Continuous Operation of Foamed Emulsion Bioreactors Treating Toluene Vapors

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Abstract: Continuous operation of a new bioreactor for air pollution control called the foamed emulsion bioreactor (FEBR) has been investigated. The effect of several liquid feeding strategies was explored. The FEBR exhibited high and steady toluene removal performance (removal efficiency of 89%–94%, elimination capacity of 214–226 g/m^{3}h at toluene inlet concentration of 1 g/m^3) for up to 360 h, when 20% of the culture was replaced every 24 h by a nutrient solution containing 4 g/L of potassium nitrate as a nitrogen source. This feeding mode supported a high cell activity measured as INT reduction potential and active cell growth without being subject to nitrogen limitation. In comparison, operating the FEBR with the liquid in a closed loop (i.e., batch) resulted in a significant decrease of both the removal efficiency of toluene and INT reduction activity. Operation with feeding active cells resulted in stable and effective treatment, but would require a significant effort for mass culture preparation. Therefore, the continuous process with periodically feeding nutrients was found to be the most practical and effective operating mode. It also allows for stable operation, as was shown during removal of low concentration of toluene or after pollutant starvation. Throughout the study, INT reduction measurements provided insight into the process. INT reduction activity data proved that under normal operating conditions, the FEBR performance was limited by both the kinetics and by mass transfer. Overall, the results illustrate that engineered gas-phase bioreactors can potentially be more effective than conventional biofilters and biotrickling filters for the treatment of air pollutants such as toluene.

Keywords: VOC control; biofilter; air pollution control; toluene; biologically activated foam; biodegradation

INTRODUCTION

Biological treatment is an established technology for air pollution control and the alternative of choice to physical and chemical treatment techniques because of its effectiveness for the control of odors and volatile organic compounds, especially in high flow rates—low concentrations cases (Devinny et al., 1999; Leson and Winer, 1991).

The most widely utilized bioreactors for air pollution control are biofilters and biotrickling filters (Cox and Deshusses, 1998; Devinny et al., 1999). Biofilters are reactors in which a humid polluted air stream is passed through a porous packed bed on which a mixed culture of pollutant-degrading organisms is naturally immobilized (Devinny et al., 1999). In biotrickling filters, a distinct free water phase containing various nutrients is trickled over a packed bed (Cox and Deshusses, 1999; Gabriel and Deshusses, 2003; Mpanias and Baltzis, 1998; Pedersen and Arvin, 1995). Both biofilters and biotrickling filters have some limitations of performance although they are currently used for treating volatile organic compounds, odorous compounds, and other air pollutants. Biofilters typically have low pollutant elimination capacities, due to the low cell activity of essentially resting cells in the reactors, while biotrickling filters have often experienced clogging from excessive biomass growth, which results in high pressure drop and process instability (Cherry and Thompson, 1997; Cox and Deshusses, 1999; Fürer and Deshusses, 2000; Laurenzis et al., 1998; Smith et al., 1996).

Recently, we have developed a new vapor phase bioreactor named the foamed emulsion bioreactor (FEBR) that overcomes some of the limitations of biofilters and biotrickling filters (Kan and Deshusses, 2003). The FEBR consists of an emulsion of highly active pollutant-degrading microorganisms culture and a water-immiscible organic phase, which is made into a foam with the air being treated (Fig. 1). After the desired treatment is achieved, the foam is continuously collapsed, and the cells with the emulsion are reused. The FEBR has high oxygen and pollutant mass transfer rates due to the large interfacial area between gas and liquid of the fine foam and a high partitioning of pollutants into the organic phase. Rapid biodegradation of the pollutants is achieved by a high-density bacterial culture actively growing. Bed clogging and associated pressure drop problems are prevented by using a moving foam rather than an immobilized culture growing on a support. We demonstrated a higher performance for toluene removal in the FEBR than in most current bioreactors. The FEBR reached an elimination capacity of 280 $g_{toluene}/m^3{}_{reactor}h$ with a removal efficiency of 95% at a gas residence time of 15 s and toluene inlet concentration of 1–1.2 g/m^3 for operation lasting 2 to 8 h (Kan and Deshusses,

