Treatment of Dynamic Mixture of n-Hexane, Benzene, and Methanol and Fungi Community Characterization in an Integrated Scheme of Cyclic Adsorption/Desorption Beds and Trickle Bed Air Biofilter

Authors: Zehraoui, Abderrahman, and Sorial, George A.

Source: Air, Soil and Water Research, 8(1)

Published By: SAGE Publishing

URL: https://doi.org/10.1177/ASWR.S23688

Treatment of Dynamic Mixture of *n*-Hexane, Benzene, and Methanol and Fungi Community Characterization in an Integrated Scheme of Cyclic Adsorption/Desorption Beds and Trickle Bed Air Biofilter

Abderrahman Zehraoui and George A. Sorial

Department of Biomedical, Chemical, and Environmental Engineering, College of Engineering and Applied Science, University of Cincinnati, Cincinnati, OH, USA.

ABSTRACT: This study investigates the effect of volatile organic compound (VOC) feed fluctuations on trickle bed air biofilters (TBABs) and the ability of a two-bed adsorption/desorption unit in dampening fluctuations. A mixture of *n*-hexane, benzene, and methanol with concentration ratios of 1:3:6.6 was fed to two parallel TBABs. To simulate feed fluctuations, four different square waves were applied. The total VOC loading rates (LRs) varied from 28.4 to 107.3 g/m^3 h. The average concentration of VOCs applied to both TBABs was within allowable limits as determined in an earlier study. One TBAB was preceded by a two-bed cyclic adsorption/desorption unit (integrated unit), while the other TBAB (control unit) was directly subjected to the high and low peaks. *n*-Hexane elimination in the integrated unit was steady, and stable performance was obtained (75%–89%) based on the LRs, whereas the control unit showed erratic performance. The other two VOCs were mostly removed.

KEYWORDS: adsorption, aerobic processes, biodegradation, biofilters, filamentus fungi, volatile organic chemicals

CITATION: Zehraoui and Sorial. Treatment of Dynamic Mixture of *n*-Hexane, Benzene, and Methanol and Fungi Community Characterization in an Integrated Scheme of Cyclic Adsorption/Desorption Beds and Trickle Bed Air Biofilter. *Air, Soil and Water Research* 2015:8 31–41 doi:10.4137/ASWR.S23688.

RECEIVED: January 14, 2015. **RESUBMITTED:** June 2, 2015. **ACCEPTED FOR PUBLICATION:** June 4, 2015.

ACADEMIC EDITOR: Carlos Alberto Martinez-Huitle, Editor in Chief

TYPE: Original Research

FUNDING: The authors acknowledge financial support from the National Science Foundation under award No. CBET 0852803. The finding and conclusions expressed in this publication are solely those of the authors and do not necessary reflect the views of the Foundation. The authors confirm that the funder had no influence over the study design, content of the article, or selection of this journal.

COMPETING INTERESTS: Authors disclose no potential conflicts of interest.

CORRESPONDENCE: george.sorial@uc.edu

Paper subject to independent expert blind peer review by minimum of two reviewers. All editorial decisions made by independent academic editor. Upon submission manuscript was subject to anti-plagiarism scanning. Prior to publication all authors have given signed confirmation of agreement to article publication and compliance with all applicable ethical and legal requirements, including the accuracy of author and contributor information, disclosure of competing interests and funding sources, compliance with ethical requirements relating to human and animal study participants, and compliance with any copyright requirements of third parties. This journal is a member of the Committee on Publication Ethics (COPE).

Published by Libertas Academica. Learn more about this journal.

Introduction

Biofiltration has emerged as an attractive option for controlling volatile organic compound (VOC) emissions from industrial processes due to its cost effectiveness as well as its benign by-products.[1,2] Most interestingly, trickle bed air biofilters (TBABs), because of their flexible, optimal, and controllable operations, combined with their efficiency in treating different type of VOCs, are preferred compared to conventional biofilters.[2–4] One of the main challenges of biofiltration is the erratic contaminant loading rate (LR), as microorganisms are not flexible in handling variability in the feed. It should be noted that biofiltration of waste gas streams is best carried out at a steady contaminant load. However, most off-gas or treatment streams for VOCs that originate in industrial processes have variable flow rates and transient loading, which limit the handling efficiency of biological oxidation processes.[3,5,6] An action is, therefore, required to dampen emission fluctuations since increased regulatory scrutiny after the Clean Air Act Amendments (CAAAs) is imposed on emitting sources.[7]

In order to eliminate the fluctuations in contaminant concentrations, which often undesirably influence the effectiveness of bioreactors for waste gas treatment, complementary techniques have been tested. Manninen et al,[8] for instance, used "coupled systems" consisting of a bioreactor followed by a biofilter for combating a complex mixture of VOCs. More interesting is the use of a biofilter preceded by an activated-carbon bed for stabilizing VOC loads. During periods of high VOC loadings, the buffering bed will act as an adsorption unit to accumulate VOCs, while during periods of low VOCs loadings the bed will release the accumulated VOCs. In this manner, the activated carbon bed could dampen fluctuations in loading to the biofilter and help maintain its long-term consistent performance. Waber and Hartmans[9] used this system in buffering the fluctuations of the concentration of contaminants in waste gases so that a constant flow of contaminants was supplied to the bioreactor. The activated-carbon buffer unit minimized the fluctuation in toluene loading, thereby improving the overall removal efficiency (RE) of the combined system. In another study, Li and Moe[10] also employed a single activated-carbon bed for equalizing discontinuous loading (eg, 8-hour loading per day) of an acetone and toluene mixture.

These previous studies have endorsed the usefulness of an adsorption unit in reducing biofilter performance fluctuations during transient VOC loadings. However, restrictions increased in the application of this system to the desired contaminant concentration as well as the frequency and duration

of contaminant loading. In practice, for high contaminant loading fluctuations, the buffering capacity of single-bed adsorption will be quickly exhausted. Furthermore, a starvation period will exist within the biofilter during the initial start-up period due to adsorption until breakthrough.

To overcome the drawback of the single buffering unit, Kim et al[11] used a fixed two-bed adsorption system to flatten and reduce waste-gas fluctuations. At transient toluene loadings of up to 65.9 g/m^3 h, a biofilter with a precyclic adsorber/desorber unit successfully reduced the waste gas toluene concentration below 5 mg/m^3, while a stand-alone biofilter could not achieve a similar behavior. This novel system, subsequently, provided stable operation of a biofilter under adverse contaminant feeding conditions. For instance, Cai and Sorial[12] evaluated an adsorption and desorption unit for dampening load fluctuation of a mixture of toluene, styrene, methyl ethyl ketone (MEK), and methyl isobutyl ketone (MIBK). The integrated system achieved consistent 99% RE since the working loading did not exceed the critical loading of 34.0 g/m^3 h, regardless of the fluctuation in feeding conditions prior to the cyclic adsorption beds. In the same way Aly Hassan and Sorial[13] employed cyclic adsorption/desorption beds to stabilize the erratic loading of a mixture of *n*-hexane and benzene while the control unit could not withstand the fluctuations incurred.

