

# Transient Performance of a Two-Phase Partitioning Bioscrubber Treating a Benzene-Contaminated Gas Stream

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The dynamic performance of a prototype, two-phase partitioning bioscrubber in response to fluctuating benzene waste gas feeds has been characterized through a series of spike, step, and shutdown–restart experiments. From stable operation at a nominal loading capacity of  $62 \pm 6 \text{ g}/(\text{m}^3 \text{ h})$  and removal efficiencies of over 99%, the bioscrubber was subjected to influent benzene concentration step changes and spikes as high as 10- and 20-fold, respectively. The bioscrubber responded rapidly and effectively to all feed concentration spikes and steps, as well as to step changes in the feed flow rate, maintaining nearly undisturbed performance. Although benzene absorption by the two liquid phases was found to dominate the early stages of each transient, *Achromobacter xylosoxidans* Y234 responded quickly to prolonged disturbances, readily consuming most of the excess absorbed substrate. The results demonstrate that the two-phase partitioning bioscrubber can rapidly acclimate to and recover from fluctuations, ensuring that stable performance can be maintained both during and after transients. The presence of *n*-hexadecane promotes benzene capture in the two-phase system by increasing the absorption driving force, an important characteristic during high intermittent loadings. Rapid recovery from two different shutdown scenarios further demonstrates the practical potential of the two-phase partitioning bioscrubber as a high-performance biotechnology alternative for the treatment of toxic waste gases.

## Introduction

Of the numerous different biologically based processes presented throughout the literature for the treatment of waste gases contaminated with volatile organic compounds (VOCs), arguably the most popular remains biofiltration. Biofilters are attractive because their uncomplicated and inexpensive design is usually coupled with long-term performance requiring minimal maintenance and operator attention (1). As such, biofilters continue to fill an important niche for the treatment of VOCs. However, because of poor mass transfer and deactivation due to inhibition by toxic substrates such as BTEX (benzene, toluene, ethylbenzene, and xylene) compounds, their application has typically been limited to the treatment of low-concentration waste gases (2).

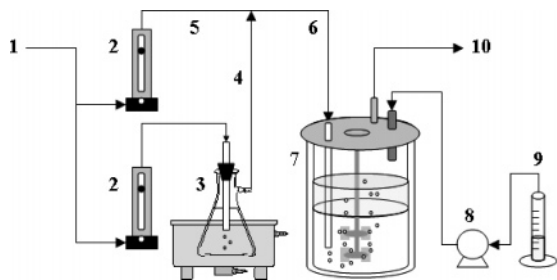
The two-phase partitioning bioscrubber (TPPB) was originally developed to address some of the inherent limitations of biofiltration through a design that could provide protection from high concentrations of toxic compounds, as well as enhanced rates of VOC absorption (3, 4). On the basis of the concept of two-phase partitioning bioreactors, the bioscrubber includes a cell-containing aqueous medium and a second, often immiscible organic phase which functions as a repository for high concentrations of toxic substrates, controlling the aqueous-phase concentrations at subinhibitory levels (5). Substrate will partition through mass transfer equilibrium to the aqueous phase according to the metabolic requirements of the cells (6). A protocol for selecting an appropriate, organic second phase, originally laid out by Collins and Daugulis (7), was subsequently followed by Yeom and Daugulis (3), who selected *n*-hexadecane as the second phase in their original TPPB design to treat benzene waste gases with *Achromobacter xylosoxidans* Y234 (formerly *Alcaligenes xylosoxidans* Y234). Absorption mass transfer into a TPPB is enhanced, first as a result of mixing that allows for greater gas–liquid contact. In addition, and in a manner analogous to the ability of a two-phase partitioning bioreactor to improve the bioavailability of poorly water soluble substrates (5), a TPPB can increase the absorption of VOCs which have high Henry's law coefficients in water, such as benzene. By selecting a second phase with a Henry's law coefficient for the volatile substrate that is lower than for water, the affinity for the compound in the two-liquid system is enhanced, resulting in greater driving forces for absorption.

To be considered for practical use, the dynamic stability of any biological air treatment strategy must be confirmed because, as noted by Zarook et al. (8), transient fluctuations will be encountered more routinely than constant conditions in most applications. Typical dynamic variations in the feed streams can include fluctuations in concentration, flow rate, and composition. Transient conditions become of particular concern when the treated compounds are toxic in nature, as fluctuations can lead to increasing substrate concentrations which may surpass the level that is inhibitory to the biocatalyst, inducing performance instabilities that may lead to process failure and emissions to the environment.

Since their inception, TPPBs have been used in various designs to treat single substrate feeds of benzene (3, 4, 9, 10), toluene (11, 12), and ethylbenzene (10), as well as a mixed feed of benzene and toluene (11). The most recent research focused on demonstrating steady-state operation with a culture whose maintenance energy requirements consumed the entire substrate supply (10). All of these previous studies demonstrated both high removal efficiencies and elimination capacities of the respective compounds. However, with the exception of the study of Nielsen et al. (10), in which a single set point change in the benzene loading rate was imposed, only constant feed streams have been studied.