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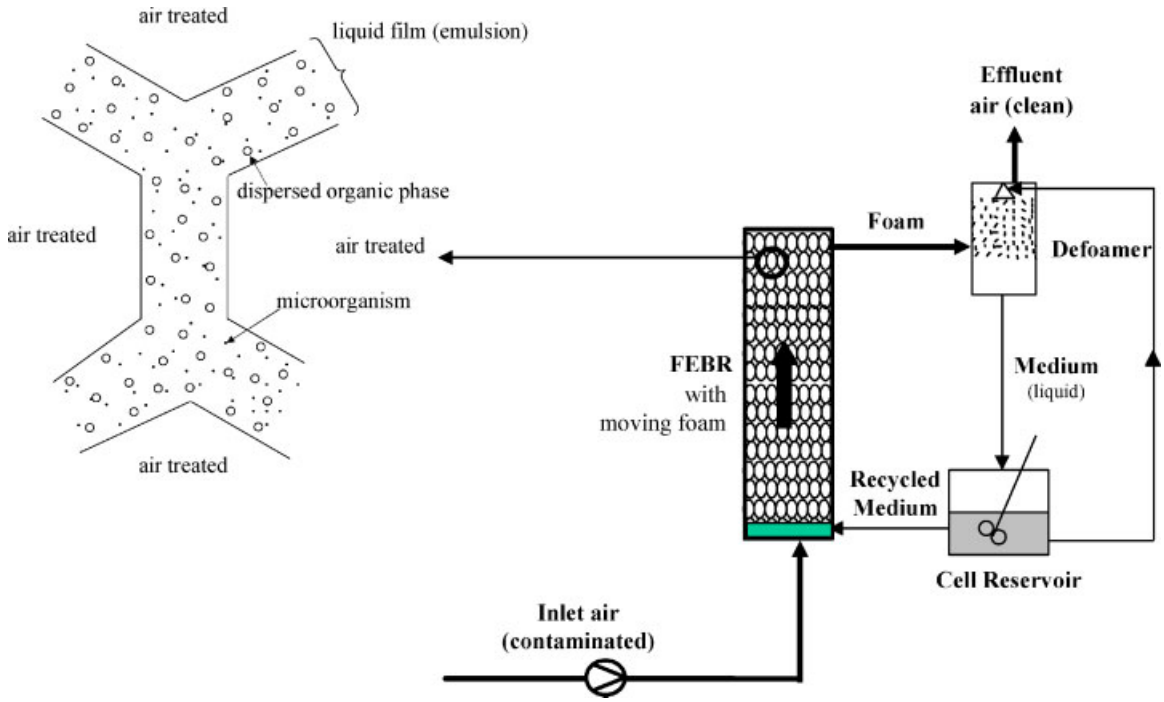

Figure 1. FEBR concept and configuration. The schematic on the left shows the proposed structure of the foamed emulsion which has dispersed organic phase droplets (4–10 μm diameter) and microorganisms in a liquid film (130–160 μm thick) as well as entrapped air (2–3 mm pockets) with pollutant vapors. The schematic on the right shows the basic configuration of the FEBR in a closed-loop batch operation. [Color figure can be seen in the online version of this article, available at www.interscience.wiley.com.]

2003). When pure oxygen was added to the inlet air stream of the FEBR (to lessen oxygen limitation), the FEBR exhibited an elimination capacity up to 408 $g/m^3_{reactor}h$ with 77% removal at the same gas contact time as above (Kan and Deshusses, 2003). Also, a conceptual mathematical model of the foamed emulsion system was developed and showed good agreement with the experimental data (Kan and Deshusses, 2004).

However, until now, FEBR operation was only shown for relatively short periods of time and batch operation of the culture, and demonstration of sustained treatment is required before practical application in the field. Deactivation of cells and decrease of performance are often observed over the long-term in biofilters because of deteriorating environmental conditions, such as accumulation of byproducts or nutrient limitation resulting in biokinetic limitations (Holubar et al., 1999; Moe and Irvine, 2001; Morgenroth et al., 1996; Song et al., 2003). Also, most of bioreactors for air pollution control are exposed to non-ideal conditions such as fluctuating pollutant loads and periods of pollutant starvation, which may affect pollutant removal efficiency (Cox and Deshusses, 2002; Metris et al., 2001).

In light of these, the objective of the present study was to develop strategies for continuous operation of the FEBR with high and stable toluene elimination, as well as high and constant cell activity and biomass concentration. Different modes of continuous operation were investigated either by periodically replacing part of the culture in the FEBR with a nutrient solution or with active toluene-degrading cells. The effect of toluene starvation on the FEBR performance, biomass concentration, and cell activity measured as INT reduction activity was also examined.

MATERIALS AND METHODS

Reactor Setup

The FEBR system was described previously by Kan and Deshusses (2003). The reactor consisted of the actual foam column, a cell reservoir, and a defoamer (Fig. 1). The foam column (4.04 cm ID, 40 cm high, volume of 0.51 L) had a fine gas sparger at the bottom of the reactor. A metered stream of toluene contaminated air was introduced through the gas sparger while an emulsion consisting of the mineral medium, the active culture (see below), the organic phase (oleyl alcohol, Sigma Co., Ltd, St. Louis, MO), and the surfactant (DC-100 silicone, Sigma) was introduced at the bottom of the reactor. The mineral medium consisted of 1 g/L KH_2PO_4, 1 g/L K_2HPO_4, 1 g/L KNO_3, 1 g/L NaCl, 0.2 g/L $MgSO_4$, 26 mg/L $CaCl_2 \cdot 2H_2O$, 5.2 mg/L EDTA $Na_4(H_2O)_2$, 1.5 mg/L $FeCl_2 \cdot 4H_2O$, 0.12 mg/L $CoCl_2 \cdot 6H_2O$, 0.1 mg/L $MnCl_2 \cdot 2H_2O$, 0.07 mg/L $ZnCl_2$, 0.06 mg/L H_3BO_3, 0.025 mg/L $NiCl_2 \cdot 6H_2O$, 0.025 mg/L $NaMoO_4 \cdot 2H_2O$, 0.015 mg/L $CuCl_2 \cdot 2H_2O$. After rising through the reactor, the foam leaving through a side port was defoamed in a defoamer by continuously spraying the foam with the emulsion from the cell reservoir. The liquid was returned to the cell reservoir (a 0.8 L flask) prior to be recycled to the foam generation column. The total amount of liquid in the system was about 0.5 L. The air was not humidified prior to

entering the FEBR and in all experiments, about 60 mL of distilled water were added to the FEBR every day to compensate for evaporation.

