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Performance of a hybrid-loop bioreactor system in biological treatment of 2,4,6-tri-chlorophenol containing synthetic wastewater: Effects of hydraulic residence time

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Abstract

A hybrid-loop bioreactor system consisting of a packed column biofilm and an aerated tank bioreactor with effluent recycle was used for biological treatment of 2,4,6-tri-chlorophenol (TCP) containing synthetic wastewater. Effects of hydraulic residence time (HRT) on COD, TCP and toxicity removal performance of the reactor were investigated for the HRT values between 5 and 30 h, while the feed COD ($2700 \pm 100 \, \mathrm{mg \, l^{-1}}$), TCP ($300 \pm 10 \, \mathrm{mg \, l^{-1}}$) and the solids retention time (sludge age, SRT, 20 d) were constant. Percent TCP, COD and toxicity removals increased with increasing HRT resulting in more than 90% COD, TCP and toxicity removals at HRT values above 25 h. Biomass concentrations in the packed column and in the aeration tank increased with increasing HRT resulting in low reactor TCP concentrations and therefore high TCP, COD and toxicity removals at high HRT values. Volumetric and specific rates of TCP and COD removals decreased with increasing HRT due to increased biomass and decreased flow rates at high HRT levels. Volumetric and specific removal rates of COD and TCP were maximum at an HRT of 5 h.
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Keywords: Biological treatment; Hybrid loop-bioreactor; 2,4,6-tri-chlorophenol (TCP); Toxicity removal

1. Introduction

Due to toxic effects of chlorophenol compounds present in some chemical industry wastewaters such as petrochemicals, pesticides, plastics, pulp and paper, performance of conventional biological treatment systems treating such wastewaters is usually low yielding highly toxic effluents [1–4]. Therefore, effective treatment methods should be developed to reduce the concentrations of chlorophenols to non-toxic levels before discharging such effluents to receiving media. Effluent toxicity should also be considered as a discharge parameter in evaluation of such effluents.

One of the most toxic chlorophenol compounds is 2,4,6-tri-chlorophenol (TCP) because of three chloride groups attached to the phenol ring. Some physico-chemical methods may be used such as activated carbon adsorption or chemical oxidation by ozon for the removal of TCP from wastewaters [1–3]. Adsorption is not an ultimate method for mineralization of such compounds

and also is expensive which can only be used to concentrate TCP on activated carbon for further chemical or biological mineralization. Chemical oxidation methods may result in formation of toxic products and may also be very costly. Adsorption and chemical oxidations are usually used in tertiary treatment after biological treatment where the concentrations of chlorophenols are low enough to justify the cost of adsorption or chemical oxidation. Therefore, anaerobic and aerobic biological treatment methods are the most cost effective methods for complete mineralization of chlorophenols [2,3]. Biological treatment of chlorophenol containing wastewaters is relatively inexpensive and usually results in complete mineralization [2–5]. A number of studies were reported in literature concerning biodegradation of chlorophenols by pure suspended cultures of bacteria and fungi [6–15]. In most of the literature studies, chlorophenols were used as co-substrate at low concentrations in the presence of a major carbohydrate substrate [9,10,14]. Some adaptation procedures were used to promote the capabilities of different cultures to improve the rate and the extent of biodegradation of chlorophenols [5,16]. Utilization of immobilized cells or biofilm reactors was reported to improve the removal efficiency of chlorophenols due to high biomass concentrations and con-

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centration gradients of chlorophenols in the bioreactor and in the biofilm [17–24]. However, due to heterogenous nature of the biofilm reactors, it is difficult to control some parameters such as the biofilm thickness, dissolved oxygen concentration, pH, and redox potential. Well mixed suspended culture reactors are easier to control and may be effective with partial cell recycle. Utilization of hybrid reactors consisting of suspended and biofilm cultures in the biological treatment system with a fast effluent recycle between the reactors may be an effective approach for treatment of chlorophenol containing wastewaters since each reactor would compensate for the other's disadvantages.

Biodegradability of chlorophenols varies depending on the position and the number of chlorine groups and usually biodegradability decreases and as a result toxicity increases with increasing number of chlorine groups [4]. Toxicity of individual chemicals or complex effluents can be determined by using different biological tests [25]. Recently developed 'Resazurin Assay' is relatively simple, inexpensive and rapid method for assessment of the toxicity of chemicals and toxic effluents [26–28]. The resazurin assay is based on measurement of percent inhibition on dehydrogenase activity of bacteria in the presence of toxic compounds. Toxicity values obtained with the resazurin assay are comparable to those obtained with the more commonly used biological methods such as *Daphnia magna*, and Microtox TM [25].

