The Journal of Microbiology, March 2000, p.31-35
Vol. 38, No: 1

Selection of Suitable Packing Material for Biofiltration of Toluene, *m*- and *p*-Xylene Vapors

Young-Sook Oh and Sung-Chan Choi*

Division of Life Sciences, Hallym University, Chunchon 200-702, Korea

(Received December 28, 1999 / Accepted February 7, 2000)

A suitable packing material for biofiltration of monoaromatic solvent vapors was selected among various types of packing materials such as peat, bark chips, vermiculite, and Hydroballs. A previously isolated strain, *Pseudomonas pseudoalcaligenes* BTXO2, which could utilize toluene, *m*- and *p*-xylene as carbon and energy sources was used as a biofilter inoculum. Four glass biofilters (6 cm dia. × 60 cm) were individually packed with each of the packing materials and solvent vapors were passed through the columns. During three weeks of peat biofilter operation, average removal efficiencies of toluene, *m*- and *p*-xylene were 90.4%, 95.3%, and 82.1%, respectively. With the other packings, the efficiencies were in the range of 10.1 to 58.6% which were significantly lower than those of the peat biofilter. The peat biofilter was continually operated for approximately nine months and the biofilter sustained its degradation activity during the operation period with minimal maintenance. At steady state, average removal rates of toluene, *m*- and *p*-xylene vapors were estimated as 14.2, 5.5, and 8.1 g m^{-3} packing h^{-1}, respectively.

Key words: Biofiltration, degradation, peat packing, toluene, xylenes

Benzene, toluene, and xylenes (BTXs) are substantive constituents of gasolines (10) and are also produced to serve as industrial solvents and/or feedstocks for synthesis (13). Due to their wide usage, BTXs have been commonly found in air, soil, and groundwaters worldwide. During the remediation process BTXs need to be treated by gas cleaning techniques because BTXs in contaminated soils or groundwaters are easily volatilized into air.

To date, a number of physical and chemical gas cleaning techniques have been developed which achieve highly efficient removal of various compounds from industrial waste gases. Chemical methods such as thermal and catalytic destruction, ozonization and chlorination are usually capable of removing a broad spectrum of compounds, but energy consumption and/or consumption of chemicals (oxidants or catalysts) are the disadvantages of these methods (7, 16). Physical methods, such as condensation, adsorption on solids or absorption in liquids have the disadvantage of the pollutants not being destroyed. The solid adsorbents or liquids need to be regenerated and often new polluted materials are created. As a cost-effective and environmentally safe alternative, biofiltration using soil beds (12) or enclosed biofilters (9) has been successfully applied for the treatment of effluent gas streams containing low concentrations of air pollutants. Biofiltration is now a proven air-pollution-control technology and recently adapted for use in controlling emissions of volatile organic carbons (VOCs) in remediation projects (5, 18).

During biofiltration, the type of packing materials are essential so that the proper design of the materials is the basis of the vendor patents and commercial technology. Packing materials need to provide optimum conditions for inhabiting microbial populations, and therefore they need to provide a large reactive surface, a low pressure drop, and a minimum tendency for compaction. As requirements for a suitable packing material, a pH between 7 and 8, pore volume of greater than 80%, particle diameter of greater than 4 mm, and total organic matter content of more than 55% were recommended by Eitner (4).

In this study, a previously isolated strain (8) of *Pseudomonas pseudoalcaligenes* was used as a biofilter inoculum and different types of packing materials such as peat, bark chips, vermiculite, and Hydroballs were used for the biofilter bed. The biofilters were operated under similar conditions and their capabilities to degrade toluene, *m*- and *p*-xylene vapors were measured and compared.