The toluene-degrading consortium used in the experiments was initially obtained from a biotrickling filter in our laboratory. The mixed culture was grown prior to each experiment by bubbling toluene-laden air (1–2 g/m^3) through mineral medium in a 10 L bubble column reactor, and concentrated by centrifugation before each experiment.

Effect of Operating Conditions on the Performance of the FEBR

The study of the operation of the FEBR was carried out with continuously feeding toluene-contaminated air, while the cell suspension/emulsion was recycled in the FEBR-defoamer-cell reservoir. To better understand the effect of cell and mineral medium feeding on the performance of the FEBR, batch and continuous liquid feeding were investigated. During the batch experiment, no toluene-degrading cells or nutrients were supplied, and the cell culture was in a closed-loop. Only water was added daily to compensate for evaporation. For continuous operation, two modes of operation were investigated. The first mode was to replace 20% (vol.) of the FEBR culture once per day with a fresh culture of toluene-degrading cells. This was to compensate for the deactivation of cells in the FEBR. In the second mode, 20% (vol.) of the culture was withdrawn daily from the FEBR, and replaced by a concentrated nutrient solution. The concentrated nutrient solution was the same mineral medium described above except that the potassium nitrate was increased fourfold to 4 g/L. Under the latter conditions, active growth of toluene-degrading cells in the reactor was stimulated. In all cases, the starting operating conditions were 12 g_{dw}/L of toluene-degrading consortium, 5% (v/v) oleyl alcohol, 1 g/m^3 inlet concentration of toluene at 15 s gas retention time. Continuous operation of the FEBR at low toluene inlet concentration was also conducted by periodically replacing 10% (vol.) of the reactor culture with the concentrated nutrient solution. The same initial operating conditions as above were used except that the inlet toluene concentration was 0.5 g/m^3, and that the initial cell concentration was 13 g_{dw}/L. During the experiments, biomass concentration, cell activity in terms of INT reduction potential (thereafter INT reduction activity activity) (see below), toluene removal efficiency, and residual nitrate concentration in the culture were monitored.

Effect of Toluene Starvation on the FEBR

After steady state continuous operation of the FEBR at 0.5 g/m^3 for 48 h, the supply of toluene in the air was stopped for 48 h and then restarted at a concentration of 1 g/m^3. During starvation, only fresh air was supplied to the FEBR at a gas retention time of 15 s; no nutrient solution or culture were supplied. The changes in biomass concentration, INT reduction activity and toluene removal efficiency were monitored.

Analytical Methods

Biomass concentration was determined by measuring protein concentration using the BCA protein assay (Pierce Chemical Co., Rockford, IL) and assuming that cell dry weight was twice the protein concentration. Cell activity was measured as the INT reduction activity. The INT (2-(*p*-iodo-phenyl)-3-(*p*-nitrophenyl)-s-phenyl tetrazolium chloride) assay was selected because its response is proportional to the fraction of active cells in a culture, and the test provides relatively easy, yet accurate measurement of microbial activities (Fonseca et al., 2001; Kim et al., 1994; Rodriguez et al., 1992; Yu et al., 1995). The method was similar to Rodriguez et al. (1992) except for the preparation and extraction of culture samples. The final concentration of INT and the incubation time for the measured samples were investigated prior to analyzing the INT reduction activity during the experiments; 0.12% (w/v) INT and 2 h incubation were found to be the optimum conditions for the INT reduction activity of the culture samples (200–250 mg protein/L in the experiments). Cells from the FEBR were harvested every 24 h, and incubated with 0.12% (w/v) INT in the dark for 2 h. Abiotic controls were also incubated with 0.12% (w/v) INT for 2 h, after killing the cells with 37% (w/v) formaldehyde. After incubation, 37% (w/v) formaldehyde was added to the cultures to stop the reaction and methanol was then added as to extract INT formazan (INTF), the reduced form of INT, which is a red crystal. Samples were sonicated and extracted with methanol in the dark for 30 min, after which the absorbance at 480 nm was measured with a spectrophotometer. The INT reduction activity (mmol of INTF per g of protein) were calculated by using the net optical density of the samples and calibration of optical density at 480 nm versus INT reduction activity. The net optical density of the samples at 480 nm was obtained by subtracting the optical density of the controls from the optical density of the sample cells and dividing it by the protein concentration. Also, the calibration of optical density at 480 nm versus INTF concentration (mmol of INTF/L) was the following: INTF concentration $= 0.19 \times OD_{480nm}$ ($R^2 = 0.99$).

