes sp.	√	√	√			
Litonotus sp.	√		√		√	
Pelomyxa sp.						√
Paramecium sp.	√	√				
Colpoda sp.			√	√	√	
Euglena sp.						√
Rotifers						
Euchlanis sp.	√					
Philodina sp.	√	√	√	√	√	√
Lecane sp.	√					
Keratella sp.					√	
Nematodes						
Nematodes	√				√	√

samples compared to other studies [36,37], a fact that might be attributed to the low concentrations of organic load and nutrients present in the municipal influent. However, it should be stressed out that all the species present are indicative of "well dominated microfauna with good biodegradation ability" [38]. Moreover, the presence of at least three trophic levels identified, bacteria (essential to all activated sludge processes), protozoa, and metazoa (Rotifers), indicates the stability of the activated sludge "ecosystem" [25]. Low diversity of species was observed in all reactors, as in the inoculum. In most reactors, protistan species were not affected neither by the addition of biocarriers nor by the addition of phenanthrene, while metazoa diversity decreased from 4 to 1 representative species. However, reactor R3 presented significant loss, operating as a single activated sludge process and under 150 μg/L phenanthrene, of protistan species, resulting in single species protistan community of *Euglena* sp. Thus, these two factors affected the biological activity and growth of micro-organisms. Moreover, it is worth mentioning that in the reactors containing biocarriers, a larger number of species were identified. This fact could be explained by the growth of the micro-organisms on or in biocarrier matrices and the formation of biofilms, which makes them more tolerant toward harsh conditions and provides an amenable environment for their growth.

Succession on the biocarriers creates dense and compacted core zones, which are usually composed by bacteria and are bound by the extracellular polymer substances (EPS) excreted by them, while the outer fringe zones are composed mainly by stalked ciliates. Previous studies have reported changes of the biofilm structure during its succession [30,39]. The succession process of a biofilm can be divided into four distinct phases:

(1) dominance of crawling ciliates on the sludge flocs,
(2) proliferation of stalked ciliates, serving as platform for the attachment of bacteria,
(3) complete dominance of the stalked ciliates, and
(4) overgrowth of the stalked ciliates, having as a result their subsequent death or escape from the biofilm and the prevalence of the free-swimming ciliates.

Increased tolerance of the microfauna presented in the reactors operating with biocarriers may also be attributed to the increased EPS production that enables the protection of the cells toward toxic substances [40].

4. Conclusions

The main conclusions of the present study are as follows:

(1) COD levels and NH_4^+–N concentrations changed uniformly with no significant differences among the three reactors with activated sludge (R1, R2, and R3) and varying concentrations of phenanthrene.
(2) Only the reactor containing the PVA-gel beads achieved a reduction of total nitrogen to levels well below the discharge limit due to a superior denitrification performance.
(3) No significant differences in the phosphate removal efficiencies between the different reactors were observed over the period of testing, suggesting that the use of biocarriers has no effect on this parameter.
(4) A relatively larger number of species were identified in the reactors containing biocarriers versus the reactor with the activated sludge, when higher phenanthrene concentration was applied.

Thus, the biofilm reactors had similar treatment performance as that of the activated sludge reactor, with the exception that superior denitrification was demonstrated with the PVA-gel biocarrier, evidently due to the porous matrix of the gel beads providing a niche for the formation of a stable anoxic zone. In addition, when higher phenanthrene concentration was applied, a higher degree of biodiversity was observed with the two biofilm reactors than that of the activated sludge reactor.

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