Materials and Methods

Bacterial strain

A microbial consortium was enriched using equal parts of benzene, toluene, and xylene isomers from wastewater

* To whom correspondence should be addressed.
(Tel) 82-361-240-1531; (Fax) 82-361-242-9300
(E-mail) scchoi@sun.hallym.ac.kr

obtained from the Kuro industrial complex, and a *Pseudomonas pseudoalcaligenes* strain BTXO2 (hereafter referred to as strain BTXO2) was isolated from this consortium. The details of the isolation and identification process have been previously described (8).

Utilization of solvents

The model substrate mixture consisted of equal parts of toluene, *m*- and *p*-xylene isomers purchased from Sigma Chemical Co. with the highest grades available. The strain was pregrown on a mineral salts basal (MSB) medium (14) with the substrate mixture as the sole carbon and energy source. Cells were harvested by centrifugation (8,000 × *g*, 20 min) and the resulting cell pellet was resuspended in fresh MSB medium with the cell concentration in the range of 1.5 to 2 g dry biomass/l. Ten milliliter aliquots of the suspension were dispensed into 160 ml serum bottles (Wheaton Glass Co., USA) closed with Teflon-faced gray butyl rubber septa (Wheaton) and aluminum seals. Either each of the solvents or their mixture was added by a microsyringe, and incubated at 30°C using a rotary shaker (200 rpm). Identical bottles without cells served as negative controls, and any losses from these control bottles were subtracted from the amount of substrate which disappeared in the active bottles. Decrease of substrate was monitored by taking 100 μl gas samples from the headspace of the serum bottles using a gas-tight syringe (Hamilton Co.). The contents in the sample were determined using a gas chromatography system (Hewlett-Packard Model 5890 Series II Plus, USA) equipped with a flame ionization detector and a capillary column of 0.53 mm diameter and 30 m length (Hewlett-Packard HP-1). Chromatography conditions were: injector temp., 150°C; oven temp., 70°C; detector temp., 250°C; and nitrogen carrier gas flow rate, 10 mL/min.

Packing materials

Packing materials tested in this study were peat moss (Seungjin Co., Korea), vermiculite (Hyponex Co., USA), bark chips (Seungjin Co., Korea), and Hydroball (Haeran Co., Korea). Water holding capacities of the four packing materials were measured using the Hilgard cup method (11). Filter bed pH was measured with solutions which contained distilled water and 10% (v/v) of each packing materials. At the end of the experiments, pressure drops were measured prior to and after exit from the packed biofilters with a pressure gauge at a surface load of 12.5 m^3 m^{-2} h^{-1}.

Operation of biofilters

The glass columns used for the solvent vapor removal consisted of immobilized microbial cells and a stationary aqueous phase in a porous medium. The arrangement shown in Fig. 1 allowed adjustment of flow rates and solvent vapor concentrations independently by directing a

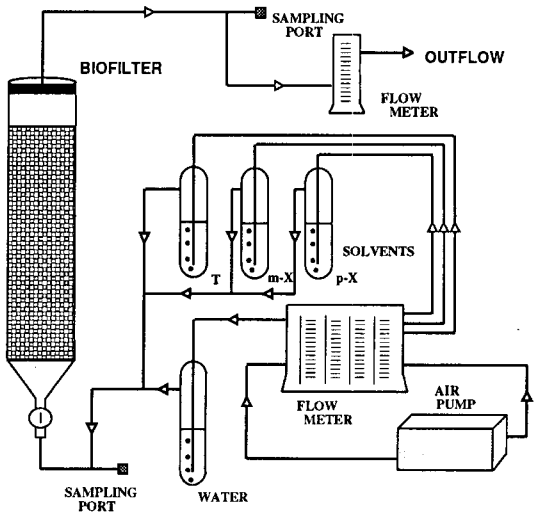

Fig. 1. Schematic diagram of biofilter used for removing toluene, *m*- and *p*-xylene vapors. The actual dimension of the glass column was 6 cm dia. × 60 cm length. Four identical columns prepared with different packing materials were used. Solvent (T, toluene; X, xylene) vapor concentrations in the air stream were measured prior to and after exit from the biofilter column by gas chromatographic analyses.