This study experimentally characterizes the dynamic response of a TPPB to a transient benzene feed, focusing on fluctuations in concentration and flow rate, in the form of both spikes and step changes. The response to different modes of shutdown and restart have also been investigated, as this represents a type of dynamic variation and a practical aspect of operation that will be important in most applications. Biofiltration will serve as a benchmark for comparison since it has been so widely studied, particularly under transient conditions.

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**FIGURE 1.** Experimental arrangement, including (1) sterile supply air, (2) air flow rotameter, (3) sparged benzene flask in 30 °C water bath, (4) enriched benzene gas stream, (5) makeup air stream, (6) combined feed gas stream, (7) two-phase partitioning bioscrubber, (8) base pump, (9) 6 M KOH, and (10) off-gas stream.

## Materials and Methods

**Microorganism and Media.** *A. xylosoxidans* Y234 was used in this work to degrade benzene, in previous successful studies (3, 4, 10). The enrichment medium formulation, inoculum preparation procedure, and details regarding all nutrients and chemicals used have all been previously reported (10).

### Bioscrubber Arrangement and Sampling Procedures.

The experimental arrangement used for all of the dynamic TPPB studies described in this paper is depicted in Figure 1. The bioscrubber consisted of a 5 L New Brunswick BioFlo III bioreactor containing 2 L of aqueous medium and 1 L of *n*-hexadecane, representing an organic fraction of 33% (volume). The aqueous medium was twice as concentrated as the enrichment medium used (10) but contained no carbon or energy source. Although considered conceptually as two distinct liquid phases as shown in Figure 1, the TPPB was agitated with two Rushton turbines and always operated as a well-mixed dispersion to ensure high mass transfer rates of both oxygen and benzene. Operating conditions that were maintained included pH 6.6 (controlled automatically by adding 6 M KOH), 30 °C, and agitation at 800 rpm. The nominal operating point from which fluctuations were imposed included aeration at a combined feed gas flow rate of 60 L/h (enriched benzene gas flow plus makeup aeration). These operating conditions produced a gas holdup volume of approximately 25% of the total liquid volume. The average benzene concentration in the combined feed gas stream was  $3.1 \pm 0.3 \text{ g/m}^3$  (as determined by gas chromatography). After normalization by the total liquid volume of the bioscrubber, these nominal settings constitute an average loading capacity of  $62 \pm 6 \text{ g/(m}^3 \text{ h)}$ .

After inoculation, the benzene loading rate was set to and held constant at its nominal value to allow the biomass, which was retained indefinitely, to accumulate and gradually approach a characteristic steady-state concentration which arises in response to cellular maintenance energy, as demonstrated by Nielsen et al. (10). Throughout the experiments, feed and off gases were concurrently sampled for benzene content via GC/FID as previously described (10). Periodically (every 12–24 h), 10 mL liquid samples of the reactor contents were drawn into 15 mL centrifuge tubes prior to phase separation by centrifugation (15 min at 4 °C and 3400 rpm). Benzene in *n*-hexadecane was measured by GC/FID (10). Aqueous benzene levels were estimated using an experimentally determined equilibrium partitioning coefficient for benzene between *n*-hexadecane and the aqueous medium. Dissolved oxygen levels were measured via a polarographic-membrane electrode (Broadley and James Corp.) and automatically recorded by a computer interface. Biomass concentrations in the aqueous phase were determined by optical density measurements with a Biochrom Ultraspec 3000 UV/vis spectrophotometer (Biochrom, Ltd.,

U.K.) at 650 nm using an external cell dry weight (CDW) calibration. Each biomass measurement was performed in duplicate. Culture purity was routinely analyzed throughout the experiments using phase microscopy, as well as by culturing samples of the bioscrubber contents for 24 h at 30 °C on solid agar plates. Water losses incurred by stripping into the passing air stream or by sampling were largely compensated for by base addition for pH control; however, fresh sterile water (approximately 150 mL) was added to the bioscrubber every 5–7 days to maintain the original liquid level.

**Dynamic Fluctuations.** Perturbations were imposed in the form of both spikes and steps after the TPPB had reached its steady-state biomass concentration. The 5 min long spikes and 4 h long steps in the feed rate were produced by increasing the enriched benzene feed stream flow rate while maintaining the total aeration rate constant at its nominal value of 60 L/h. Spikes in the benzene feed concentration of approximately 4, 8, and 20 times the nominal value were introduced, while step changes of approximately 2, 4, and 10 times the nominal value were studied. The response of the system to a 2-fold step change in the loading rate was also examined at a constant benzene feed concentration by doubling the total flow rate to 120 L/h from its nominal value.