In this regard, the aim of the current study was to evaluate the effectiveness of an integrated system consisting of a two-bed adsorption/desorption unit followed by a TBAB in treating a mixture of *n*-hexane, benzene, and methanol. These VOCs are hazardous air pollutants included in the 1990 CAAA.[7] Industries such as printing and publishing, polymer and man-made fiber, pulp and paper, and organic chemicals are major contributors of these VOCs, which also represent a combination of hydrophilic and hydrophobic compounds. Evaluations in this study were conducted under dynamic contaminant VOCs loadings and compared with a stand-alone biofilter system. Also, the evaluations were conducted under acidic conditions in the biofilter to favor the growth of a fungi consortium. Such environment has been found to be very effective for the chosen contaminants.[14] The evaluation of the buffering capacity of the cyclic two-bed adsorption unit as well as comparisons of its performance to the control unit, carbon mass balance closures, kinetic analysis, and characterization of microbial communities within the biofilters were performed.

Experimental Methods

The experimental work was performed on two lab-scale reactors for controlling a mixture of *n*-hexane, methanol, and benzene. *n*-Hexane and benzene were obtained from Fisher Scientific with 95% and 99% purity, respectively. Methanol was obtained from Tedia with 99.9% purity. The dimensionless Henry's law constant at 25°C is 40.7 ± 2.78 for *n*-hexane,[13] 2.28×10^{-1} for benzene,[15] and 1.9×10^{-4} for methanol.[16] One system consisted of the two-bed adsorption unit followed by a TBAB, named the integrated unit. The other system was a control unit in which a stand-alone TBAB was operated. A scheme of the experimental setup is illustrated in Figure S1 (Supplementary Material).

Adsorption unit. The system was designed to operate two adsorption beds in series. While one of the beds was adsorbing, the other bed was desorbing. The cycle was switched every four hours, and the desorbing bed started to adsorb, and vice versa. Figure S2 (Supplementary Material) shows a two-step cycle for the two-bed adsorption unit used in this study. The unit was adopted based on a previous study.[17] The empty bed retention time (EBRT) in the two-bed adsorption was designed to be nine seconds (at an air flow rate of 1.36 L/min) with the corresponding total volume of two cylindrical beds of 2.06×10^{-4} m^3. The beds were constructed of stainless steel with an external diameter of 2.54 cm and a length of 20.3 cm, and were packed with bituminous-base BPL activated carbon (Calgon Carbon Co., apparent density 0.85 g/mL) to provide a total mass of activated carbon of 164 g.

Cyclic operation was achieved through an electrically operated four-way solenoid valve (ASCO 8342G 701), which was controlled by an electronic timer (Digi 42A-120; Grasslin Controls Corp.). The cycle duration was set at eight hours, providing each bed with four hours of feeding and four hours of purging. The air supplied to the system was purified with the complete removal of water, oil, carbon dioxide, VOCs, and particles by a Balston FTIR purge gas generator (Paker Hannifin Corporation). Liquid VOCs was injected via syringe pumps (Harvard Apparatus, model NP-70-2208) into the air stream, where it vaporized and entered the equalizing vessel before the adsorption bed for isolating the adsorption bed from unexpected concentration fluctuations in the upstream air supply. Sampling ports were installed for both the feed and exhaust gases from the cyclic adsorption/desorption unit.

The choice of BPL activated carbon packing in the adsorption/desorption unit and the required design capacity for this unit, such as the adsorption capacity of an adsorbent for an adsorbate, were mainly based on our previous studies.[11–13] Parameters for the fixed bed and the adsorbent employed in this study are summarized in Table S1 (Supplementary Material). Adsorption equilibrium was described by the Freundlich equation.[11] The Freundlich parameters for *n*-hexane and benzene were obtained through an isotherm study of these compounds,[13] while those of methanol were adapted from Refs 18 and 19. The adsorption theory is provided in one of our previous publications[11] and therefore is not provided in this study, which concentrates on the effectiveness on the use of the integrated technology.

Trickle bed air biofilter unit. The two TBABs employed were constructed of seven cylindrical glass sections with an internal diameter of 7.6 cm and a total length of 130 cm (corresponding to an EBRT of two minutes at an air flow rate of 1.36 L/min). A liquid mixture of benzene, *n*-hexane, and methanol was injected via a syringe pump to provide the

desired influent concentration and vaporized into the air stream. Buffered nutrient solution was supplied at the rate of 2.0 L/day; more details about the formulation of the buffered nutrient solution can be found in a previous publication.[3] Each section was equipped with a sampling port that extended to the end of the column. Each biofilter was packed with pelletized diatomaceous earth biological support media (Celite® 6 mm R-635 Bio-Catalyst Carrier; Celite Corp.) to a depth of ~60 cm. The pellets were made from sintered diatomaceous earth and were therefore principally silica (SiO_2). Their physical properties were investigated in a previous study.[20] The biofilter was seeded with an aerobic microbial culture obtained from a wastewater treatment plant and pre-acclimated to *n*-hexane, benzene, and methanol.[14] The biofilters were maintained at a constant operating temperature of 20°C in a constant temperature chamber. To simulate the transient contaminant loading in the industry, four different square wave changes of *n*-hexane, benzene, and methanol concentrations were considered in this study, as detailed in Table 1 and illustrated in Figure S3 (Supplementary Material).

Analytical methods. Gas-phase samples were taken in gas-tight syringes through low-bleed and high-puncture-tolerance silicone gas chromatography (GC) septa installed in the sampling ports. Samples of *n*-hexane, benzene, and methanol were immediately analyzed by using a gas chromatograph (Agilent 6890 series) equipped with a flame ionization detector (FID) and 30 m × 320 µm × 0.25 µm column (HP-5, 5% phenyl methyl siloxane). The GC oven was programmed isothermally at 60°C. The carrier gas (He) flow rate was set at 2.7 mL/min. FID was used with He make-up gas at a flow rate of 45 mL/min, a fuel gas flow (H_2) of 35.3 mL/min, and an oxidizing gas flow (air) of 450 mL/min. The detector temperature was set at 250°C. Retention times of 1.064, 1.231, and 1.382 minutes for methanol, *n*-hexane, and benzene, respectively, were obtained under the conditions used. Carbon dioxide samples were also taken by using gas-tight syringes through sampling ports in the TBABs. A GC unit (HP 5890, Series II, Hewlett-Packard) equipped with a thermal conductivity detector (TCD) was used for determining the CO_2 concentrations in the effluent gas phase. The detection limit was 0.001%v CO_2. Liquid-phase measurements included influent and effluent concentrations of nitrate, dissolved total carbon (TC), dissolved inorganic carbon (IC), and volatile suspended solids (VSS). Nitrate concentration was determined by measuring the UV absorption at 220 nm using a Shimadzu UV mini 1240 UV–vis spectrophotometer (Shimadzu Corp.). TC and IC contents of the aqueous samples were determined by using a Shimadzu TOC 5000 analyzer. VSS analysis was carried out according to Standard Methods 2540G.[21]

Both TBABs were run using the starvation technique for a period of 2 days per week in the co-current air and water downward flow mode as means of biomass growth control within the bed. The starvation technique could be explained as no flow of VOC passing through the TBAB in order to let the excess biomass die out. Occasionally, backwashing was used to reduce the accumulation of excess biomass within the bed media as the VOCs LRs increased. More details about biomass control strategies could be found in previous publications.[3,4,20]

Microbiology analysis. Initially, TBABs A and B were mostly dominated by fungi communities, very specifically by *Fusarium solani* in the top section of the TBABs and *Gibberella moniliformis* (*Fusarium verticillioides*) in the bottom section.[14] In continuation of our previous study focusing on the characterization of fungi communities within the TBABs, samples were collected at the end of the experimental run in the current study.