Selected samples were analyzed for toluene-induced oxygen uptake rate (OUR) as a second measure of the culture activity. OUR determinations were conducted at room temperature in a 2.7 mL custom-made vessel fitted with a YSI oxygen probe and meter (YSI Inc., Yellow Springs, OH). Samples were first saturated with air and monitored for endogenous respiration rate for 2–5 min depending on the activity. The toluene-induced OUR was determined after addition of a concentrated toluene solution to reach a final concentration of 0.19 mM in the vessel, and the OUR was corrected for the endogenous respiration.

Nitrate concentration in the culture was analyzed by using Nitrate Kit (CHEMetrices, Inc., Calverton, VA). The reactor setup included on-line monitoring of CO_2 (non-dispersive

infrared) and dissolved oxygen (electrochemical sensor) both from Vernier Instruments (Beaverton, OR). Gaseous toluene concentrations were measured by gas chromatography (HP 5890) and a FID detector. The foam stability was assessed using the 50% drain time according to the method described by Ripley et al. (2000).

The size of each foam element and the diameter of the organic phase droplets were determined experimentally by image analysis, while the liquid film thickness was calculated from the liquid holdup assuming the foam bubbles to be dodecahedral in shape.

RESULTS AND DISCUSSION

For stable operation of the FEBR and sustained pollutant treatment, foam stability and cell activity are of prime importance. In all experiments, a very good foam stability was observed, even after several hundred hours of batch or continuous operation. The 50% drain time of foam (Ripley et al., 2000), that is, the time required for foam to loose half of its volume when at rest, was in the range of 3–5 min, which is much larger than the foam life in the system. Therefore, maintaining cell activity in the FEBR is probably the most important parameter required to sustain a high pollutant removal efficiency during long term operation.

The results of batch operation of the FEBR are shown in Figure 2. The operating conditions were designed to achieve about 90% of toluene removal, as predicted by a mathematical model of the FEBR (Kan and Deshusses, 2004) and previous experimental results (Kan and Deshusses, 2003). During the 144 h experiment with the liquid recycled in a closed loop, no external nutrient solution and new cells were added to the FEBR while toluene-contaminated air was supplied continuously. Toluene removal efficiency decreased continuously from 90% to 64% after 144 h (Fig. 2). At the same time, biomass concentration and INT reduction activity also dropped from 12 to 8 g/L and 5.2 to 0.6 mmol INTF/$g_{protein}$, respectively. This is a 30% loss in the biomass and 88% loss of the INT reduction activity. The INT reduction activity dropped to half of the initial activity after 24 h and slightly decreased after 48 h, while biomass concentration kept decreasing over time. The decay of the cell activity measured as INT reduction is most likely due to the accumulation of unfavorable by-products in the culture, the damage to the enzymatic system and the absence of cell growth due to nutrient limitation (Bailey and Ollis, 1986; Duetz et al., 1994; Kirchner et al., 1991; Weber and Hartmans, 1996). After 48 h, a much slower decrease in INT reduction activity is observed, possibly because of some shifts in the cell metabolism. The concurrent decrease of the biomass concentration is most likely due to biomass death and lysis and possibly secondary processes such as predation by higher organisms (Cox and Deshusses, 2002). Nitrogen is usually one of the most critical nutrients for biomass growth, although some nitrogen recycling has been identified in complex biological systems and in gas-phase bioreactors, such as biofilters and biotrickling filters (Bailey and Ollis, 1986; Gribbins and Loehr, 1998; Rittmann and McCarthy, 2001; Song et al., 2003). During the batch experiment, little free nitrate (0–1 ppm) was detected in the culture of the FEBR, indicating that nitrogen limitation probably contributed to cell decay. The decrease in INT reduction activity over time indicates that cells shifted to a lower metabolism, as a result of their low or negative growth rate. From Figure 2, first order INT activity and biomass decay constants of 0.216/day ($R^2 = 0.89$) and 0.0768/day ($R^2 = 0.93$), respectively, were calculated. Those decay constants can be included in the mathematical model of the FEBR (Kan and Deshusses, 2004), in order to improve the accuracy of the model. As mentioned, INT reduction activity decreased 88% during the experiment, while toluene removal only decreased 28%. Other experiments in shake flasks with suspended mixed cultures (not shown) revealed that the INT reduction activity was proportional to the toluene degradation activity of the culture. Hence, the lack of linear correlation between the decrease in INT reduction activity and the FEBR performance shown in Figure 2 suggests that the reactor was initially subjected to some limitation(s) other than a kinetic limitation. These could be diffusion limitations of either toluene or of oxygen.