Two modes of shutdown and subsequent restart were investigated. The first involved stoppage of the benzene feed from its nominal value for 24 h while maintaining makeup aeration at 60 L/h. The second halted all air flow to the bioscrubber for 24 h. Agitation was performed throughout both shutdown experiments, and the system was monitored for nearly 24 h after restart to investigate stability. Dynamic experiments were performed in the following order: spike, step changes (constant flow rate), step changes (constant concentration), and shutdowns. However, the different magnitudes of each type of feed dynamic were performed in randomized order. Biomass concentrations were measured prior to each dynamic experiment to verify that no major accumulation or depletion (beyond the error associated with the measurement) had occurred and that the values were within 1 standard deviation of the characteristic experimental steady-state value.

**Gas–Liquid Equilibrium.** Henry's law coefficients for the distribution of benzene between the gas and each of the aqueous (same composition as used in the bioscrubber) and organic (*n*-hexadecane) phases were estimated at 30 °C to aid in analysis. Known volumes of liquid benzene were injected into 125 mL amber serum bottles containing 45 mL of aqueous medium or *n*-hexadecane and capped with Teflon-lined septa. After 1 h of equilibration at 30 °C, headspace samples were analyzed for benzene content via gas chromatography (10). Equilibrated gas-phase concentrations in the range of 1–20 g/m<sup>3</sup> were tested for each liquid phase. Liquid-phase benzene concentrations were calculated by material balance and, in the case of *n*-hexadecane, were also verified by gas chromatography. Linear least-squares regression of the equilibrated liquid- and gas-phase concentration data provided estimates of the Henry's law constants for each phase, assuming ideal gas behavior. The data remained highly correlated throughout the range of concentrations studied (not shown). The Henry's law constants of the aqueous and organic phases were determined to be  $0.33 \pm 0.01 \text{ (g/m}^3\text{)}_{\text{gas}}/\text{(g/m}^3\text{)}_{\text{aqueous}}$  and  $0.0020 \pm 0.0001 \text{ (g/m}^3\text{)}_{\text{gas}}/\text{(g/m}^3\text{)}_{\text{organic}}$  at 30 °C, respectively.

**Quantifying Performance.** The performance of the bioscrubber unit was assessed by instantaneous measurement of the apparent elimination capacity (EC)

$$EC = \frac{Q_g(S_g^{\text{in}} - S_g^{\text{out}})}{V_{\text{total}}} \quad (1)$$

and the apparent removal efficiency (RE)

$$RE = \frac{Q_g(S_g^{\text{in}} - S_g^{\text{out}})}{Q_g S_g^{\text{in}}} \times 100\% \quad (2)$$

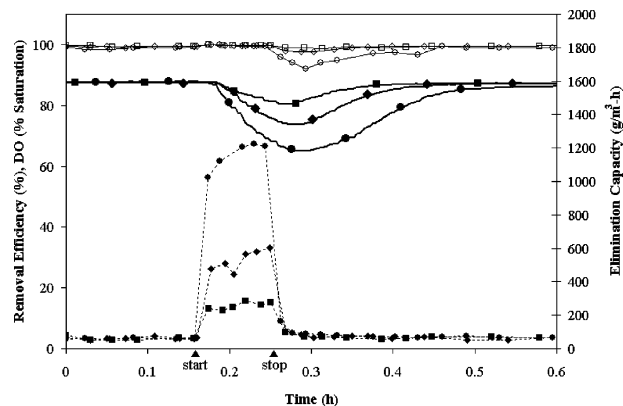
where  $Q_g$  represents the volumetric flow rate of the waste gas stream,  $V_{\text{total}}$  is the total volume of the bioscrubber liquid contents, and  $S_g^{\text{in}}$  and  $S_g^{\text{out}}$  are the gaseous benzene concentrations measured at the inlet and outlet of the bioscrubber, respectively.

## Results

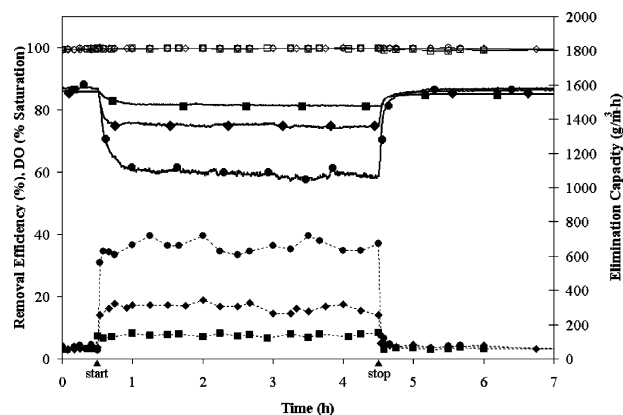
During the preliminary stage of the experiment, a steady-state biomass concentration of  $5.8 \pm 0.2$  g (CDW)/L was attained after approximately 200 h of operation. An average apparent elimination capacity of  $61.6 \pm 6.4$  g/(m<sup>3</sup> h) (average of all instantaneous measurements) was achieved throughout, representing an average removal efficiency of over 99%. Dissolved oxygen (DO) levels were maintained relatively constant at  $89.4 \pm 1.4\%$  of saturation, well above the limiting conditions. This steady state served as the condition from which all dynamic fluctuations were imposed.