2,4,6 TCP is one of the most refractory chlorophenol compounds with an IC_{50} value of nearly $45\,mg\,l^{-1}$ for the test organisms. Limited number of studies was reported in literature on biodegradation of TCP [2,3,30–33]. Most of those studies were performed with either pure cultures or utilized combined anaerobic–aerobic treatment and also used low TCP concentrations below $100\,mg\,l^{-1}$. Some industrial effluents from pesticide, plastics and petrochemical industries may contain chlorophenols above $100\,mg\,l^{-1}$ in certain waste streams. Biodegradation of TCP is different from those for mono and dichlorophenols [33]. The *para*-Cl group is replaced by another OH-group forming 2,6 dichlorohydroquinone in biodegradation of 2,4,6 TCP [11,33]. There are no literature reports on aerobic biological treatment of TCP containing wastewater at high TCP concentrations such as $300\,mg\,l^{-1}$ at different hydraulic residence times. Therefore, the major objective of this study is to investigate the biological treatment performance of a hybrid (packed column and suspended culture) loop-bioreactor system for biological treatment of synthetic wastewater containing high TCP concentration ($300\,mg\,l^{-1}$) for a large range of HRT (5–30 h). Effects of HRT on TCP, COD and toxicity removal performance of the system were investigated at constant feed COD, TCP and SRT of $2700 \pm 100\,mg\,l^{-1}$, $300 \pm 10\,mg\,l^{-1}$ and 20 days, respectively.

2. Materials and methods

2.1. Experimental set-up

A schematic of the experimental set-up is shown in Fig. 1. The hybrid-loop bioreactor system consisted of a plexi-glass
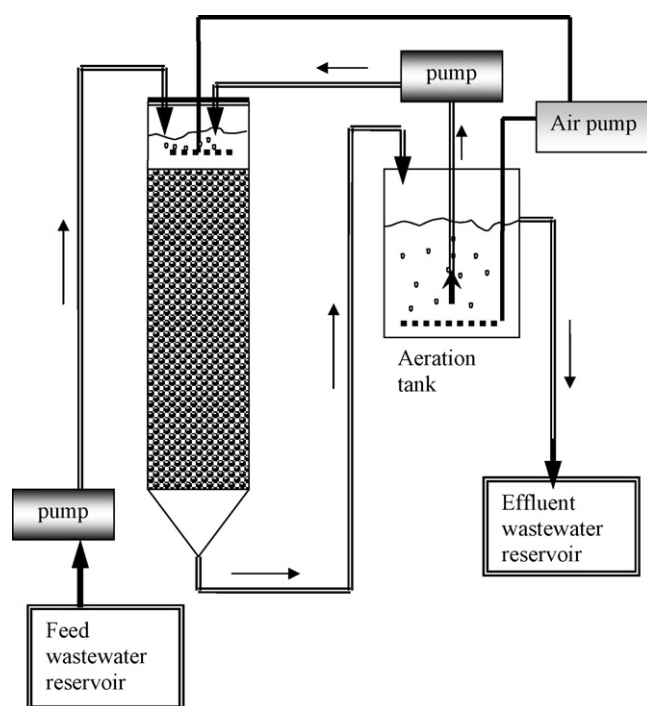

Fig. 1. A schematic diagram of the experimental set-up.