Table 1. *n*-Hexane, benzene, and methanol square wave inlet loading conditions.

	TYPE "A"	TYPE "B"	TYPE "C"	TYPE "D"**
Average *n*-hexane loading rate (g m^{-3} h^{-1})	4.63	7.16	9.25	17.48
Average benzene loading rate (g m^{-3} h^{-1})	12.42	19.20	24.79	47.10
Average methanol loading rate (g m^{-3} h^{-1})	11.30	17.49	22.57	42.68
Average total loading rate (g m^{-3} h^{-1})	28.35	43.85	56.61	107.26
Inlet concentration (*n*-hexane)				
Peak concentration (ppmv)	500	500	225	181/281
Base concentration (ppmv)	20	20	75	100
Inlet concentration (benzene)				
Peak concentration (ppmv)	543	543	672	538/837
Base concentration (ppmv)	60	20	223	299
Inlet concentration (methanol)				
Peak concentration (ppmv)	1749	1749	1478	1185/1843
Base concentration (ppmv)	130	130	492	658
Duration of peak (min)*	6	12	10	15/15

Notes: *The duration of peak is repeated every 1-h cycle. **Wave type "D" consisted of two peak concentrations, medium and high.

Sample collection and DNA isolation. Biological samples were collected from the first sampling port at the top section within the biofilter media and the last sampling port at the bottom section within the media. The samples consisted of five inert pellets covered with the biomass. The pellets were immersed in 20 mL of sterilized water, and a 20-second vortex was necessary to fluff off the biomass. The samples were preserved at around –20°C until analysis. The DNA template was extracted using the QIAgen kit according to the manufacturer's instructions. Based on our previous study,[14] a minimum 10 ng/µL concentration of the DNA template was used for the next steps.

Polymerase chain reaction analyses. The same primers as used in the previous study were used in order to identify specific strains such *Aspergillus niger* and *Fusarium solani*, as well as the universal primers ITS86M-F and ITS4M-R. PCR was performed using the 18S rRNA gene. The primers used were the forward primer FF102 (5′-CTGAAAGCGTGCAGTCTGAGT-3′) and reverse primer FRev 102 (5′-TTCAGCGGGTATCC CTACCT-3′) for *A. niger* detection. For fungus *F. solani*, the forward and reverse primers were, respectively, 107F (5′-AG AGGACCCCTAACTCTGTTTCT-3′) and 107R (5′- TTT CCTCCGTCTTATTGATATGC-3′). The universal primers ITS86M-F (5′-TGAATCATCGACTCTTTGA-3′) and ITS4M-R (5′-ATAGTTATTCGCCTCC-3′) were previously successfully used[14] and also used by Ref. 22 for a wide range of fungi detection. The 18S PCRs 50 µL contained 0.2 mM of primers, 0.8 µL of DFS Tag DNA polymerase, 5 µL of buffer (Boca Scientific Inc.), and 1 µL of dNTP Mix N0447L (10 mM) (New England Biolabs) as well as 2 µL of sample DNA. Initial denaturation was at 94 µC for two minutes, followed by 35 cycles of denaturation (at 94°C), annealing (at 56°C for 107F/107R, at 54°C for FF102/FRev102, and at 66°C for ITS86M-F/ITS4M-R) all at 30 seconds and extension at 72°C for 60 seconds.

Microbial sequencing. The PCRs products were then cloned using TA Cloning® kits with pCR®2.1 vector and One Shot® TOP10 chemically competent *E. coli* (Life Technologies) according to the manufacturer's protocol. Selected single large white colonies were placed in an LB/ampicillin broth and cultured at 37°C overnight. The samples were centrifuged and the pellets were sent for further DNA extraction and sequencing to the CCHMC Genetic Variation and Gene Discovery Core Facility at Cincinnati, OH. Vector forward T7 (pCR®2.1) was used as promoter for in vitro RNA transcription and sequencing. Sequences were then submitted to BLAST homology search algorithms to assess similarity to sequences in the 18S rRNA sequences (Fungus) database at the National Center for Biotechnology Information (NCBI).

Results and Discussion

TBAB performance. The integrated unit and a standalone biofilter serving as a control unit were operated under different square wave feeding conditions, as shown in Table 1. The table also summarizes the different VOC loads applied. The VOCs were previously mixed in the desired ratio and then introduced to the air stream with two syringe pumps running at different flow rates. One of the syringe pumps was connected to an automatic time-programmed on/off switch to establish the square wave effect, and the other one was allowed to run continuously.

The experimental setup was designed so that the system would be receiving dynamic VOC LRs. The first run was with a Type A square wave with a total VOC LR of 28.35 g/m^3 h and operated for 30 days. The square wave was designed to provide an inlet concentration of *n*-hexane, benzene, and methanol base at 20, 60, and 130 ppmv, respectively, for 54 minutes, and then for 6 minutes, whereupon the concentrations sharply increased to reach peaks of 500, 543, and 1749 ppmv for *n*-hexane, benzene, and methanol, respectively. The cycle was then repeated every 60 minutes. The results of this run are presented in Figure 1. The removal efficiency is plotted in a box plot, which is a summary that plots graph data as a box representing statistical values. The boundary of the box closest to zero indicates the 25th percentile, a line within the box marks the median, and the boundary of the box farthest from zero indicates the 75th percentile. The error bars above and below the box indicate the 90th and 10th percentiles, respectively. The figure clearly indicates that for *n*-hexane, which is the most hydrophobic contaminant among the VOCs, the performance obtained from the integrated unit is higher and more stable (89% ± 7%) than that obtained for the control unit (72% ± 15%). It seems that the change in the influent concentration stressed the control unit, which could not handle the surge in the concentration. A longer acclimation period of 2 days was required after the 2-day starvation period per week to stabilize the control unit. On the other hand, the integrated unit did not require any apparent acclimation period because the inlet concentration did not exceed the critical concentration of 152 ppmv for *n*-hexane and the biofilter was continuously fed during the starvation period from the desorption of the cyclic adsorption/desorption unit. The highly influent VOC concentrations fed to the control unit provided an LR of 172 g/m^3 h. Such LRs are far beyond the capacity of the biofilter, which had been determined previously to be 117.7 g/m^3 h.[14] On the other hand, both the integrated and control units were very efficient with respect to the other contaminants, namely benzene and methanol. More than 98% and 94% removal efficiencies were obtained in both units for benzene and methanol, respectively. The high removal efficiency of benzene might have been triggered by methanol that enhanced the bioavailability of benzene. The other important factor that enhanced VOC removal efficiencies, specifically for *n*-hexane, was the stable influent concentrations to the TBAB in the integrated unit. The adsorption/desorption unit positively alleviated the surge in high peak concentrations and stabilized them to around an average effluent concentration