Dynamic experiments were initiated from the steady state with a series of 5 min spikes of increased benzene concentration in the combined feed gas stream. Figure 2 shows that, as the magnitude of the spike was increased from 4 to 8 and to 20 times the nominal load, the process was able to operate with no change in removal efficiency during the initial period after the spike was introduced. However, after the spike was completed the removal efficiency dropped temporarily, with the magnitude of the decrease reflecting the spike size. By applying a higher loading, even for just 5 min, the rate of benzene absorption increases, leading to greater benzene retention in the liquid phases. The elevated benzene content in the system promotes increased bioactivity and, thus, higher consumption rates of dissolved oxygen, as seen in Figure 2. Larger spikes resulted in greater dissolved oxygen consumption rates. When the loading rate was returned to its nominal value after the spike, the excess absorbed benzene began to strip from the liquid phases, resulting in higher off-gas benzene concentrations and a decrease in the measured instantaneous removal efficiency. The most dramatic decrease in removal efficiency resulted after the 20-fold spike; however, it did not decrease below 92%, corresponding to a maximum emitted off-gas benzene concentration of only 0.4 g/m<sup>3</sup>. This off-gas concentration, assumed to be in equilibrium with the liquid phases of the bioscrubber, would correspond to an aqueous-phase benzene concentration of approximately 1.2 g/m<sup>3</sup>, well below the inhibitory threshold of this organism (unpublished data). As the magnitude of the spikes was increased, greater recovery times were required for the process to return to stable performance since more time was required to strip and/or consume the greater quantities of benzene absorbed during the spike. Furthermore, since the aqueous-phase benzene concentrations remained low throughout the dynamic fluctuations, the observed recovery period is likely not related to any sort of cellular deactivation, but is due strictly to the physical phenomena discussed above.

A series of step increases to the benzene concentration in the combined feed gas stream of 2, 4, and 10 times the nominal value were also introduced to the TPPB from the nominal steady state. Monitoring was conducted throughout the 4 h step input as well as for an additional 3 h after returning to the nominal conditions to confirm the stability of the process. As seen from Figure 3, the TPPB handles each of the step change inputs well, adapting to the temporary load changes to achieve average elimination capacities of up to 650 g/(m<sup>3</sup> h) while continuing to maintain removal efficien-



**FIGURE 2.** Dynamic response of the two-phase partitioning bioscrubber to 5 min feed stream benzene spikes of 4-fold (squares), 8-fold (tilted squares), and 20-fold (circles) concentration increases from the nominal loading capacity ( $3.1 \pm 0.3$  g/m<sup>3</sup>,  $62 \pm 6$  g/(m<sup>3</sup> h)). The removal efficiency (solid line, hollow symbols), dissolved oxygen (solid line, solid symbols), and elimination capacity (dashed line, solid symbols) are also shown.

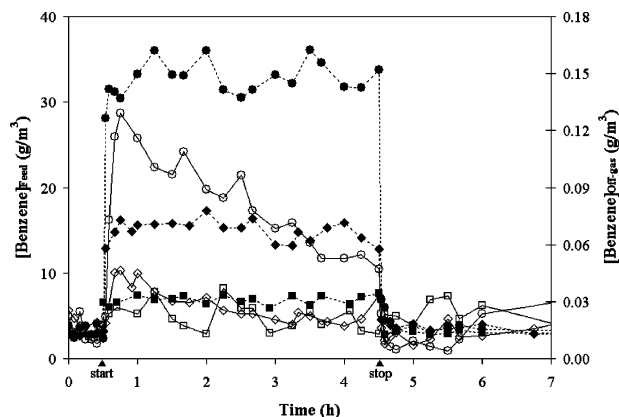


**FIGURE 3.** Dynamic response of the two-phase partitioning bioscrubber to 4 h feed stream benzene step changes of 2-fold (squares), 4-fold (tilted squares), and 10-fold (circles) concentration increases from the nominal loading capacity ( $3.1 \pm 0.3$  g/m<sup>3</sup>,  $62 \pm 6$  g/(m<sup>3</sup> h)). The removal efficiency (solid line, hollow symbols), dissolved oxygen (solid line, solid symbols), and elimination capacity (dashed line, solid symbols) are also shown.