greater or smaller part of an air stream through each solvent reservoir. The balance of the air passed through water to partially humidify the air stream. Ports for gas sampling were used to determine solvent concentrations prior to and after exit from the biofilter column. Air streams generated by aquarium pumps were measured by low-capacity rotameters (Rate-Master, Dwyer Instruments, Inc.) and passed through water or solvent reservoirs. Separate flow meters allowed the measurement of the total air flow and the portions passing through water and solvents. The air streams were then combined and passed through the column. The biofilter columns were installed in exhaust hoods and operated at ambient room temperatures of 20~25°C. In preparation of the biofilter, sufficient amounts of dry packing materials were weighed to pack a glass column (6 cm dia. × 60 cm length). Strain BTXO2, pregrown on toluene, was harvested by centrifugation and suspended in fresh MSB medium. Each packing material received an equal volume of microbial suspension plus sufficient MSB medium to fill 50% of its water holding capacity. Four glass columns were individually packed with each of the moistened packing materials and solvent vapors were passed through the columns at an air flow rate of 0.018 m^3 h^{-1}. Solvent concentrations in the air streams were measured prior to and after exit from the biofilter columns using gas chromatography. Knowing the concentration drop and the volume of air passing through the column, total and its individual component removal rates could be calculated. Removal rates were expressed as grams of solvent removed per cubic meter of packing material per hour (g m^{-3} h^{-1}).

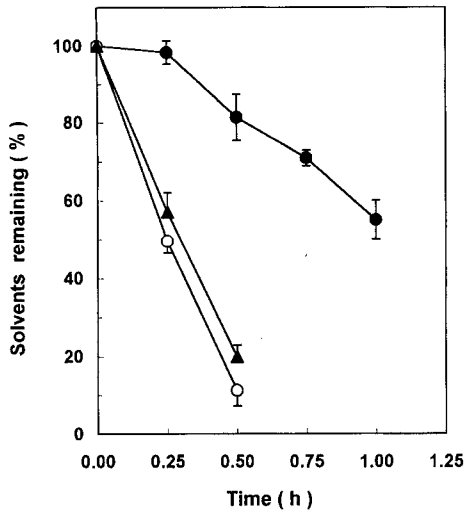

Fig. 2. Removal of toluene(●), *m*-xylene(○), and *p*-xylene(▲) by strain BTXO2 in serum bottles at initial concentrations of 92, 106, and 106 mg/l, respectively. Remaining solvents were analyzed by headspace analysis using a gas chromatograph. An identical but uninoculated sample showed no loss of solvents during the incubation period. Error bars represent SD.

Results and Discussion

Removal of toluene, m- and p-xylene by strain BTXO2

Strain BTXO2 seemed to use toluene, *m*- and *p*-xylene as carbon and energy sources, but could not utilize either benzene or *o*-xylene. Fig. 2 shows the removal of individual solvent vapor in closed serum bottles. Approximately 50% of the added toluene disappeared within 1 h of incubation. However both *m*- and *p*-xylene were rapidly degraded leaving approximately 10~20% of the added substrates within 0.5 h. When cells received the solvent mixture, *p*-xylene and *m*-xylene were preferred to toluene and removal of toluene was competitively inhibited by the presence of *m*- or *p*-xylene (data not shown). Approximately a 50% decrease in the specific toluene removal rate was observed with a 1:3 (toluene : xylenes) mixture. The removal of *m*- and *p*-xylene in the presence of toluene was also inhibited but the intensity of inhibition turned out to be much less severe than that of toluene degradation by xylenes. The substrate range of the BTXO2 strain and its inhibition pattern did not contradict its previous report (7) on the degradation of toluene and xylenes by the *Pseudomonas putida strain* (ATCC 33015) which harbors the TOL plasmid (17).