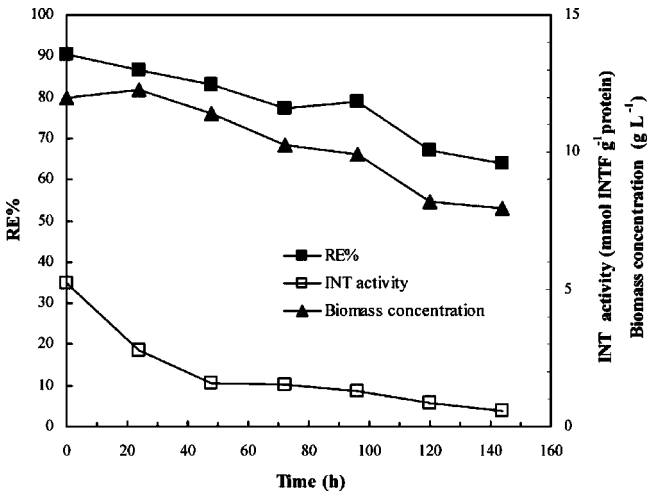

Figure 2. Batch operation of the FEBR. Conditions: toluene inlet concentration, 1 g/m^3; oleyl alcohol concentration, 5% (v/v); silicone surfactant concentration, 0.4% (v/v); initial biomass concentration, 12 g_{dw}/L; gas retention time, 15 s. During the experiment, no external nutrient solution and new cells were added to the FEBR while toluene-contaminated air was supplied continuously. INTF represents INT formazan, a reduced form of INT.

The experiment of Figure 2 suggested that replacing or replenishing part of the culture was necessary to maintain high biodegradation activity and high toluene removal in the FEBR. Hence, two different modes of continuous operations were investigated and results are presented in Figures 3 and 4. In the first case (Fig. 3), the FEBR was operated such that 20% (vol.) of the FEBR culture was replaced every 24 h by the same volume of an active toluene-degrading cells suspension in order to compensate for the decay of INT reduction activity and maintain the same biomass concentration. The active toluene-degrading cells were grown on toluene (1–2 g/m^3) in a bubble column bioreactor for 24–48 h and fed to the

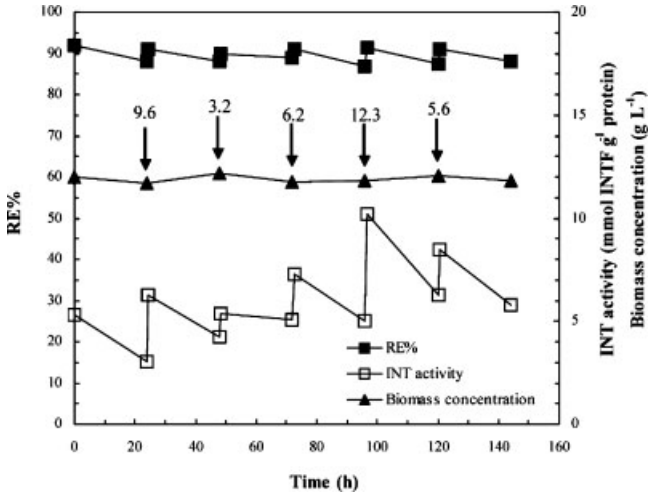

Figure 3. Continuous operation of the FEBR by periodically replacing 20% (v/v) of the reactor culture with active cells. Conditions: toluene inlet concentration, 1 g/m^3; oleyl alcohol concentration, 5% (v/v); silicone surfactant concentration, 0.4% (v/v); initial biomass concentration, 12 g_{dw}/L; gas retention time, 15 s. The arrows indicate feeding of the active cells, after purging 20% (v/v) of the FEBR culture every 24 h. The numbers in the legend show the toluene-induced oxygen uptake rates (OUR) of the added cells (in mg O_2/ming of protein).

FEBR after being harvested and concentrated by centrifugation. The 20% vol. fraction replacement was determined from the first order deactivation rate (0.22/day) plotted in Figure 2 and model simulations (not shown) that revealed that a dilution rate of 0.2/day would result in a high and constant biomass concentration. As shown in Figure 3, cell activity in terms of INT reduction potential decreased significantly during each cycle (32%–43% loss), which is consistent with the results of the batch experiments. However, the decrease of INT reduction activity was compensated by the feeding of active toluene-degrading cells to the FEBR. Since the activity of the cells that were fed varied (see the toluene-induced OUR values in Fig. 3), a different

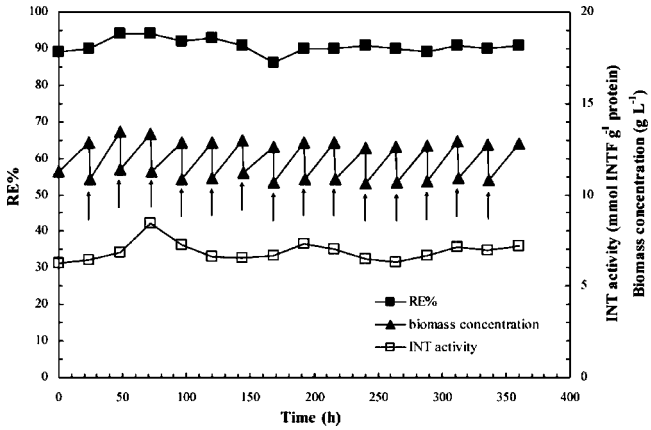

Figure 4. Continuous operation of the FEBR by periodically replacing 20% (v/v) of the reactor culture with a nutrient solution. Conditions: toluene inlet concentration, 1 g/m^3; oleyl alcohol concentration, 5% (v/v); silicone surfactant concentration, 0.4% (v/v); initial biomass concentration, 12 g_{dw}/L; gas retention time, 15 s. The arrows indicate feeding of the nutrient solution, after purging 20% (v/v) of the FEBR culture every 24 h.