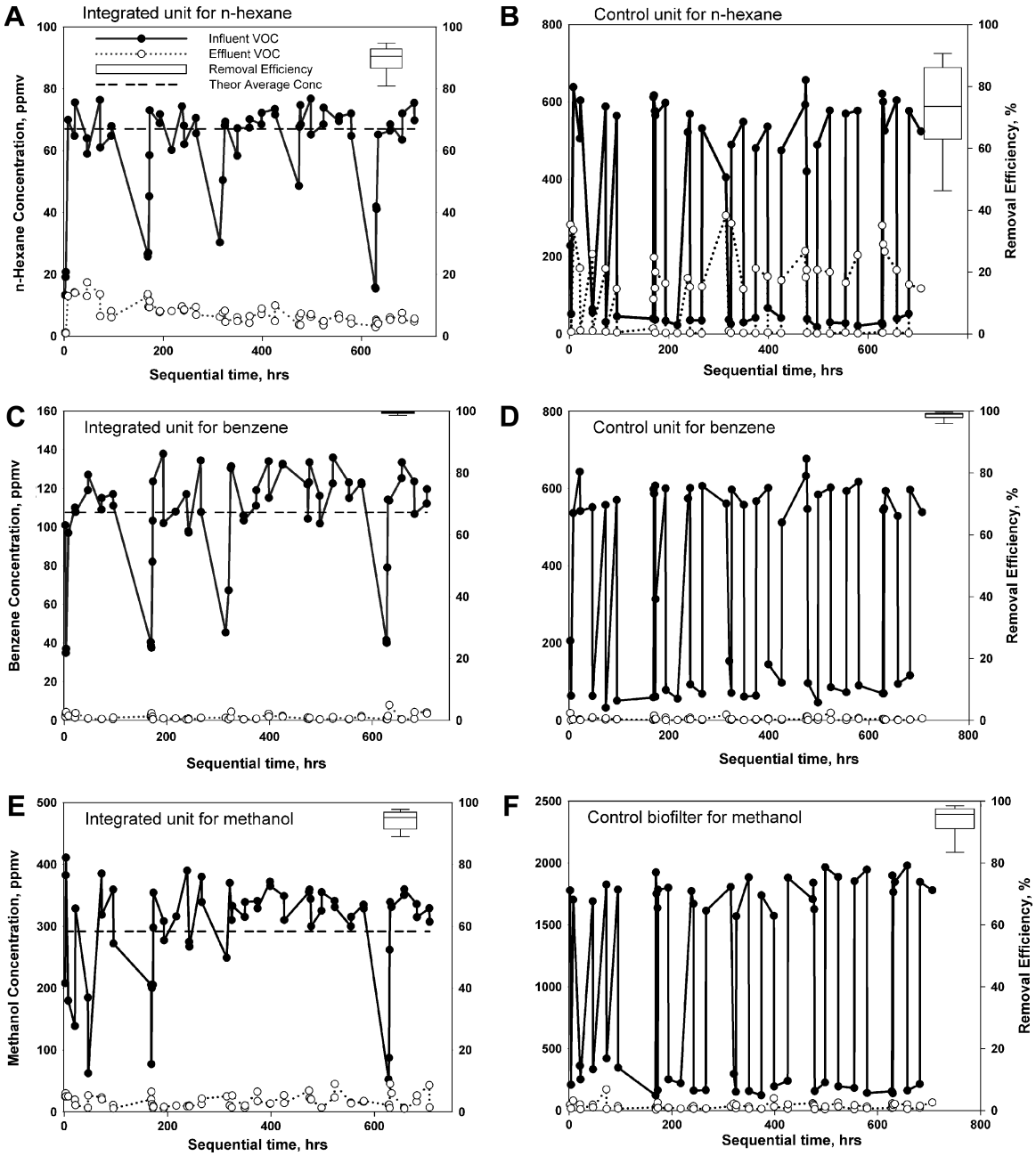

Figure 1. Biofilter performances under wave Type A feeding condition for *n*-hexane, benzene, and methanol.

of 64 ± 14, 112 ± 21, 315 ± 56 ppmv for *n*-hexane, benzene, and methanol, respectively. These values were oscillating around the average theoretical (time-weighted) values of 67, 107, 292 ppmv for *n*-hexane, benzene, and methanol, respectively. The stabilized influent concentrations to the biofilter due to the cyclic adsorption/desorption beds greatly enhanced *n*-hexane biodegradation and allowed the elimination of benzene and methanol in the integrated unit. Therefore, the biofilter's overall performance was controlled by that of *n*-hexane in the mixture. The same observation had previously been reported while treating a mixture of toluene, styrene, MEK, and MIBK with the same integrated unit. Toluene was controlling the biofilter performance of the control unit.[12] It is worth noting that the current removal efficiencies are higher for the same influent concentrations fed continuously to the biofilter than in our previous study.[14] In our previous study treating VOC mixtures of *n*-hexane, methanol, and benzene, REs of *n*-hexane and benzene were, respectively, 67% and 91% as compared to 89% and 99% obtained in this study.[14] Methanol was always the most biodegradable due to its hydrophilic properties, and there was no difference in performance between the previous study and the current one.

The system was then subjected to the second square wave of Type B, with the same base and high peak concentrations maintained but with the duration time doubled for the high peak concentrations. This run was aimed at investigating the

change in behavior of the integrated and control units for longer high peak durations (12 minutes). Because of the change of the duration, the total VOC loading rate increased to 43.85 g/m^3 h and lasted for 25 days of operation. This provides an average concentration of 116 ppmv for *n*-hexane, 156 ppmv for benzene, and 454 ppmv for methanol in the biofilter preceded by the adsorption/desorption unit. Figure S4 (Supplementary Material) summarizes the performance results of this run for square wave Type B. The duration time of 12 minutes for the high peak concentrations had affected all VOC biodegradation in the integrated unit but more extensively the control unit in eliminating the most hydrophobic VOC, namely *n*-hexane. *n*-Hexane removal decreased from 89% to 85% ± 9% and from 72% ± 15% to 56% ± 24% in the integrated unit and control unit, respectively. The erratic behavior in the control unit is an indication of its inability in handing such high concentrations loads at such duration (12 minutes compared to 6 minutes in the previous run). Similar to Type A square wave, benzene and methanol were the most eliminated in the integrated unit and were not affected by the current loads in concentrations. This is an indication that the integrated unit could handle higher loads of benzene and methanol. However, for the control unit, benzene RE was affected and 91% ± 10% RE was obtained. Methanol, on the other hand, was not affected. The decrease in *n*-hexane and benzene removal efficiencies in the integrated unit could be attributed to the increase of the LRs. For *n*-hexane, the average LR was 4.63 g/m^3 h (Type A) as compared to 7.16 g/m^3 h (Type B), while for benzene it was 12.42 g/m^3 h (Type A) as compared to 19.20 g/m^3 h (Type B). For the control unit, the long duration of high peak impacted significantly the biodegradation of *n*-hexane. The behavior of the integrated unit is in harmony with that in previous studies for treating a mixture of *n*-hexane and benzene[23] as well as mixtures of toluene, styrene, MEK, and MIBK.[12] Doubling the peak duration time while keeping the same base and high peak VOC concentrations led to a decrease in the RE of the more hydrophobic VOCs and to a lesser extent the hydrophilic ones.