Biofiltration of toluene, m- and p-xylene vapors

In choosing packing materials, one should note a large surface area for microbial adhesion and efficient mass transfer, along with a minimal pressure drop and a minimum tendency for compaction within the biofilter. Microbial compatibility, low cost, and convenient availability are additional considerations. The packing materials we tested were peat moss, vermiculite, bark chips, and Hydroball. All of the packing materials could be easily purchased from local horticultural shops and the unit price was less than $2 per kg. Physicochemical characteristics such as filter bed pH, filter media porosity, pressure drop, filter media dry density, and filter media water holding capacity are summarized in Table 1. The parameters of used packing materials were in the range of the recommended operational parameters for biofiltration systems as suggested by Eitner (4) or Dharmavaram (3).

To choose the best packing for biofiltration of toluene, *m*- and *p*-xylene vapor, biofilters composed of each of the four packing materials were used. Table 2 shows average values of inlet concentrations, removal efficiencies, and removal rates of toluene, *m*- and *p*-xylene vapor in biofilters composed of Hydroballs, vermiculite, bark chips, and peat. At the beginning of three weeks of Hydroball biofilter operation, toluene and *p*-xylene were mainly removed but their removal efficiencies gradually

Table 1. Comparison of four different packing materials utilized for the operation of biofiltration systems

Packing materials / Parameter	Peat	Vermiculite	Bark chips	Hydroball
Filter bed pH[a]	6.7	6.9	6.8	6.9
Filter media porosity	0.74	0.71	0.80	0.61
Pressure drop (per meter bed)[b]	0	0	0	0
Filter media dry density (g/ml)	0.21	0.15	0.24	0.27
Filter media water holding capacity (ml/g)	1.55	4.84	2.11	0.65

[a] pH of 100 ml distilled water containing 10 g of each packing materials
[b] At surface load of 12.5 $m^3 m^{-2} h^{-1}$

Table 2. Average values of inlet concentrations, removal efficiencies, and removal rates of toluene and *m*-, *p*-xylene vapor during three weeks of biofilters operation composed of four different packing materials

Packings	Surface loading ($m^3m^{-2}h^{-1}$)	Parameters	Substrates		
			Toluene	*m*-Xylene	*p*-Xylene
Hydroball	0.83	Conc.[a]	2.44	0.25	0.57
		RE[b]	10.3	25.2	36.2
		RR[c]	0.85	0.19	0.55
Vermiculite	1.02	Conc.	2.16	0.24	0.46
		RE	16.2	15.2	58.6
		RR	1.20	0.15	0.91
Bark chip	0.68	Conc.	1.06	0.96	2.22
		RE	10.1	29.3	24.5
		RR	0.64	0.15	0.22
Peat	0.57	Conc.	1.52	0.16	0.50
		RE	90.4	95.3	82.1
		RR	3.21	0.34	0.98

[a]Conc., inlet concentrations (g m^{-3})
[b]RE, removal efficiencies (%)
[c]RR, removal rates (g m^{-3} h^{-1})

decreased to 10% and 30% of the inlet concentrations (data not shown). In contrast to toluene or *p*-xylene, removal efficiencies of *m*-xylene gradually increased and reached around 30%. After the Hydroball biofilter reached a stabilized condition, *m*- and *p*-xylene were mainly removed and this result was due to the substrate preference of the strain BTXO2 which was already observed in batch culture experiments. Similar to the pattern of removal efficiencies, removal rates of toluene and *p*-xylene were initially the highest during the operation, but subsequently decreased. Nevertheless, removal rates of *m*-xylene gradually increased from 0.06 to 0.31 g m^{-3} h^{-1} and total removal rate of toluene, *m*- and *p*-xylene was in the range of 0.45~4.07 g m^{-3} h^{-1} with an average of 1.59 g m^{-3} h^{-1}.