packed-column biofilm reactor and a plexi-glass aeration tank. The packed-column biofilm reactor had dimensions of 7.1 cm diameter and 40.5 cm height with a total empty volume of 1.25 l in the packed section. The column was filled with 1300 olive pits as support material for biofilm formation. Void fraction in the column was 0.46 with a total solids volume of 0.675 l and liquid volume of 0.575 l. Total biofilm surface area in the column was $0.6175\,m^2$ yielding specific surface area of $494\,m^2\,m^{-3}$ empty column or $1074\,m^2\,m^{-3}$ liquid in the packed column. The column contained 100 ml wastewater on top, which was aerated using a diffuser and air pump and also 75 ml wastewater was present in the conical section at the bottom of the packed section. A perforated plate was used at the bottom of the column to separate the support particles from the circulating liquid. The cylindrical aeration tank contained 1.5 l mixed liquor and was aerated with an aeration rate of nearly 5 vvm (vol air/vol water min) to yield a dissolved oxygen concentration (DO) of above $2\,mg\,l^{-1}$. Feed wastewater was kept in a deep refrigerator at $4\,°C$ to avoid any decomposition and was fed to the packed column with a desired flow rate to adjust the HRT to 5–30 h for the whole system volume of 2.3 l. The aeration tank was fed with the recirculation effluent and the wastewater in the aeration tank was re-circulated to the column reactor as shown in Fig. 1 using a peristaltic pump with a flow rate of $2.4\,l\,h^{-1}$ yielding recirculation ratio (R = recycle flow rate/ feed flow rate) between 5.2 and 31.3. The effluent from the aeration tank was collected in a reservoir. The system was operated as a closed-loop hybrid bioreactor system containing a packed column biofilm reactor and an aeration tank. Temperature, pH and dissolved oxygen (DO) concentrations in the aeration tank were measured twice a day and were adjusted to $T = 25 \pm 2\,°C$, pH $7. \pm 0.2$ and DO $= 2 \pm 0.5\,mg\,l^{-1}$, respectively.

2.2. Wastewater composition

Synthetic feed wastewater used throughout the study was composed of diluted molasses, urea, KH_2PO_4 and $MgSO_4$ resulting in COD/N/P = 100/8/1.5. Typical composition of the feed wastewater was $COD_0 = 2700 \pm 100\,mg\,l^{-1}$ including COD content of TCP, N_T (total nitrogen) = $215 \pm 10\,mg\,l^{-1}$, PO_4-P = $40 \pm 2\,mg\,l^{-1}$, $MgSO_4 = 50\,mg\,l^{-1}$ and 2,4,6 $TCP_0 = 300 \pm 10\,mg\,l^{-1}$ with a COD content of $340\,mg\,l^{-1}$ (COD/TCP = 1.134). pH of the feed wastewater was nearly 6.9 which increased to nearly pH 7.5–8 in the reactors due to ammonia released by biodegradation of urea. TCP was dissolved in hot water at $50\,^{\circ}C$ before adding to the synthetic wastewater.

2.3. Organisms

The activated sludge culture obtained from PAK MAYA Bakers Yeast Company wastewater treatment plant in Izmir, Turkey was used as the seed culture. The activated sludge culture was grown in an aeration tank using the same synthetic wastewater in the presence of $50\,mg\,l^{-1}$ TCP for several days and was used for inoculation of the experimental system.

2.4. Experimental procedure

Experiments were started batchwise by placing about 1 l synthetic wastewater containing $50\,mg\,l^{-1}$ TCP in aeration tank, which was inoculated with 0.5 l of the seed activated sludge culture. The system was operated in batch mode with effluent recycle between the reactors for 10 days to obtain a dense culture of the activated sludge before starting the continuous operation. Feed wastewater was fed to the reactor with a desired flow rate (1.8 to $11\,l\,d^{-1}$) using a peristaltic pump to obtain HRT values between 5 and 30 h and was removed with the same flow rate. Temperature and pH were approximately $25 \pm 2\,^{\circ}C$ and $7. \pm 0.2$, respectively in the system. Dilute sulfuric acid was added to the aeration tank several times a day to adjust the pH around 7. Dissolved oxygen (DO) concentration was above $3\,mg\,l^{-1}$ in the aeration tank and more than $1.5\,mg\,l^{-1}$ in the effluent of the packed column reactor indicating no DO limitations in the system. Solids retention time (sludge age, SRT) was kept constant at 20 days by removing 5% of the sludge (115 ml) from the system everyday one-half of which was from the aeration tank and one-half from the packed column including the peats. Every experiment was conducted until the system reached the steady-state yielding the same COD and DCP contents in the effluent for the last 3 days. The average time elapsed for each experiment was nearly 15–20 days. The samples collected from the feed reservoir and effluents of packed column and the aeration tank at the steady-state were analyzed for COD, TCP and toxicity after centrifugation.