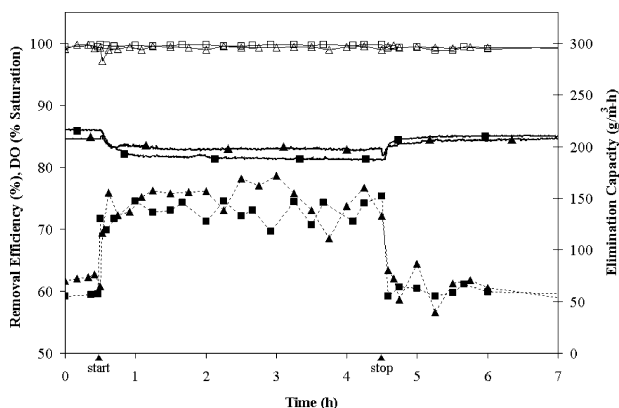
cies above 99% throughout. Periods of reduced performance, observed after each of the spike experiments due to stripping of absorbed benzene, were not observed after any of the step changes returned to the nominal conditions. This is attributed to the ability of the cells to rapidly adapt to the increased benzene load over the course of the longer (4 h) dynamic period. Biomass concentrations measured immediately after the 4 h dynamic period increased from their steady-state concentration by 6.5%, 8%, and 11% after the 2-, 4-, and 10-fold step increases in feed concentration, respectively. Biomass growth occurred during the step changes because benzene was supplied in excess of the requirements for cellular maintenance energy alone. As biomass increased, so too did the total utilization rate of benzene, which led to a gradual decrease in absorbed benzene levels and, as seen in Figure 4 (most dramatically with the 10-fold step change), a steady decline in off-gas benzene concentrations.

A step change in loading capacity was also introduced at constant feed gas benzene concentration by changing the total feed gas flow rate. A 2-fold step increase in the feed gas flow rate from 60 to 120 L/h was imposed for 4 h while the average benzene feed gas concentration was maintained constant at its nominal value of  $3.1 \pm 0.3$  g/m<sup>3</sup>. The results are shown in Figure 5 and compared with the results of a





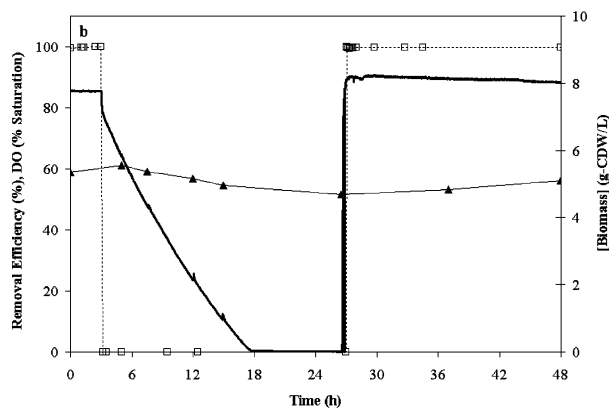
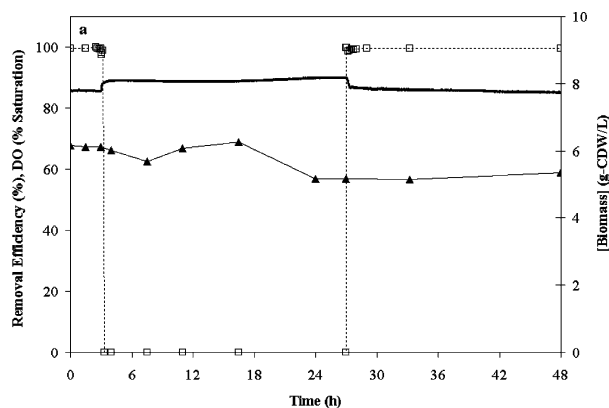
**FIGURE 4.** Feed (dashed lines, solid symbols) and off-gas (solid lines, hollow symbols) benzene concentrations during 2-fold (squares), 4-fold (tilted squares), and 10-fold (circles) step changes in concentration for 4 h from the nominal feed ( $3.1 \pm 0.3 \text{ g/m}^3$ ).



**FIGURE 5.** Dynamic response of the two-phase partitioning bioscrubber to 2-fold, 4 h benzene loading rate step changes due to changes in the concentration (squares) and flow rate (triangles) from the nominal loading capacity ( $3.1 \pm 0.3 \text{ g/m}^3$ ,  $62 \pm 6 \text{ g/(m}^3 \text{ h)}$ ) while monitoring the removal efficiency (solid line, hollow symbols), dissolved oxygen (solid line, solid symbols), and elimination capacity (dashed line, solid symbols).

2-fold step change in loading caused by fluctuation of the feed gas concentration at a constant feed gas flow rate. Similar performance is observed for both 2-fold step changes in terms of elimination capacities and removal efficiencies. Dissolved oxygen levels remain higher through the dynamic period when the total flow rate is changed because this also represents an increase in aeration. At the onset of the step change when the total flow rate is perturbed, the removal efficiency briefly plunges to 97% as higher rates of stripping from the bioscrubber are experienced due to the higher air flow. The process quickly adapts to this change, however, and removal efficiencies of over 99% are then maintained throughout the duration of the experiment. Both the 4-fold spike and the 10-fold step change were repeated to investigate reproducibility, and nearly identical results were obtained (data not shown).