Though its water holding capacity was exceptionally high (Table 1), performance of the biofilter composed of vermiculite was somewhat improved, if any, as the total removal rate of toluene, *m*- and *p*-xylene was in the 1.38~5.78 g m^{-3} h^{-1} range and the average was 2.26 g m^{-3} h^{-1} (Table 2). The biofilter packed with bark chips was operated at a surface load of 0.68 m^3 m^{-2} h^{-1}, and all of the three compounds were poorly degraded when compared with other biofilters. Total removal rate was in the range of 0.18~3.24 g m^{-3} h^{-1} and the average rate was 1.01 g m^{-3} h^{-1}. Wood bark has been widely used as a packing material because of its excellent air permeability, easy availability and low cost (15). The results we obtained with bark chips, however, suggested that not all the organic support materials were superior to inorganic materials, Hydroball or vermiculite, for the packings of biofilter.

Peat has been also used as a support media for microorganisms due to its absorption/adsorption properties, high cellulose content, buffering capacity and easy availability (2, 6). In this study, the biofilter packed with peat removed more than 80% of the incoming toluene, *m*-, and *p*-xylene and it performed the best when compared with other biofilters. The total removal rate was in the range of 2.24~6.11 g m^{-3} h^{-1} with an average of 4.53 g m^{-3} h^{-1}. The peat biofilter was continually operated for about 9 months and the biofilter maintained sustained activity during the operation (Fig. 3). Initially, the surface loading was maintained at 0.57 m^3 m^{-2} h^{-1} and gradually increased to 1.82 m^3 m^{-2} h^{-1} at 74 days after the start-up. The same surface loading value was maintained up to the end of the experiment and the retention time was 8.7 min at this surface load. With an assumption that the biofilter reached steady state (Day 120), the average removal rates of toluene, *m*-, and *p*-xylene vapors were 14.20 ± 2.32, 5.51 ± 1.43, and 8.10 ± 2.55 g m^{-3} h^{-1}, respectively.

As a result of peat biofilter operation, a total of 88.6 g carbon was removed, but only 0.107 g nitrogen and 0.268 g phosphorus were occasionally added via diluted MSB. Assuming that the C : N : P ratio generally required for microbial degradation is 10 : 1 : 0.5 (1), our result of

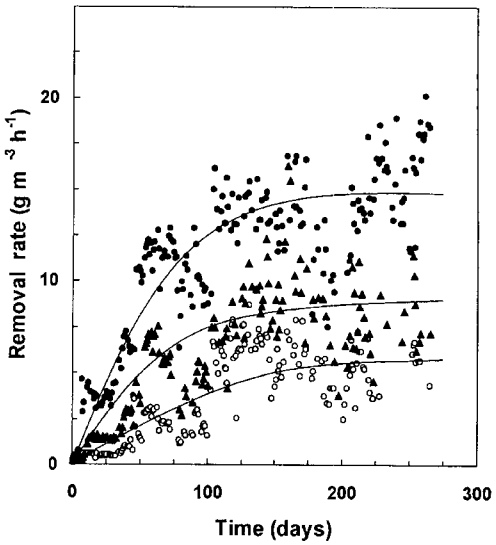

Fig. 3. Removal rates of toluene(●), *m*-xylene(○), and *p*-xylene(▲) obtained from the operation of peat-biofilter inoculated with strain BTXO2. Surface loading was initially started at 0.57 m^3 m^{-2} h^{-1} and gradually increased to 1.82 m^3 m^{-2} h^{-1} at 74 days after the start-up and maintained constant thereafter.

88.6 : 0.107 : 0.268 (=10 : 0.012 : 0.03) suggested that the biofilter required minimal maintenance. This was probably caused either by continuous regeneration of required mineral salts during cell cycles of bacteria within the biofilter or by the dissolution of mineral salts from the packing itself. If the regeneration was a primary mechanism for the supply of required mineral nutrients, possible clogging of the biofilter by the continuous increase in the biomass could be avoided by the careful control of mineral nutrient concentrations so that they limit further change of microbial growth. By doing so, one can extend the average lifetime of biofilter packings resulting in a more cost-effective overall treatment process. In addition to these advantages, the fact that organic packing wastes are usually easier and cheaper for disposal than synthetic packings favors peat as an adequate packing material for the biofiltration of volatile organic carbon emissions.