2.5. Analytical methods

Samples were withdrawn everyday from the feed reservoir, column effluent and aeration tank and centrifuged at 8000 rpm

(7000 g) for 20 min to remove biomass from the liquid phase before analysis. Clear supernatants were analyzed for COD and TCP contents. 4-Aminoantipyrene colorimetric method developed for determination of phenol and derivatives in form of phenol index was used for TCP analysis as specified in the standard methods [29]. Some samples were also analysed by using an Agilent 1100 model HPLC (Agilent Technologies, USA) to determine potential end products of TCP biodegradation. The HPLC was equipped with a ZORBAX Eclipse XDB-C18 column ($4.6 \times 150\,mm \times 5\,\mu m$). The mobile phase was methanol: water (65:35) with a flow rate of 1 ml/min. Chemical oxygen demand (COD) was determined using the closed reflux method according to the standard methods [29]. Biomass concentrations in the aeration tank were determined by filtering the samples (10 ml) through $0.45\,\mu m$ milipore filter and drying in an oven at $105\,^{\circ}C$ until constant weight. The biomass concentrations in the packed column was measured by removing samples from the column including the olive peats and the mixed liquor. The organisms on the peat surfaces were washed and the total biomass was determined after filtering through a $45\,\mu m$ milipore filter and drying until constant weight. The COD and TCP samples were analyzed in triplicates with less than 5% standard deviations from the average.

Resazurin reduction method was used to determine the toxicity of the feed and effluent wastewater from each reactor [26–28]. The test organisms (activated sludge) were cultivated on nutrient broth and were transferred to a new medium everyday to keep the sludge age constant. Five milliliters of the test culture was centrifuged and re-suspended in distilled water resulting in an optical density of about 0.4 at 625 nm. The centrifuged cells were used in toxicity determinations in the test tubes after discarding the supernatant. Reagent control (only distilled water), cell (the test organisms + distilled water) and the test (the test organisms + influent and effluent wastewater samples) tubes were used in toxicity determinations by using the centrifuged cells. Fifty milligrams resazurin and 10 mg phosphate buffer were dissolved in 100 ml distilled water to prepare the resazurin solution. A 3.75 g of nutrient broth (Merck) was dissolved in 10 ml of distilled water and used as the growth media in the test tubes. Reagent control solution contained 0.275 ml growth medium; 9.525 ml distilled water and 0.2 ml resazurin solution. Cell control solution contained 0.2 ml growth medium; 9.6 ml distilled water; 0.2 ml resazurin solution and test tubes contained 0.2 ml growth medium; 9.6 ml centrifuged wastewater sample and 0.2 ml resazurin solution. The test tubes were incubated for 20 min at room temperature before the resazurin solution was added. After the resazurin solution addition to the tubes, the color was monitored until the cell solution was pink. After almost 30 min, 50 μl of $HgCl_2$ solution (10 mg/ml) was added to the cell control and the test tubes to stop the reaction following centrifugation for 5 min. In the presence of active bacterial culture with dehydrogenase enzyme activity, resazurin changes color from blue to pink forming the reduced compound resorufin. Inactive bacteria do not cause any change in resazurin color and remain blue. A spectrometer was used to determine the color change at a wavelength of 610 nm. Feed and effluent wastewater toxicities and percent toxicity removals were calculated as

follows.

$$\%\text{toxicity} = \frac{(A - B)}{(C - B)}$$

where A is the optical density (OD) of the test tubes containing feed or effluent wastewater; B the OD for the cell tube (cells + water with no TCP); and C is the OD for the control tube (only water).

Percent toxicity removals were calculated as $E = 1 - (\%\text{TOX}_f/\% \ \text{TOX}_e)$ where $\%\text{TOX}_f$ and $\%\text{TOX}_e$ are the percent toxicities of the feed and the effluent with respect to control.

3. Results and discussion

A set of experiments with six different hydraulic residence times (HRT) between 5 and 30 h were performed in order to determine the effects HRT on COD, TCP and toxicity removal in the hybrid reactor system. Feed COD, TCP and SRT were kept constant at $2700 \pm 100 \ \text{mg} \ l^{-1}$, $300 \pm 10 \ \text{mg} \ l^{-1}$ and 20 days, respectively.

Fig. 2 depicts variation of percent toxicity of 2,4,6-trichlorophenol (TCP) on the test organisms with the TCP concentration in distilled water. Toxicity of TCP increased with TCP concentration as expected with an IC_{50} value of nearly $45 \ \text{mg} \ l^{-1}$. TCP concentrations below $20 \ \text{mg} \ l^{-1}$ did not cause considerable toxicities whereas $80 \ \text{mg} \ l^{-1}$ TCP concentration resulted in nearly 100% toxicity for the test organisms.