The theoretical feeding condition for Type C square wave change for *n*-hexane, benzene, and methanol consisted of hourly 50-min base concentrations of 75, 223, and 492 ppmv, respectively. During the remaining 10 minutes, the peak concentrations of *n*-hexane, benzene, and methanol were 225, 672, and 1478 ppmv, respectively. The square-wave range leads to an average theoretical time-weighted concentration of 100, 298, and 656 ppmv for *n*-hexane, benzene, and methanol, respectively. With the increase of VOC loads, the effluent concentration of the cyclic adsorption/desorption beds was less stable than the previous Types A and B with an average of 108 ± 14 ppmv for *n*-hexane, 331 ± 34 ppmv for benzene, and 697 ± 42 ppmv for methanol. The RE and effluent concentration for *n*-hexane for the integrated and control units are provided in Figure S5 (Supplementary Material). It is clearly seen from the figure that the increase of VOC LRs to 56.61 g/m^3 h caused a decrease in the performance of the integrated unit for biodegrading *n*-hexane, and an RE of 72% ± 10% was obtained. On the other hand, the control unit exhibited an irregular performance in *n*-hexane removal, for which an average of 48% ± 20% was obtained (between 41% and 91% for the base concentration and from 20% to 51% for high peak concentration).

While applying this square wave, the effect of fluctuations in the influent concentration was more evident during the restart-up of the control unit after the starvation period. The control unit required more than 3 days for acclimation. It is believed that the fluctuation in the inlet concentration exceeded the capacity of the biofilm present in the control unit. Therefore, the microbial population present needed more time to be acclimated to the higher concentration of *n*-hexane and to build up sufficient biomass. In the case of the integrated unit, the adsorption unit reduced the peak concentrations to around the average VOC value to the biofilter. As for benzene and methanol elimination, both TBABs exhibited high removal efficiencies (over 95%).

Compared to those of our previous study for treating a binary mixture of *n*-hexane and benzene in the same TBABs and with the identical operational conditions (same wave peak time, EBRT, and influent concentrations),[23] the present results are significantly higher and more stable. In fact, for the integrated unit, an RE of 72% was obtained for treating *n*-hexane, whereas it did not exceed 33% in the previous study. In the same way, for benzene 98% RE was obtained while it was only 88% for the previous study. For the control unit also, the same trend was observed. The current data revealed that the introduction of methanol was very beneficial in enhancing the biofiltration of *n*-hexane and benzene. This is in conformity with our previous study in treating *n*-hexane and methanol.[24,25]

The fourth square wave applied was Type D, which was operated for 3 weeks for total VOC LRs of 107.3 g/m^3 h. This square wave has a two-step increase in the influent VOC concentrations. The base concentrations of 100, 299, and 658 ppmv for *n*-hexane, benzene, and methanol, respectively, were applied for 30 minutes. The medium peak lasted for 15 minutes and consisted of 181, 538, and 1185 ppmv for *n*-hexane, benzene, and methanol, respectively. The high peak lasted for 15 minutes and the square wave range led to average theoretical time-weighted concentrations of 100, 298, and 656 ppmv for *n*-hexane, benzene, and methanol, respectively. Figure S6 (Supplementary Material) summarizes the results of the square wave Type D. At these LRs, more fluctuations in the adsorption/desorption unit became apparent. The influent VOC concentrations were 171 ± 11, 542 ± 29, and 1227 ± 126 ppmv as compared to the theoretical average concentrations of 165, 493, and 1085 ppmv for *n*-hexane, benzene, and methanol, respectively. The variation in supplying VOCs to the biofilters affected the biodegradation of the contaminants, mainly *n*-hexane and benzene, and led to 75 ± 5% and 88.8 ± 6% in *n*-hexane and benzene REs. Methanol was not affected with these fluctuations, and more than 97% in RE was obtained. High VOC loads might also contribute to the

decrease in *n*-hexane and benzene, which could be due to methanol inhibition for *n*-hexane and benzene biodegrading microorganisms. On the other hand, the variation in the VOC LRs critically affected the stability of the control unit and diminished considerably its performance. A very low *n*-hexane RE of 32% ± 20% was obtained, while for benzene the RE was 75.8 ± 13%. As in previous runs, the methanol was largely eliminated, and a 95% RE was maintained. It seems that the combination of high loading VOC rates and the square wave type considerably decreased the capacity of the control unit in eliminating *n*-hexane and benzene.

On comparing the overall performance of the biofilters (integrated unit and control unit), it is clear that the wave duration, intensity, and frequency are the main factors that affected the VOC elimination. Keeping the same VOC loads and changing the duration of the peak concentration from 6 to 12 minutes reduced *n*-hexane and benzene REs of the control unit and to a lesser degree the REs of the integrated unit. However, increasing the VOCs loading rates and reducing the peak wave time (Type C as compared to type B) resulted in more deterioration in the performance of the biofilter in eliminating *n*-hexane as compared to other VOCs. On the other hand, increasing and varying the wave frequency and magnitude of the square wave (Type D) affected VOC biodegradation, especially of the control unit, in handling frequent change in the VOC concentration.

The integrated unit, taking advantage of the two-bed adsorption desorption unit, was less sensitive to the variations in wave duration, frequency, and loads as compared to the control unit. It is seen from Figure 2 that, at all feeding conditions, the effluent concentrations from the integrated system were varying within a very restricted range, whereas the control unit allowed wide variations.

The results noticeably indicated that the integrated system succeeded in attenuating fluctuations in the VOC influent concentrations as compared to the control unit that suffered from variability in wave frequency and intensity. As a consequence, the integrated unit sustained stable and higher performance in biodegrading VOCs as compared to the control unit.

TBAB performance after nonuse period. In industrial settings, periods of nonuse, such as, shut down for maintenance, on weekends, and on holidays, are frequent. Continuously feeding the biofiltration systems to sustain their biological activity will preclude starvation of these systems and shorten the start-up acclimation time necessary for biofilter performance.

Figure 3 summarizes the influent and effluent concentrations of *n*-hexane during the start-up of the integrated unit (Fig. 3A) and the control unit (Fig. 3B) with time after the starvation period. The plots focus on the acclimation period after starvation during the third week. As shown for the integrated unit (Fig. 3A), no acclimation periods were apparently observed, and subsequently high removal efficiencies were consistently attained during the experimental period as compared to the control unit.