degree of increase of INT reduction activity was observed after adding the active cells. The biomass concentration was constant at 11.7–12.2 g_{dw}/L and a high toluene removal efficiency (88%–92%), and high elimination capacity of toluene (211–221 g/m^{3}h) were maintained during the 144 h of the experiment. Although the INT reduction activity was not directly proportional to the toluene removal efficiency, it proved useful for assessing the metabolic activity and gaining insight into the process. Overall, operation of the FEBR was stable and satisfactory, however, this operating mode required preparing active cells to be fed periodically. This would probably be costly to implement at a larger scale. Furthermore, pollutant removal under these conditions relies mostly on cells at stationary phase rather than growing cells, which could possibly limit the performance.

In the next experiment, the FEBR was operated in a similar manner, except that mineral medium was fed instead of active cells. Twenty percentage of the reactor volume was replaced daily, whose value was determined by calculation using toluene load rate, biomass growth rate, and yield coefficient and assumption of steady state. This strategy was expected to promote cell growth, which would result in higher cell activity. The mineral medium was modified to have enough nitrate-nitrogen to support effective biomass growth. Calculations based on theoretical growth yields ($1.02 < Y_{X/C} < 1.15$ g_{dw}/g_C and $8.3 < Y_{X/N} < 20$ g_{dw}/g_N, Pirt, 1975) suggest that a carbon to nitrogen (C/N) supply ratio of 7.2–20 is theoretically required. However, C/N supply ratios in gas phase bioreactors systems are generally much higher, ranging typically from 13 to 34 (Song and Kinney, 2000; Song et al., 2003) up to 61 (Smith et al., 1996), or even much higher in biofilter systems, in which little to no continuous supply of nutrients is made. Thus, the potassium nitrate concentration in the FEBR feed was increased fourfold to 4 g/L so that supplied C/N ratio would be 32, which would prevent severe nitrogen limitation. Residual nitrate was monitored to ensure that no nitrogen limitation occurred. Other nutrients were kept unchanged, as their concentrations were in excess. The results are shown in Figure 4, which is markedly different from Figure 3. During each cycle, biomass concentration increased, as a result of biomass growth and relatively stable INT reduction activity was maintained for over 360 h of operation. The active cell growth resulted in high cell activity (measured as INT reduction activity) required for stable and high toluene removal performance. Removal efficiency ranged from 89 to 94% and toluene elimination capacity ranged from 214 to 226 g/m^{3}h. This is much higher than the 60–80 g/m^{3}h usually obtained with current bioreactors, if one excepts the high performance of up to about 270 g/m^{3}h achieved by fungal biofilters (Garcia-Pena et al., 2001; Woertz et al., 2001). The specific biomass growth rates during each FEBR cycle was 0.17–0.22/day which is almost equal to the dilution rate of 0.2/day. This meant that the reactor, which operation was initiated close to steady-state biomass concentration (12 g_{dw}/L), readily reached steady state during the experiment. During the 360 h operation, the dissolved oxygen

concentration ranged from 0.8 to 1 mg/L, which indicates that some degree of oxygen limitation occurred. This is consistent with the interpretation of the INT reduction activity data discussed above, and the speculation that both kinetic and some mass transfer limitations occurred.

Figure 5 presents carbon balances and C/N ratios during the experiment with periodical replacement of the culture with mineral medium illustrated in Figure 4. The carbon balances revealed that about 27% of carbon-toluene degraded was incorporated into the biomass, while the rest was mineralized to carbon dioxide. Interestingly, these numbers are not significantly different from those obtained in some biofilters and biotrickling filters (Cox and Deshusses, 1999; Jorio et al., 2000; Song and Kinney, 2000; Song et al., 2003). The C/N ratios, calculated by dividing the carbon-toluene degraded with the nitrate-nitrogen consumed by the microorganisms during the experiment, was 30 ± 2 (see Fig. 5). This is slightly higher than in usual fermentations, but as mentioned above, it is lower than in most biofilters or biotrickling filters. Throughout the experiments, selected liquid samples were analyzed for nitrate just prior to culture/medium replenishment. The residual nitrate (37–44 mg nitrate/L) showed that no nitrogen limitation occurred during the operation. Thus, the continuous operation with regular feeding of a concentrated nutrient solution was found to be effective in sustaining a high pollutant removal efficiency by supporting active cell growth without nitrogen limitation.

The three operating modes discussed previously are compared in Figure 6. As shown in Figure 6, the continuous processes resulted in better toluene removal efficiencies and more constant INT reduction activities than the batch process. Both continuous processes allowed to sustain a high toluene removal efficiency (>90%) as well as maintaining a high cell activity in terms of INT reduction potential (average INT activity 6.9 mmol of INTF/g protein when feeding nutrient, 6.2 mmol of INTF/g protein when feeding active cells). However, considering that continuous feeding of active cells requires cell production in another bioreactor, continuous feeding of mineral medium was found to be the most effective and practical strategy.