The two modes of shutdown–restart investigated involved stoppage of the enriched benzene gas stream while maintaining the makeup air stream at the nominal combined feed gas flow rate of 60 L/h and complete cessation of the combined feed gas stream (air and benzene). The results of these two shutdown experiments are presented in parts a and b of Figure 6, respectively. Restart posed no operational problem after either shutdown, with no reacclimation period required since the bioscrubber promptly recovered to its original high performance after the feed was reinstated to its



**FIGURE 6.** Shutdown performed from nominal conditions ( $3.1 \pm 0.3 \text{ g/m}^3$ ,  $62 \pm 6 \text{ g/(m}^3 \text{ h)}$ ) by (a) halting the enriched benzene gas stream and maintaining the makeup air stream at 60 L/h for 24 h and (b) halting all gas flow for 24 h while monitoring the removal efficiency (squares), dissolved oxygen (solid line), and biomass concentration (triangles).

nominal conditions. During the first shutdown, dissolved oxygen levels increased from an average value of 86% saturation, maintained under nominal conditions, to an average of 90% saturation (Figure 6a). In the absence of the benzene feed, the offset in dissolved oxygen from 100% saturation is associated with the dissolved oxygen requirements for the endogenous activities of the cells. During the second shutdown, dissolved oxygen levels first dropped abruptly as all residual absorbed benzene was quickly consumed and then proceeded to decline gradually over the following 15 h (Figure 6b). Again, the constant rate at which dissolved oxygen was consumed in the bioscrubber is thought to be characteristic of the culture's endogenous requirements during total starvation. Even after a 9 h period of total dissolved oxygen limitation experienced during the second shutdown (Figure 6b), the bioscrubber immediately reestablished its original performance at high removal efficiency. During each of the shutdowns, biomass levels declined gradually from their steady-state concentration as the cells survived by endogenously respiring their own storage materials. In each case, this led to a decrease in biomass of approximately 14% by the time of restart. However, after the benzene feed was reinstated to its nominal condition, the biomass began to recover, approaching the characteristic steady-state level of  $5.8 \pm 0.2 \text{ g (CDW)/L}$  over the course of 48 h after the feed was reinitiated.

## Discussion

In biofiltration studies, feed spikes lasting between 30 min (13) and 120 min (14) are typically found, with concentrations changing between 4-fold (15) and 15-fold (13) of the nominal load. Step changes usually range between 2 and 4 times the

nominal loading rate and last up to 5 h (16); however, a few studies have maintained steps for as long as 100 h (17, 18), and even 150 h (19). While the magnitude of feed perturbations imposed in this study is similar to those of past biofiltration studies, the dynamic periods of 5 min spikes and 4 h steps investigated here are much shorter. Nevertheless, the range of transient conditions encountered in industrial processes will likely include fluctuations with durations of several hours, as well as those lasting only minutes. Furthermore, the experimental results demonstrate that, when subjected to longer dynamic periods, the bioscrubber becomes acclimatized to the new conditions by using the excess substrate to produce more cells. In this manner, the response of the TPPB to dynamic conditions resembles that of a chemostat. The biomass acclimatization that occurred during the 4 h step change experiments eliminated the periods of poor performance that appeared after the shorter, 5 min spikes as a result of stripping of absorbed benzene. Acclimation could not occur during the feed spikes as they were not sustained long enough for the cells to respond. Had the dynamic inputs been prolonged indefinitely, in the form of a set point change in loading, for example, the bioscrubber would have eventually achieved a new, characteristic steady state, as previously demonstrated by Nielsen et al. (10).

Only a few of the numerous transient biofilter studies in the literature have focused solely on the treatment of benzene. Therefore, studies involving the dynamic biofiltration of other BTEX compounds will also be used for comparison, considering the compounds as structural and physical analogues. In addition, transient operation involving other substrates considered to be particularly toxic will be considered qualitatively, where appropriate. Since feed disturbances shorter than 1 h are not usually reported in biofiltration studies, it is difficult to compare the response characteristics of biofilters to loading spikes of the brief duration (5 min) performed here. Feed spikes lasting 1 h or less have, however, been studied for the biofiltration of mixtures containing BTEX compounds using toluene and xylene (20) and MEK and toluene (15). Metris et al. (20) performed a series of five spikes of 5–6 times the nominal loading, each lasting approximately 30 min with a 30 min recovery time in between. Instantaneous removal efficiencies worsened with each spike, dropping to as low as 50% by the last spike. This suggests that the 30 min recovery period allowed was insufficient for the biofilter to regain its steady-state performance. Feed concentration spikes of roughly 5 times their normal value and lasting 1 h were performed by Atoche and Moe (15). They also observed stripping of a portion of the sorbed VOC after the spike input ended and the feed returned to its baseline value. However, in their case the effects of this phenomenon were much more dramatic, resulting in instantaneous removal efficiencies which fell below 50%. For toluene, full recovery to normal performance required 25 min after the 1 h spike, while MEK required approximately 75 min. In our TPPB study, benzene feed spikes of 4–8-fold had little effect on effluent concentrations and removal efficiencies remained high. Even after the 5 min feed concentration spike of 20 times the normal load, instantaneous removal efficiencies did not drop below 92% and full performance was achieved after approximately 15 min.