Acknowledgments

This work was supported by KOSEF (Korea Science and Engineering Foundation) Research Grant No. 961-0510-078-2.

Reference

1. Alexander, M. 1994. Biodegradation and bioremediation, p. 196-225. Academic Press, Inc., New York.
2. Beerli, M. and A. Rotman. 1989. Biofilter- A unique method to reduce and/or eliminate VOCs. Proc. of Envirocon 89: 1st International Conf. on Environ. Issues for Converters, p.1-32.
3. Dharmavaram, S. 1991. Biofiltration: A lean emissions abate-

ment technology. Abstr. 91-103.2, Abstr. 84th Annu. Meet. Air & Waste Manage. Assoc. Vancouver, B.C.
4. Eitner, D. 1989. Biofilter in der Abluftreinigung. *Umwelt.* 19, L24.
5. Leson, G. and A.M. Winer. 1991. Biofiltration: An innovative air pollution control technology for VOC emissions. *J. Air & Waste Manage. Assoc.* 41, 1045-1054.
6. Martin, A.M. 1991. Peat as an agent in biological degradation: Peat biofilters, p. 341-362. In A.M. Martin (ed.), Biological Degradation of Wastes. Elsevier Science Publishers, NY.
7. Oh, Y.-S. 1993. Ph. D. thesis. Rutgers- The State University of New Jersey, New Jersey.
8. Oh, Y.-S. and S.-C. Choi. 1997. Characterization of BTX-degrading bacteria and identification of substrate interactions during their degradation. *J. Microbiol.* 35, 193-199.
9. Ottengraf, S.P.P. 1986. Exhaust gas purification, p. 427-452. In H.J. Rehm and G. Reed (eds.), Biotechnology. VCH Publishers, Weinheim, Germany.
10. Potter, T.L. 1992. Fingerprinting petroleum products: Unleaded gasolines, p. 83-92. In P.T. Kostecki and E.J. Calabrese (eds.), Petroleum Contaminated Soils. Lewis Publishers, Chelsea, MI.
11. Pramer, D. and E.L. Schmidt. 1964. Experimental Soil Microbiology. Burgess, Minneapolis, MN.
12. Prokop, W.H. and H.L. Bohn. 1985. Soil bed system for control of rendering plant odors. *J. Air Pollut. Contr. Assoc.* 35, 1332-1338.
13. Reisch, M.S. 1992. Top 50 chemicals production stagnated last year. *Chem. Eng. News* 70, 16-22.
14. Stanier, R.Y., N.J. Palleroni, and M. Doudoroff. 1966. The aerobic pseudomonads: a taxonomic study. *J. Gen. Microbiol.* 43, 159-171.
15. Van Langenhove, H., E. Wuyts, and N. Schamp. 1986. Elimination of hydrogen sulfide from odorous air by a wood bark biofilter. *Wat. Res.* 20, 1471-1476.
16. Wani, A.H., R.M.R. Branion, and A.K. Lau. 1997. Biofiltration: A promising and cost-effective control technology for odors, VOCs and air toxics. *J. Environ. Sci. Health* 32, 2027-2055.
17. Worsey, M.J. and P.A. Williams. 1975. Metabolism of toluene and xylenes by *Pseudomonas putida* (arvilla) mt-2: Evidence for a new function of the TOL plasmid. *J. Bacteriol.* 124, 7-13.
18. Yudelson, J.M. and P.D. Tihari. 1995. Economics of biofiltration for remediation projects. In R.E. Hinchee, R.S. Skeen, and G.D. Sayles (eds.), Biological Unit Processes for Hazardous Waste Treatment. Battelle Press, Columbus, OH.