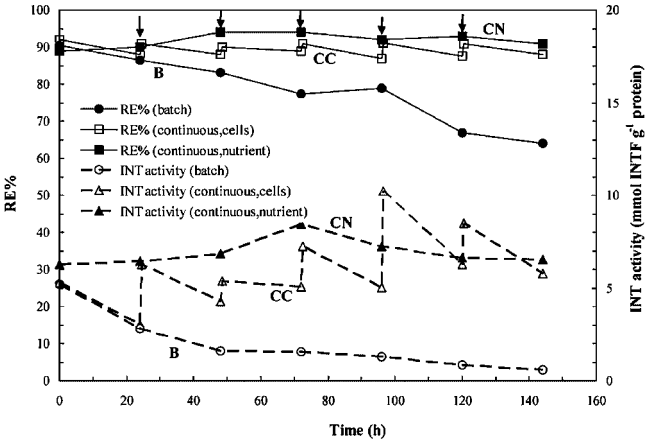

Figure 6. Comparison of batch and continuous operations of the FEBR. See Methods for detailed conditions. The arrows indicate feeding of the nutrient or the active cells every 24 h (except for the batch operation). B: represents removal efficiency and INT activity in batch operation, CC in continuous operation with feeding active cells and CN in continuous operation with feeding nutrients.

Continuous operation with feeding mineral medium was applied to a lower toluene inlet concentration (0.5 g/m^3) to determine whether growth could be sustained at less favorable conditions, and to possibly demonstrate the feasibility and the flexibility of the process (Fig. 7). The feeding rate was reduced to 10% of the culture volume per day in order to increase cell retention time in the system. As shown in Figure 7, the toluene removal efficiency was kept at 91%–92% except in the first 24 h, during which there was a brief acclimation phase due to starting the system with an inoculum of lower cell activity. After that short phase,

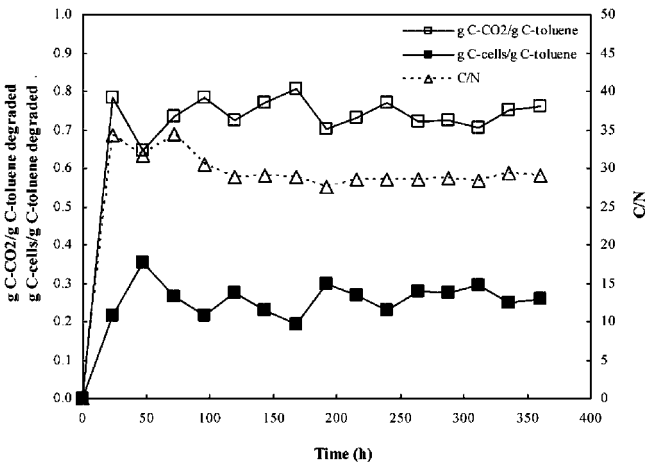

Figure 5. Carbon balances and C/N ratios (C degraded/N consumed) during continuous operation of the FEBR while periodically replacing 20% (v/v) of the reactor culture with a nutrient solution. Conditions are as in Figure 4. The arrows indicate feeding of the nutrient solution.

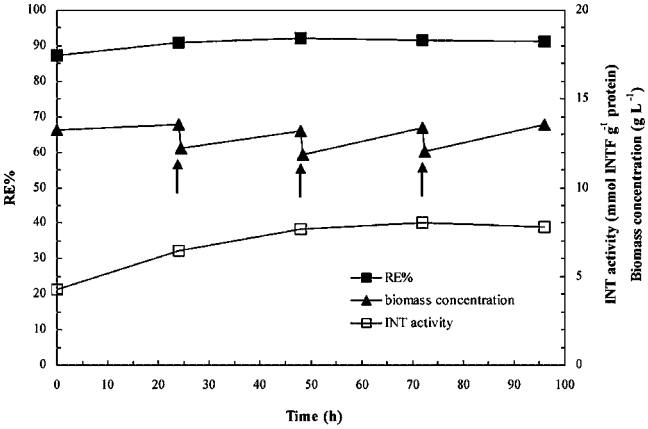

Figure 7. Continuous operation of the FEBR at low toluene inlet concentration by periodically replacing 10% (v/v) of the reactor culture with a nutrient solution. Conditions: toluene inlet concentration, 0.5 g/m^3; oleyl alcohol concentration, 5% (v/v); silicone surfactant concentration, 0.4% (v/v); initial biomass concentration, 13 g_{dw}/L; gas retention time, 15 s. The arrows indicate feeding of the nutrient solution, after purging 10% (v/v) of the FEBR culture every 24 h.

24 h, biomass concentration and INT reduction activity remained steady for over 96 h, indicating reasonable cell growth. At steady state, the specific growth rate was 0.08–0.12/day, that is, close to the dilution rate of 0.1/day. The results of Figure 7 illustrate that the FEBR can provide long-term stable and effective treatment at a lower toluene inlet concentration.