Variation of percent TCP removal and effluent TCP concentrations with hydraulic residence time (HRT) are depicted in Fig. 3 for the packed column and the system (or aeration tank) effluents. Percent TCP removal increased and the effluent TCP decreased with increasing HRT. The effluent TCP's for the system were always lower than those of the packed column effluents indicating TCP removals in both reactors. However, most of the TCP was removed in the packed column at low HRT levels below 15 h. The contribution of the aeration tank for TCP removal was more pronounced at high HRT levels as shown in Fig. 3. The results indicated that the hybrid bioreactor system behaved like a homogenous single reactor at low HRT levels because of high recirculation ratio between the reactors yielding short recirculation or mixing times. The aeration tank did not only contribute to TCP and COD removals, but also provided dissolved oxygen to the biofilm culture in the packed column by effluent recir-
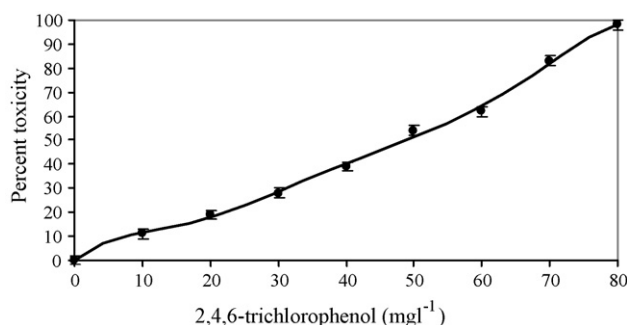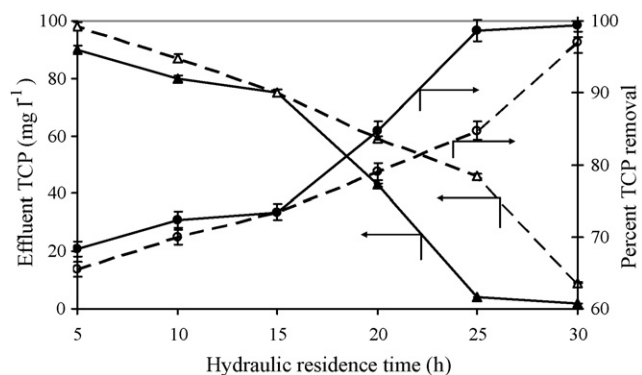

Fig. 3. Variation of percent TCP removal and effluent TCP with hydraulic residence time for both reactors. ▲, ● System effluent, △, ○ packed column effluent (broken lines). $COD_0 = 2700 \pm 100 \ \text{mg} \ l^{-1}$, $TCP_0 = 300 \pm 10 \ \text{mg} \ l^{-1}$, SRT = 20 days.

culation. Percent TCP removal decreased from 99% to 73% and further to 68% corresponding to the effluent TCP levels of $2 \ \text{mg} \ l^{-1}$ ($TCP_0 = 299 \ \text{mg} \ l^{-1}$), $75 \ \text{mg} \ l^{-1}$ ($TCP_0 = 282 \ \text{mg} \ l^{-1}$) and $90 \ \text{mg} \ l^{-1}$ ($TCP_0 = 284 \ \text{mg} \ l^{-1}$) when the HRT decreased from 30 to 15 h and further to 5 h, respectively. The system should be operated at an HRT of at least 25 h to obtain 99% TCP removal at a 20 days of sludge age.

Fig. 4 depicts variations of percent COD removal and the effluent COD concentrations with hydraulic residence time (HRT) for both reactors. Percent COD removal increased and the effluent COD decreased with increasing HRT. Percent COD removals were higher and effluent COD's for the system was lower than those of the packed column effluents indicating COD removals in both reactors. Aeration tank contributed considerably to the COD removal from the system. Percent COD removal increased from 78% to 84% and further to 90% with the effluent COD contents of $595 \ \text{mg} \ l^{-1}$ ($COD_0 = 2695 \ \text{mg} \ l^{-1}$), $450 \ \text{mg} \ l^{-1}$ ($COD_0 = 2880 \ \text{mg} \ l^{-1}$) and $279 \ \text{mg} \ l^{-1}$ ($COD_0 = 2790 \ \text{mg} \ l^{-1}$), respectively when the HRT increased from 5 to 15 h and further to 30 h. Non-toxic reactor or effluent TCP contents of below $5 \ \text{mg} \ l^{-1}$ obtained at high HRT levels above 25 h are the major reason for high COD and TCP removals. Since
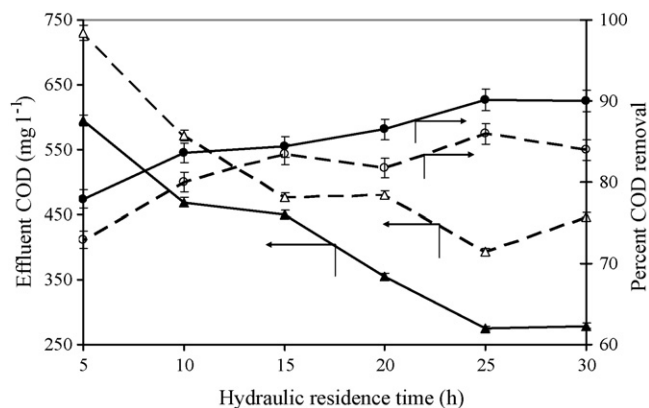