Furthermore, the biofilter in the integrated unit was fed during the starvation period continuously due to desorption from the two-bed adsorption/desorption unit, while the control unit (Fig. 3B) was exposed to substrate starvation condition. Therefore, elongated reacclimation was needed, and biofilter acclimation was never achieved in the first 24 hours of operation.

Carbon mass balance closure. The cumulative CO_2 equivalent of VOCs at the influent as well as at the effluent (gas and liquid) of TBABs at different waves was computed. The only sources of energy and electron donors were VOCs besides sodium formate, which was used in the nutrient solution.

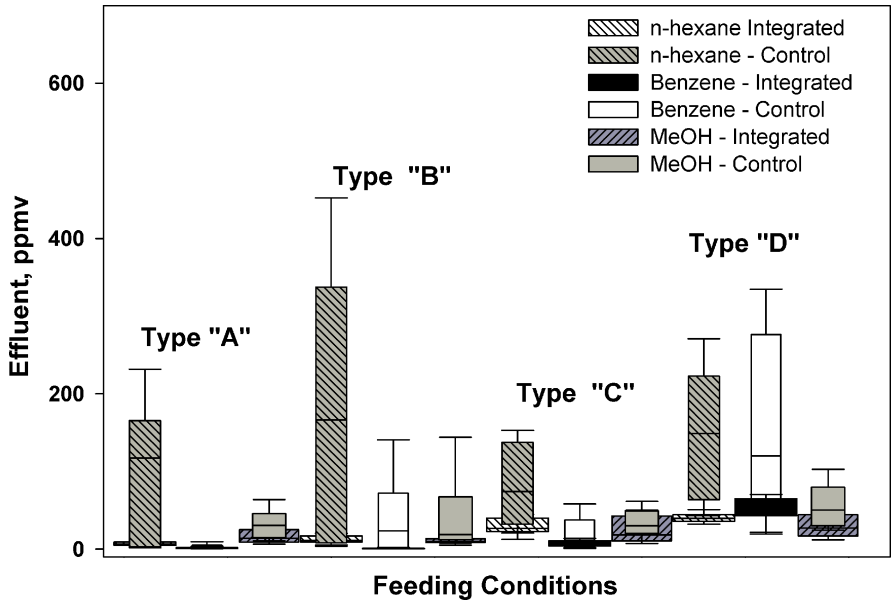

Figure 2. Summary of performances for all applied waves.

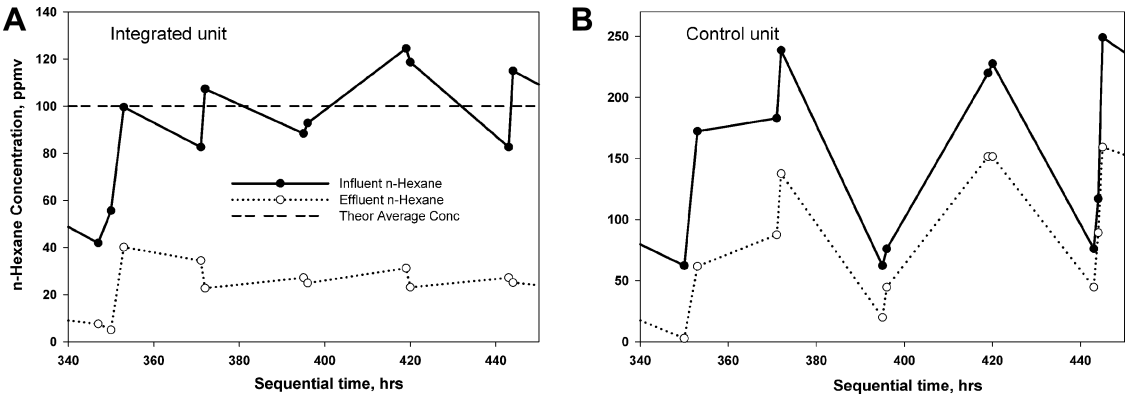

Figure 3. Removal performances of integrated unit and control unit with time after starvation: data presented for few hours after starting up the TBAB.

Consequently, the different contributors to the influent carbon represented as carbon dioxide equivalents consisted of the VOC influent gaseous concentration as well as the influent CO_2 in the liquid nutrients, whereas the effluent cumulative CO_2 was made up of VOC effluent gaseous concentrations, effluent aqueous carbon, effluent gaseous CO_2, and the carbon equivalent of effluent VSS. The CO_2-equivalent closure for the square wave Type C is shown in Figure 4. The data for the other waves are provided in (Figs. S7–S9 – Supplementary Material).

The carbon recovery rates for the waves A, B, C, and D were, respectively, 80%, 75%, 73%, and 72% for the integrated unit and 95%, 94%, 92%, and 90% for the control unit. As we were applying the starvation strategy for biomass control, it is seen from Figure 4 that there are obvious differences between the two cumulative values. These differences could be due to unwashed biomass within the biofilter, as was proven previously.[25] As the cumulative CO_2 influent for both systems is equal for each square wave applied, it is very important to note the difference between cumulative CO_2 of the integrated unit and that the control unit. It is speculated that this difference is caused by the accumulation of VOCs within the adsorption/desorption beds. Within the experimental accuracy, the VOCs accumulated were 15%, 19%, 19%, and 18% for the waves A, B, C, and D, respectively. The quantity of VOCs accumulated is in the approximate range of previous published work of 16%[26,27] and 14.8%.[12]

Removal kinetics of biofilters performance. The removal of both TBABs with respect to *n*-hexane, benzene, and methanol as a function of depth was measured weekly, 1 day following stagnation for both TBABs. The sampling ports were located at 7.6, 23, 38, 53 and 60 cm measured from the media top. These data were used to develop the pseudo-first-order reaction rate constant as a function of time. In order to compute the reaction

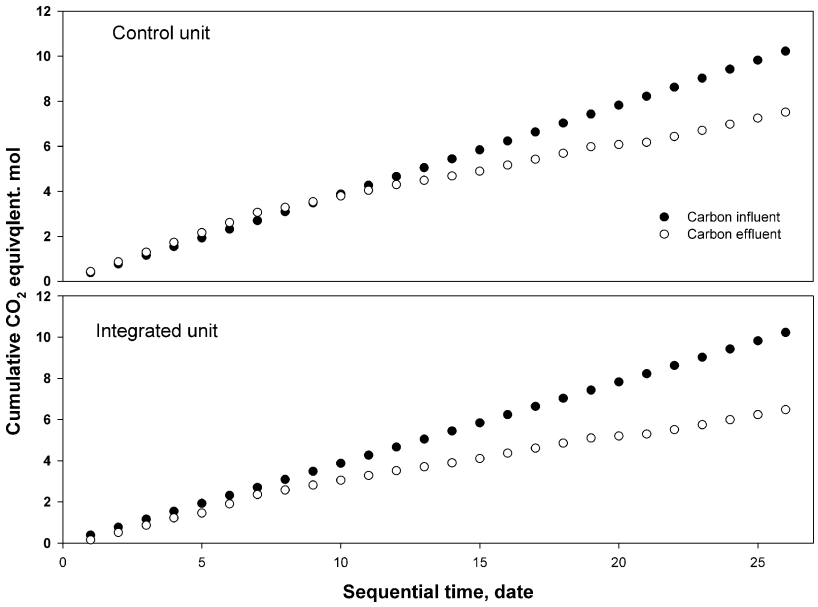

Figure 4. CO_2 closure for wave Type C.