Bioreactors for air pollution control are often exposed to non-ideal conditions such as fluctuating inlet concentrations or air flows, or pollutant starvation rather than steady conditions typical of laboratory experiments (Deshusses et al., 1996; Shareefdeen and Baltzis, 1994). Among non-ideal conditions, pollutant starvation is one frequently occurring in practical situations (Cox and Deshusses, 2002; Metris et al., 2001) and one of concern for FEBR operation. Pollutant starvation may be the result of interruptions in the plant operation, weekend recess, or equipment malfunctions leading to interruptions in the feed of pollutant. Definition of the response of the system in the absence of pollutant feed, and characterization of the recovery of the reactor when resuming treatment are important aspects in practical applications. The effect of toluene starvation on the biomass concentration and on the INT reduction activity in the FEBR was investigated by stopping the feed of toluene contaminated air for 48 h (Fig. 8). The biomass concentration decreased slightly within the first 24 h, and then decreased drastically between 24 and 48 h, whereas the INT activity decreased more significantly from 0 to 24 h than from 24 to 48 h. From the results of Figure 8, the first order decay constant of the INT activity and of the biomass concentration were 0.36/day ($R^2 = 0.94$) and 0.15/day ($R^2 = 0.92$). It is likely that the process culture switched from a highly active metabolism (growth) to a lower metabolism (maintenance) within the first 24 h. Both the biomass concentration and the INT activity significantly increased after resuming supply of toluene in the inlet air and feeding the nutrient solution. The toluene removal efficiencies after 24 and 36 h re-acclimation with 1 g/m^3 toluene were 86% and 89%, respectively, while the toluene removal after 12 h was only 58%. The latter value does not compare well with the 89% removal observed prior to the starvation. This indicates that it may take 24 to 36 h to recover full treatment capacity, if one relies on growth within the FEBR only. One can reasonably speculate that temporary feeding of high activity cells after a major upset instead of feeding mineral medium may be able to reduce the recovery time.

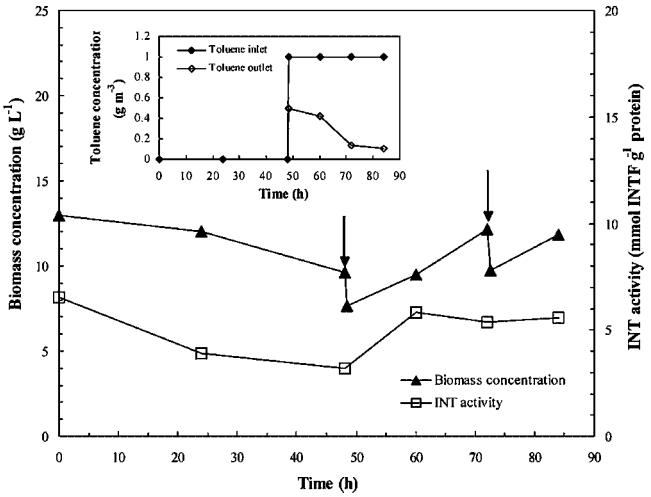

Figure 8. Effect of toluene starvation on the FEBR. Conditions: toluene inlet concentration: 1 g/m^3 after 48 hours, 0 g/m^3 during starvation (0–48 hours); oleyl alcohol concentration, 5% (v/v); silicone surfactant concentration, 0.4% (v/v); initial biomass concentration, 13 g_{dw}/L; gas retention time, 15 s. The arrows indicate feeding of the nutrient solution. The inset shows the recovery of toluene removal when resuming operation.

CONCLUSIONS

Batch and continuous operations were investigated in order to develop the baseline engineering necessary to operate effectively the FEBR for toluene contaminated air treatment. During a 144 h batch operation without addition of external nutrients or cells, toluene removal efficiency dropped from 90% to 64%, while a 30% decrease of the biomass concentration and a 88% loss of the INT reduction activity were experienced. It was concluded that batch operation of the liquid could not support stable and high toluene removal efficiency over time. Continuous feeding of either active cells or of mineral medium was investigated. The most efficient and practical operating mode was achieved when 20% of the culture was replaced daily by a concentrated nutrient solution. In doing so, significant cell growth was obtained and nutrient limitation was avoided. A toluene removal of 89%–94% and a high cell activity measured as INT reduction activity could be maintained over 360 h. Under these conditions, 27% of carbon-toluene degraded was incorporated into biomass and the rest was mineralized to carbon dioxide. The effect of toluene inlet concentration and toluene starvation were also investigated. The results showed that the FEBR was flexible in adapting to other conditions or in recovering from abnormal operation. Throughout the experiments, monitoring of the cell activity measured as INT reduction was useful in providing insight into the process. INT reduction activity measurements proved that under normal operating conditions, the FEBR was subject to both a kinetic and a mass transfer limitations. Overall, the results of this study indicate that the FEBR might be an interesting alternative to biofilters and biotrickling filters, if high performance is required.

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