Fig. 2. Variation of percent toxicity of 2,4,6 TCP with the TCP concentration for the test organisms.

Fig. 4. Variation of percent COD removal and effluent COD with hydraulic residence time for both reactors. ▲, ● System effluent, △, ○ packed column effluent (broken lines). $COD_0 = 2700 \pm 100 \ \text{mg} \ l^{-1}$, $TCP_0 = 300 \pm 10 \ \text{mg} \ l^{-1}$, SRT = 20 days.
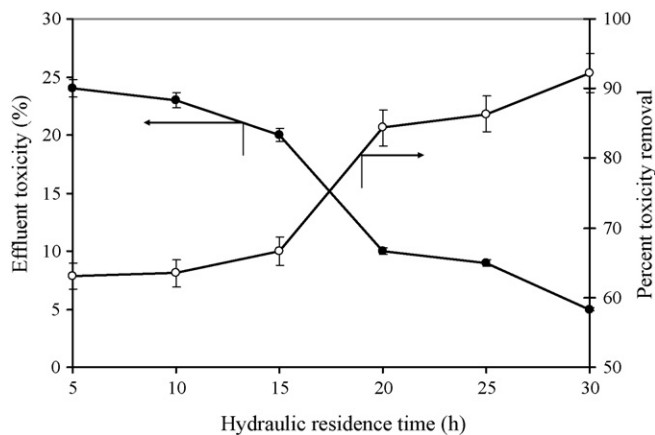
Fig. 5. Variation of percent toxicity removal and effluent toxicity with hydraulic residence time. $COD_0 = 2700 \pm 100 \, \text{mg l}^{-1}$, $TCP_0 = 300 \pm 10 \, \text{mg l}^{-1}$, SRT = 20 days.

the IC_{50} value of TCP is approximately $45 \, \text{mg l}^{-1}$, TCP contents lower than $30 \, \text{mg l}^{-1}$ did not cause considerable inhibitions yielding high COD and TCP removals.

Fig. 5 depicts variations of percent toxicity removals and the effluent toxicity levels with the hydraulic residence time (HRT). Variations in effluent toxicity showed somewhat similar trend to the effluent TCP since most of the toxicity results from the residual TCP in the effluent. HPLC analysis of the effluents did not show any intermediates from TCP biodegradation. Therefore, inhibition was only due to TCP content. Percent toxicity removals increased and the effluent toxicity levels decreased with increasing HRT. When the toxicity of the feed wastewater was nearly 65% with respect to TCP free wastewater (control), the effluent toxicities decreased from 24% to 20% and further to 5% with the corresponding percent toxicity removals from the feed of 63%, 67% and 92%, respectively when the HRT increased from 5 to 15 h and further to 30 h. The system should be operated at HRT levels above 25 h at a sludge age of 20 days for more than 92% toxicity removal.

Variations of biomass concentrations in the column reactor and in the aeration tank with the hydraulic residence time (HRT) are depicted in Fig. 6. Biomass concentration in the packed column reactor was always higher than those of the aeration tank as expected because of high cell retention in the column due to biofilm formation. Operation at high HRT levels resulted in high percent removals of COD, TCP and toxicity yielding high biomass concentrations since the biomass yield is proportional to the substrate (COD, TCP) utilization. As shown in Fig. 5, biomass concentration in both the column reactor and the aeration tank increased with increasing HRT because of decreasing reactor COD and TCP concentrations. Biomass concentrations in the packed column reactor and aeration tank increased from 2780 to $5320 \, \text{mg l}^{-1}$ and from 1300 to $2440 \, \text{mg l}^{-1}$ when HRT increased from 5 to 30 h. High biomass concentrations in the reactors at high HRT values above 25 h resulted in high percent COD, TCP, and toxicity removals of above 90%.