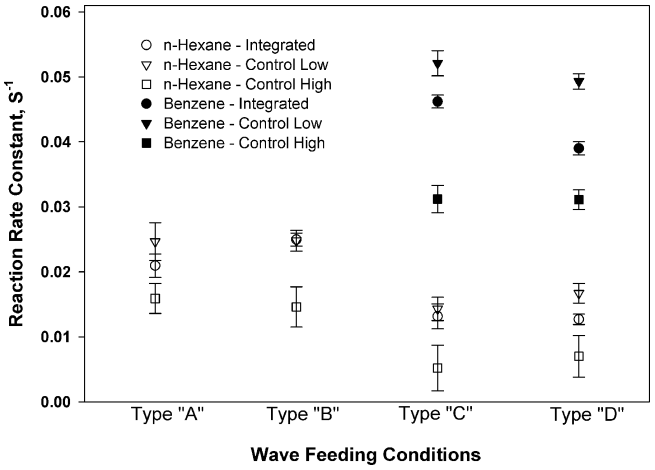

Figure 5. Removal rate constants for different VOC loading rates.

rate constant, it was assumed that the TBABs could be modeled as a plug flow reactor. The data were fitted with a linear model with the independent variable time (seconds) and the dependent variable $\log_e(C/C_0)$, where C is the effluent concentration and C_0 is the influent concentration.

As has been reported in previous studies,[24] methanol was almost eliminated in the top port of the biofilter, so it was not possible to develop a kinetics reaction rate. Similarly, for benzene, as removal was nearly 97% in the first two wave types A and B, it was not possible to compute its reaction rate constant for these two runs.

Figure 5 noticeably shows that, as the VOC loading rate increased, the drop in the reaction rate constants decreased for all the contaminants. However, the integrated unit performance was more stable and the reaction rate constants were in approximately in the same order of magnitude as the values obtained for the control unit at low peak concentrations despite the difference in the VOC loading rates. The high peak concentrations were causing deterioration in the behavior of the control unit, mainly for biodegrading *n*-hexane, which was clear from the values and standard deviations ranges. The reaction rate constant of *n*-hexane varied from 0.0127 to 0.0250/s for the integrated unit. For the control unit it varied from 0.0143 to 0.0248/s during low concentrations and from 0.007 to 0.0159/s during peak concentrations.

Microbial analyses and TBABs performances. Previously, the TBABs were fed by a mixture of *n*-hexane, benzene, and methanol in the ratio ($C_H/C_B/C_M$ = 1:3:6.6). Both TBABs were dominantly populated with the fungi *Fusarium solani* species in the top part of the TBABs and by the *Gibberella moniliformis* (*Fusarium verticillioides*) species at the bottom port of the biofilters media.

Before the start of the experiment when the TBABs were fed by the *n*-hexane, benzene, and methanol mixture, *F. solani* and *G. moniliformis* (*Fusarium verticillioides*) species were dominant in the top part and the bottom port of the biofilters media, respectively. Clustering of the species distribution remained identical in the integrated and control units. The nonchange of microbial structures in both TBABs combined by different VOC removal, mainly for *n*-hexane and benzene, might be due to feeding the TBABs with the same VOCs. This is in contrast to the results of our previous study, where change in VOC types provided different microbial structures.[27] However, when the system reached the steady state at the end of the previous experimental run, it could be speculated that a change in concentrations of VOCs did not change the dominance of the microbial species. It might be inferred from this finding that, as there were no changes in type of VOCs, nutrients, and mode of operation, the ecology of biofiltration systems should remain stable. However, for the bottom port of the control unit, only 1 of 10 cloned samples was positive, but only *G. moniliformis* (*Fusarium verticillioides*) species were detected. Limited and therefore non-representation of sample of only 10 cloned samples for each port could be another factor why the other species might not be detected.

On the other hand, the outcomes of this experiment verified that *F. solani* species is well suited for dealing with VOC mixtures, whereas *G. moniliformis* (*Fusarium verticillioides*) is the best at biodegrading the sole VOC (*n*-hexane) at high loading rates or a mixture of VOCs at low concentrations, which explains its existence at the bottom port of the TBABs. It could be speculated that higher VOC loads inhibit the species *G. moniliformis* (*Fusarium verticillioides*). Also, benzene and methanol are mostly biodegraded at the top port of the bio filters, which explains their high removal rate as confirmed by the reaction rate constants. As there was no change in the dominance of the fungi species within the biofilters, the fluctuations in VOC feeding the control unit would be the key factor for the erratic behavior of the biofilter. As consequence, incomparable performances were obtained for the integrated unit and the control unit, mainly for eliminating *n*-hexane and, to a lesser degree, benzene. In fact, the fluctuation in the feed VOC to the control unit might negatively influence the rate of VOC uptake by the microorganisms that need more time to metabolize the contaminants, whereas such fluctuations are not encountered in the integrated unit. It should be noted that biofiltration systems are very sensitive to changes in VOCs types, flows, and intensity.

Placing the two-bed adsorption/desorption unit before the biofilter bed could be a very good technique in reducing the fluctuations in VOC influent as well as serving as a buffer during nonuse periods, which will affect positively the uptake of these VOCs by microorganisms and hence stabilize the performance of biofiltration systems. The experimental data obtained in this study confirmed the effectiveness of placing the two-bed adsorption/desorption unit prior to the biofiltration systems. This confirms the results of our previous studies.[11–13,23]

Conclusions

This study evaluated the effectiveness of an integrated system composed of cyclic adsorption/desorption beds and a

biofilter in treating a dynamic mixture of *n*-hexane, benzene, and methanol. Four different square waves of the tri-mixture were applied to the cyclic beds. The performance of the integrated system was compared to a stand-alone biofilter receiving the four different square waves of influent concentrations. The experimental results clearly indicated that the cyclic adsorption beds buffered the fluctuations in the inlet concentrations, which in turn provided the following biofilter with a stable influent concentration. Furthermore, the cyclic adsorption/desorption beds provided continuous feed to the biofilter during the starvation period, which was 2 days per week, during which there was no contaminant feed to the cyclic beds.

The stand-alone biofilter provided erratic performance especially with respect to the hydrophobic contaminant *n*-hexane. Methanol and to some extent benzene were tolerant to concentration changes. The utilization of the hydrophilic compound methanol in the mixture aided the removal of benzene as compared to our previous study, where only benzene and *n*-hexane were evaluated.

The proposed integrated system is cost effective since the cost introduced will be mainly the initial cost of the two beds. There is no extra operational energy cost introduced since the flow is diverted from one bed to the other by the use of a solenoid valve. Furthermore, the proposed integrated technology, which was effective for various contaminants in our laboratories as illustrated by our previous publications, can be extended to industrial facilities where poor biofiltraion performance is attained due to fluctuating inlet feed.