Although recovery from step inputs in toluene loading was not investigated by Rene et al. (21), their study did subject a compost biofilter to a consecutive series of short step changes. Their biofilter performance was found to be quite sensitive to influent toluene concentrations, the removal efficiency dropping rapidly from 92% to 63% and then to 50% as the feed concentration changed from 0.09 to 1.4 and finally to 2.3 g/m<sup>3</sup>. Performance remained constant and low during each step (which lasted 5–10 days), indicating that

no significant acclimatization was taking place prior to the next dynamic change. Since the relative magnitude of these steps was quite large, it was concluded in that study that substrate inhibition was the likely cause of the poor performance. Substrate inhibition of *A. xylosoxidans* Y234 was not observed in our studies as low aqueous benzene concentrations were maintained through partitioning into the organic phase. Tang et al. (19) investigated toluene biofiltration with step inputs in both the feed concentration and flow rate that were sustained for periods of up to 8 days. Although their step changes lasted substantially longer than those performed here, these extended periods were necessary as acclimatization of the biofilter did not occur until 2–3 days after the onset of the fluctuation. This slow acclimatization was preceded by a period where sorption by the filter media was predominantly responsible for toluene removal from the gas stream. However, even after 8 days was allowed for acclimation, following the transient period when the feed concentrations returned to normal, significant amounts of toluene which had remained sorbed to the media began to strip and resulted in drastically low instantaneous removal efficiencies and elimination capacities. In response to sustained loading changes, biofilters have typically been found to require an intermediate period of bed acclimation of roughly 2 days for “moderate” increases and up to 5 days for “large shock loads” (2). On the other hand, the culture in the TPPB was able to adapt to each of the loading changes studied within just 4 h, ensuring that absorbed benzene concentrations in the TPPB were low and that no excessive stripping would occur after the transient period ended.

Flow rate changes at a constant feed concentration represent an additional mode by which operating conditions and loading rates can fluctuate. Since flow increases lead to decreased residence times, the mass transfer limitations associated with high flow rates often result in poor performance in biofilters, making the flow rate one of the most important process variables affecting biofilter performance (18). When Tang et al. (19) introduced an 11 day step input in flow from 0.37 to 1.12 cm/s in a feed stream containing 0.85 g/m<sup>3</sup> toluene, the elimination capacity of their chaff/compost biofilter dropped from 33 to 20 g/(m<sup>3</sup> h) and the removal efficiency fell to 60% from nearly 100%. Although our study investigated only 4 h periods in which the feed air flow rate was doubled, the unwavering elimination capacity and removal efficiencies achieved suggest that the performance is quite stable, though additional, longer and larger step inputs would help support this notion. The TPPB is less susceptible to flow rate changes because its enhanced mass transfer characteristics and well-mixed design translate into highly effective absorption, thus requiring much shorter residence times.

The first of the two shutdown experiments was designed to simulate a situation in which an upstream benzene-emitting process was halted for 24 h while the waste gas collection system (which feeds the TPPB) remained in operation. The second shutdown experiment, however, simulated a complete shutdown of the waste gas collection system. Both of these different modes of starvation have also been simulated in previous biofiltration studies (22, 23). In each case, the TPPB experienced no loss of performance after restart, responding almost immediately with no observable reacclimation periods required. Several biofiltration studies have also reported equivalently rapid recoveries after brief starvation periods (1–2 days) in treatment of hydrogen sulfide (17), ammonia (22), and a mixture of VOCs (24), while longer starvation periods, in excess of 7 days, typically required several days for full recovery to preshutdown performance (19, 22, 23). In general, the length of time required for biofilter reacclimation after a shutdown period is proportional to the duration of the shutdown (25). Wani

et al. (23) also found that reacclimation was made easier when aeration was continued through the biofilter while only the substrate feed was halted. From Figure 6, no differences in reacclimation could be deduced between the two 24 h shutdown modes using the TPPB. Shutdowns or starvation periods imposed in biofilter studies have typically been longer than the 24 h period selected here, ranging from 2 to 7 days (22), and even as long as 3 months (23). Therefore, extended periods of starvation imposed during the shutdown of the TPPB should be studied to ensure that its resilience can at least match that of biofiltration.

It has been argued that oxygen transfer is unlikely to become limiting during the biofiltration of loads below 100 g of carbon/(m<sup>3</sup> h) (26), which would translate into 108 g/(m<sup>3</sup> h) benzene. As such, oxygen is often assumed to be available in abundant supplies and the importance of its presence is overlooked. However, diffusional limitations have been predicted to affect the performance of a toluene-degrading biofilter (8). The TPPB never became oxygen-limited in response to the nominal loading rate or the imposed feed dynamics, even when interim elimination capacities of over 1200 g/(m<sup>3</sup> h) during spikes and up to 650 g/(m<sup>3</sup> h) during step inputs were achieved. As additional substrate becomes available during transient loads, the adapting cells rapidly respond by consuming additional oxygen. As the magnitude of the perturbations increased, correspondingly greater decreases in the dissolved oxygen levels were observed. In this study, rates of oxygen absorption remained sufficiently high in the well-mixed system such that limitation did not occur. Although dissolved oxygen levels never reached critically low levels under the conditions examined, the possibility of such limitations (and the poor performance that would result) in response to even larger fluctuations is a valid concern and will be further studied experimentally, as well as with the aid of a mathematical model which is currently being developed. Despite being subjected to completely depleted dissolved oxygen levels for a 9 h period during the second shutdown, the rapid recovery of the bioscrubber after restart to its previous performance indicates that the culture was not negatively impacted as a result.