Fig. 7 depicts variation of specific (mg COD. g biomass^{-1} d^{-1}) and volumetric (mg COD l^{-1} d^{-1}) rates of COD removals with the hydraulic residence time. The
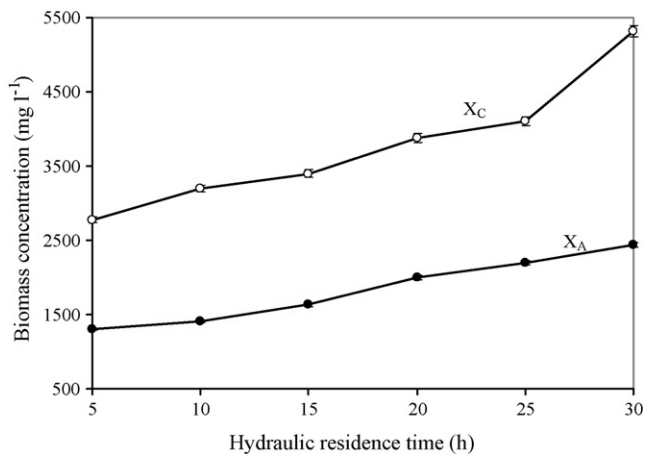

Fig. 6. Variation of biomass concentration in the packed column and the aeration tank with hydraulic residence time. $COD_0 = 2700 \pm 100 \, \text{mg l}^{-1}$, $TCP_0 = 300 \pm 10 \, \text{mg l}^{-1}$, SRT = 20 days.

volumetric and the specific rates were calculated by using the following equations:

$$R_V = \frac{Q(S_0 - S)}{V_T} = \frac{S_0 - S}{HRT}, \qquad R_x = \frac{Q(S_0 - S)}{V_C X_C + V_A X_A} \quad (1)$$

where Q is the flow rate of the feed wastewater which varied between 1.8 an $11.01 \, \text{d}^{-1}$; S_0 and S are the feed and effluent COD (or TCP) contents; V_T is the total liquid volume in both reactors (2.3 l); V_C (0.8 l) and X_C (g l^{-1}) are the liquid volume and the biomass concentrations in the column reactor, respectively; V_A (1.5 l) and X_A (g l^{-1}) are the liquid volume and biomass concentrations in the aeration tank, respectively. Both the volumetric and the specific rates of COD removal decreased with increasing HRT as expected from Eq. (1), although the effluent COD (S) also decreased with increasing HRT while the feed COD (S_0) was constant. Biomass concentrations in the packed column and also in the aeration tank also increased with HRT resulting in decreased specific COD removal rates at high HRT values since R_X is inversely related to the biomass concentration according to Eq. (1). High biomass concentrations obtained at high HRT operations resulted in low specific rates of COD removal. Both
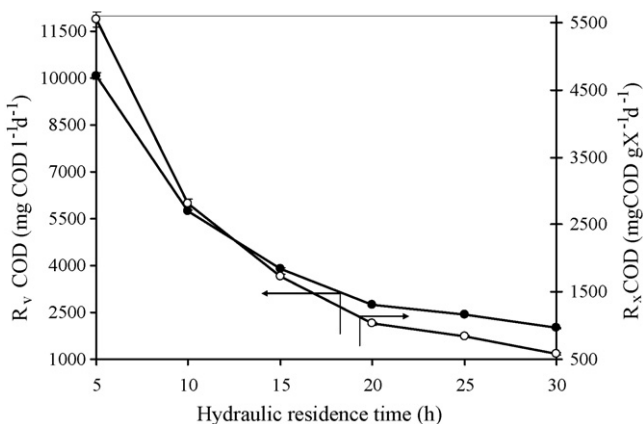

Fig. 7. Variation of volumetric and specific rates of COD removal with hydraulic residence time. $COD_0 = 2700 \pm 100 \, \text{mg l}^{-1}$, $TCP_0 = 300 \pm 10 \, \text{mg l}^{-1}$, SRT = 20 days.
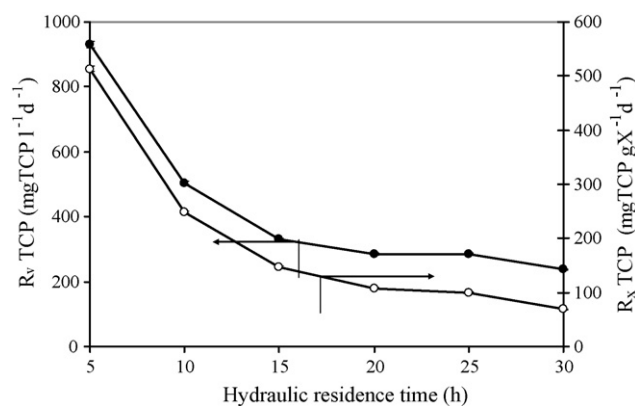
Fig. 8. Variation of volumetric and specific rates of TCP removal with hydraulic residence time. $COD_0 = 2700 \pm 100\,\mathrm{mg\,l^{-1}}$, $TCP_0 = 300 \pm 10\,\mathrm{mg\,l^{-1}}$, SRT = 20 days.