Author Contributions

Conceived and designed the experiments: AZ. Analyzed the data: AZ. Wrote the first draft of the manuscript: AZ. Contributed to the writing of the manuscript: GS. Agree with manuscript results and conclusions: GS. Jointly developed the structure and arguments for the paper: AZ and GS. Made critical revisions and approved final version: GS. All authors reviewed and approved of the final manuscript.

Supplementary Materials

Supplementary Figure 1. Schematic of TBABs.

Supplementary Figure 2. Cyclic adsorption/desorption beds system.

Supplementary Figure 3. Type of square waves of VOCs influent concentration.

Supplementary Figure 4. Biofilters performances under wave type "B" feeding condition for n-hexane, benzene, and methanol.

Supplementary Figure 5. Biofilters performances under wave type "C" feeding condition for n-hexane, benzene, and methanol.

Supplementary Figure 6. Biofilters performances under wave type "D" feeding condition for n-hexane, benzene, and methanol.

Supplementary Figure 7. Carbon mass balance closure for wave type "A".

Supplementary Figure 8. Carbon mass balance closure for wave type "B".

Supplementary Figure 9. Carbon mass balance closure for wave type "D".

Supplementary Table 1. Parameters for adsorption model.

REFERENCES

1. Deshusses MA, Cox HHJ. Biotrickling filters for air pollution control. In: Bitton G, ed. *The Encyclopedia of Environmental Microbiology*. New York, NY: Willey; 2000:782–795.
2. Smith FL, Sorial GA, Suidan MT, Pandit A, Biswas P, Brenner RC. Evaluation of trickle bed air biofilter performance as a function of inlet VOC concentration and loading, and biomass control. *J Air Waste Manag Assoc*. 1998;48:627–636.
3. Kim D, Cai Z, Sorial GA. Evaluation of trickle-bed air biofilter performance under periodic operating conditions as a function of styrene loading. *J Air Waste Manag Assoc*. 2005;55:200–209.
4. Sorial GA, Smith FL, Suidan MT, Pandit A, Biswas P, Brenner RC. Evaluation of trickle bed air biofilter performance for BTEX removal. *J Environ Eng*. 1997;123:530–537.
5. Cox HHJ, Deshusses MA. Effect of starvation on the performance and reacclimation of biotrickling filters for air pollution control. *Environ Sci Technol*. 2002;36:3069–3073.
6. Fitch MW, England E, Zhang B. 1-Butanol removal from a contaminated airstream under continuous and diurnal loading conditions. *J Air Waste Manag Assoc*. 2002;52:1288–1297.
7. USEPA. The Clean Air Act Amendments of 1990 List of Hazardous Air Pollutants. http://www.epa.gov/ttnatw01/orig189.html (accessed June 19, 2015).
8. Manninen MR, Niemi BA, Kleinheinz GT. Use of a coupled biological system to treat a chemically complex air stream. *Arch Environ Contam Toxicol*. 2003; 45:1–10.
9. Weber FJ, Hartmans S. Use of activated carbon as a buffer in biofiltration of waste gases with fluctuating concentrations of toluene. *Appl Microbiol Biotechnol*. 1995;43:365–369.
10. Li C, Moe WM. Activated carbon load equalization of discontinuously generated acetone and toluene mixtures treated by biofiltration. *Environ Sci Technol*. 2005;39:2349–2356.
11. Kim D, Cai Z, Sorial GA, Shin H, Knaebel K. Integrated treatment scheme of a biofilter preceded by a two-bed cyclic adsorption unit treating dynamic toluene loading. *Chem Eng J*. 2007;130:45–52.
12. Cai Z, Sorial GA. Treatment of dynamic VOC mixture in a trickling-bed air biofilter integrated with cyclic adsorption/desorption beds. *Chem Eng J*. 2009;151: 105–112.
13. Aly Hassan A, Sorial GA. Treatment of dynamic mixture of hexane and benzene vapors in a trickle bed air biofilter integrated with cyclic adsorption/desorption beds. *Chemosphere*. 2011;82:521–528.
14. Zehraoui A, David W, Sorial GA. Biodegradation of a mixture of hydrophobic and hydrophilic VOCs in trickle-bed-air-biofilters. *Water Air Soil Pollut*. 2014;225:2075.
15. US EPA. *Superfund Chemical Data Matrix (SCDM); Part 5: Chemical-Specific Parameters*. 2013. Available at: http://www.epa.gov/superfund/health/conmedia/soil/pdfs/part5.pdf. Accessed June 1, 2015.
16. Verschueren K. *Handbook of Environmental Data on Organic Chemicals*. Vol 1. New York, NY: John Wiley and Sons, Inc.; 2001.
17. Moe WM, Li C. A design methodology for activated carbon load equalization systems applied to biofilters treating intermittent toluene loading. *Chem Eng J*. 2005;113:175–185.
18. Reucroft PJ, Simpson WH, Jonas LA. Sorption properties of activated carbon. *J Phys Chem*. 1971;75:3526–3531.
19. Duchowicz PR, Castañeta H, Castro EA, Fernández FM, Vicente JL. QSPR prediction of the Dubinin-Radushkevich's k parameter for the adsorption of organic vapors on BPL carbon. *Atmos Environ*. 2006;40:2929–2934.
20. Smith FL, Sorial GA, Suidan MT, Breen AW, Biswas P, Brenner RC. Development of two biomass control strategies for extended, stable operation of highly efficient biofilters with high toluene loadings. *Environ Sci Technol*. 1996;30: 1744–1751.
21. Eaton AD, Clesceri LS, Rice EW, Greenberg AE, Franson MAH. *Standard Methods for Examination of Water & Wastewater*. 21st ed. Washington, DC: American Public Health Association, American Water Works Association, Water Environment Federation; 2005.

22. Soeta N, Terashima M, Gotoh M, et al. An improved rapid quantitative detection and identification method for a wide range of fungi. *J Med Microbiol*. 2009;58: 1037–1044.
23. Aly Hassan A, Zehraoui A, Sorial G. Achieving stable biological oxidation of erratic waste gas streams by utilizing cyclic adsorption/desorption beds. *J Adv Oxid Technol*. 2010;13:9–14.
24. Zehraoui A, Hassan AA, Sorial GA. Effect of methanol on the biofiltration of n-hexane. *J Hazard Mater*. 2012;219–220:176–182.
25. Zehraoui A, Hassan AA, Sorial GA. Biological treatment of n-hexane and methanol in trickle bed air biofilters under acidic conditions. *Biochem Eng J*. 2013; 77:129–135.
26. Cai Z, Kim D, Sorial GA. A comparative study in treating two VOC mixtures in trickle bed air biofilters. *Chemosphere*. 2007;68:1090–1097.
27. Zehraoui A, Kapoor V, Wendell D, Sorial GA. Impact of alternate use of methanol on n-hexane biofiltration and microbial community structure diversity. *Biochem Eng J*. 2014;85:110–118.