The stability of a dynamic process in response to transient conditions depends in part upon where the system is operating in relation to the performance boundaries which are established by physical limitations. Although previous TPPB studies have reported benzene elimination capacities much higher than  $61.6 \pm 6.4$  g/(m<sup>3</sup> h) reported here (4, 10), the performance achieved in this study is still quite high. The ability of the system to readily respond to substantial perturbations provides direct evidence that limitations of this process are far from being reached.

Toxicity aside, it is the relatively large Henry's law coefficient of benzene in water that represents an additional design limitation that has routinely hampered the performance of many biological waste gas treatment strategies, particularly biofilters (27). Reported for water as  $0.27$  (g/m<sup>3</sup>)<sub>gas</sub>/(g/m<sup>3</sup>)<sub>water</sub> at 30 °C by Ashworth et al. (28), the presence of salts in an aqueous bioreactor medium further reduces benzene solubility and leads to an even higher Henry's law constant, measured experimentally at 30 °C for our medium as  $0.33 \pm 0.01$  (g/m<sup>3</sup>)<sub>gas</sub>/(g/m<sup>3</sup>)<sub>aqueous</sub>. As such, the mass transfer driving force for benzene absorption from the gas phase into either water or aqueous medium is quite low. On the other hand, the much lower benzene Henry's law coefficient of *n*-hexadecane, experimentally determined as  $0.0020 \pm 0.0001$  (g/m<sup>3</sup>)<sub>gas</sub>/(g/m<sup>3</sup>)<sub>organic</sub> at 30 °C, translates into a mass transfer driving force for benzene absorption into *n*-hexadecane that is 165 times that of the aqueous medium. Therefore, the addition of *n*-hexadecane to the bioscrubber not only serves to partition toxic concentrations of benzene away from the cells but also greatly enhances benzene

absorption. Similar reasoning by van Groenestijn and Lake (29) led to the inclusion of silicone oil as a second phase in their biotrickling filter design to promote absorption and achieve higher elimination capacities of hexane, also a VOC with a high aqueous Henry's law coefficient.

The improved mass transfer characteristics arising from organic-phase addition are perhaps most important during dynamic fluctuations where substrate loading rates may temporarily exceed the maximum elimination capacity of the organism. The organic phase ensures that, although some substrate accumulation may occur under transient conditions, high absorption rates will continue as the overall mass transfer driving force remains high because the organic phase is far from saturated. The organic phase serves as an "overflow reservoir" for high substrate concentrations, providing a buffering capacity for absorption during transient fluctuations. Of course, the organic phase still serves its original role, ensuring that subinhibitory aqueous-phase concentrations are maintained throughout the transient periods. The ideal organic-phase volume fraction with which to operate to consistently realize a target minimum removal efficiency, while assuaging toxic concentrations of benzene, will depend on the anticipated amplitude of the fluctuations. Optimizing this quantity will translate into material cost savings and will be the focus of future work.

Previous studies have provided direct evidence that sorption will often limit the effectiveness of a biofilter during loading transients. After achieving varied success in the transient biofiltration of several VOCs, Deshusses (13) concluded that absorption processes are of primary importance during dynamic loading. Their conclusion was supported by CO<sub>2</sub> data which suggested that VOC consumption did not peak until 2–5 h after the transient period ended. Some biofilter designs have taken measures to infuse their filter bed with a greater sorption potential by incorporating granular activated carbon (GAC) into the media. Abumaizar et al. (30) demonstrated that increasing the content of GAC to as high as 13.3% (dry weight basis) in a compost biofilter also helped to mitigate the impact of shock loadings of BTEX waste gases, and that the positive effect was directly proportional to the amount of GAC present. They similarly concluded that the GAC serves as a buffer in a biofilter, initially adsorbing much of the excess substrate from a feed disturbance, and then subsequently supplying it to the adapting culture at more acceptable rates.

Although it is the biocatalyst that ultimately destroys target pollutants, the primary function of any biological waste gas treatment technology is to remove VOCs from the contaminated air stream. Be it through adsorption or absorption, the effective removal of VOCs from the waste gas dictates such performance criteria as elimination capacities and removal efficiencies and guarantees regulatory compliance. That being said, a highly active biocatalyst will minimize substrate accumulation in the media, resulting in maximal driving forces for mass transfer. Therefore, bioreactors should be designed both to provide sufficient mass transfer and to establish an environment in which bioactivity can be maintained under all conditions. Satisfying both of these objectives has allowed the TPPB to achieve success during both steady and transient operation.

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