the volumetric and the specific rates of COD removal were maximum at an HRT of 5 h. The rates did not change significantly for HRT values above 25 h since the reactor COD and TCP concentrations were low.

Variations of specific (mg TCP. g biomass^{-1} d^{-1}) and volumetric (mg TCP l^{-1} d^{-1}) rates of TCP removals with the hydraulic residence time (HRT) are depicted in Fig. 8. The volumetric and the specific rates were calculated using Eq. (1) by replacing COD with TCP where S_0 and S are the feed and the effluent (or reactor) TCP concentrations in this case. Again, both the volumetric and the specific TCP removal rates decreased with increasing HRT as estimated by Eq. (1). Specific rates of TCP removal decreased with increasing HRT since the biomass concentrations in the reactors increased with HRT. The maximum volumetric and the specific rates of TCP removals were obtained at an HRT of 5 h. Decreases in the rates were not that significant for HRT levels above 20 h because of low levels of reactor COD and TCP at high HRT values above 20 h. The reactor or effluent TCP concentrations were very low ($<10\,\mathrm{mg\,l^{-1}}$) for HRT values above 25 h while the biomass concentrations were high resulting in low specific rates of TCP removal for HRT values above 25 h.

Relatively low TCP removals of less than 65% with low feed TCP contents of less than $100\,\mathrm{mg\,l^{-1}}$ were reported in literature studies on aerobic treatment of TCP containing wastewaters [11,33]. Anaerobic–aerobic combined treatment systems resulted in relatively higher TCP removals due to anaerobic degradation of TCP [2,3,32]. However, the feed TCP contents in those studies were much lower than $100\,\mathrm{mg\,l^{-1}}$. As compared to the literature studies on biological treatment of TCP containing wastewaters our study resulted in more than 90% COD, TCP and toxicity removals from synthetic wastewater containing much higher TCP concentrations of $300\,\mathrm{mg\,l^{-1}}$ at hydraulic residence times (HRT) above 25 h at a sludge age of 20 days.

4. Conclusions

Synthetic wastewater containing $300\,\mathrm{mg\,l^{-1}}$ of 2,4,6 TCP was biologically treated at different hydraulic residence times (HRT) between 5 and 30 h using a hybrid loop-bioreactor system consisting of a packed column and an aeration tank with effluent

recycle. The feed TCP, COD and SRT (sludge age) were kept constant at $300 \pm 10\,\mathrm{mg\,l^{-1}}$, $2700 \pm 100\,\mathrm{mg\,l^{-1}}$ and 20 days, respectively throughout the study. Percent COD, TCP and toxicity removals increased with increasing HRT resulting in low effluent TCP, COD and toxicity levels at high HRT levels.

Both the column reactor and the aeration tank contributed TCP and COD removals. However, the packed column reactor removed most of the TCP at low HRT levels below 15 h. Biomass concentrations in the packed column and the aeration tank increased with increasing HRT due to extensive utilization of substrate and low reactor TCP concentrations causing negligible TCP inhibitions at high HRT operations. Biomass concentrations in the column reactor were higher than those of the aeration tank because of high cell retention by biofilm formation. Low effluent TCP and high biomass concentrations at high HRT operations above 25 h resulted in more than 90% COD, TCP and toxicity removals.

Both the specific and the volumetric rates of COD removal decreased with the HRT due to high biomass concentrations at high HRT operations and also because of the inverse relationship between the rates and the HRT. Specific and the volumetric rate of TCP removals showed the same trend as that of the COD and decreased with increasing HRT because of the same reasons.

The rates of COD and TCP removals somewhat levelled off at high HRT levels above 20 h due to low TCP and COD contents. The system should be operated at HRT values of 25 h and above in order to accomplish more than 90% TCP, COD and toxicity removals under the experimental conditions employed.

Acknowledgements

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