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Characterization of a biofilter treating toluene contaminated air

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Abstract

The removal of toluene from an experimental gas-stream was studied in an industrial biofilter filled with poplar wood bark. Toluene degradation, approximately 85% through the operating period, resulted in low levels of toluene in the off-gas effluent. For a toluene load of 6.7 g m^{-3} h^{-1} the elimination capacity of the biofilter was found to be 6.0 g m^{-3} h^{-1}. Toluene removal was due to biodegradative activity of microorganisms in the filter bed; the most probable number counts of toluene degraders increased from 2.4×10^2 to 6.4×10^7 MPN/g dry packing material in about seven months of air-toluene supply. The degradative capacity of a *Burkholderia (Pseudomonas) cepacia* strain, isolated from the biofilter material, as an example of the effectiveness of microbial toluene removal was tested in batch culture. The microorganism degraded completely 250 ppm of toluene supplied as sole carbon source in 24 hours. The high performance demonstrated for a long period and the mechanical and physico-chemical stability of the biofilter favour its use in industrial full-scale off-gas control.

Introduction

Biofiltration is an attractive technique for the elimination of low concentrations of volatile organic compounds and for control of malodorous gases and vapour emissions from waste air mixtures. Biological filters have drawn attention in view of lower operation costs as compared with physical or chemical processes in treating off-gases with low concentrations of contaminants. The biofiltration process has been developed to remove odorous compounds such as hydrogen sulphide, dimethyl sulphide and now the application has been extended to the removal of many easily biodegradable volatile compounds (Fukuyama & Honda 1976; Hirai et al. 1990; Leson & Winer 1991; Groenestijn van & Hesselink 1993). Arcangeli & Arvin (1992), Shareefdeen & Baltzis (1994), Pedersen & Arvin (1995), Weber & Hartmans (1995) recently reported on the kinetics of toluene degradation in aerobic biofilters. The removal of waste gases and volatile chemicals can also be realised by biofiltration of the gas streams formed during on-site treatment of air stripper off-gas from contaminated soil and groundwater treatment processes (Douglass et al. 1991).

Materials used for biofiltration processes require a good air permeability to prevent high pressure drops, high specific surface area, good wetting characteristics and good sorptive capacity. Packing materials for traditional biofilters consisted of soil or peat/compost (Pomeroy 1982). Among different available filling materials, wood bark may offer suitable characteristics as it gives low back pressure and is cheaper in costs (v. Langenhove et al. 1986). This work was carried out to investigate the removal of toluene from an air toluene stream in an industrial biofilter by indigenous microorganisms naturally present in poplar wood bark utilised as filling material. We report the overall performance of the biofilter system and the count and activity of the indigenous microflora involved in metabolising toluene and other related aromatic compounds such as benzene and isomers of xylene during a long running period. Finally, we report on the isolation and identification of a strain capable of growing on toluene and benzene as sole carbon and energy source.

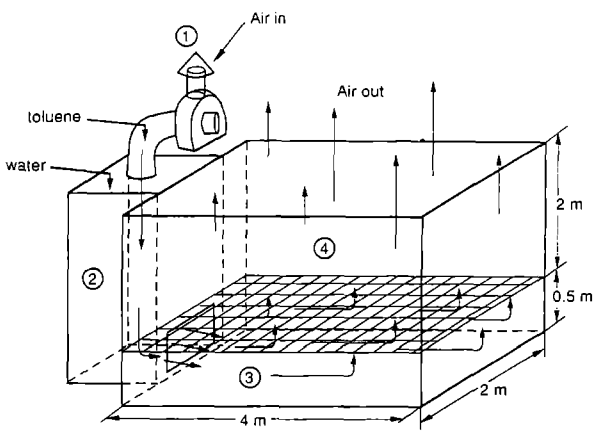

Figure 1. Biofilter scheme: 1. Air inlet; 2. Air toluene mixing and humidifing chamber (fulfilled with Rashing-rings); 3. Diffusion chamber (inlet of biofilter); 4. Wood bark, biofilter material.

Table 1. Experimental conditions

Operation period	1994, July-1995, June (324 days) toluene was supplied from 120th day
Packing material	Wood bark (C% 45; N% 0.72; P% 0.10; K% 0.52; NH_4 106 ppm; Ash% 19)
Packing material density (kg/l)	0.6
Filter bed moisture content (%)	80
pH	9.5
Initial cell number of toluene degraders (MPN/g dry wood barks)	4.4×10^2
Packing height (m)	1
Packing volume (m^3)	15
Air flow rate (m^3/h)	2400
Space velocity (h^{-1})	160
Operating pressure (mm H_2O)	10
Retention time (seconds)	25

Materials and methods

Experimental set up

A scheme of the biofilter is shown in Figure 1. The filter material was packed in a specifically designed steel container with a bed volume of 15 m^3. The wood bark had a different size distribution of the material with an apparent density of 0.6 kg l^{-1}. The air supply system for inducing toluene flow through the column was designed to provide air with a constant contaminant concentration and to achieve a high degree of humidity (95%). The toluene vapour was produced by bubbling a constant regulated air flow through liquid pure toluene. To maintain a fixed toluene air concentration, the bubbling cylinder was equipped with a water bath thermostatic chamber.

This study was carried out from July 1994 through June 1995. After filling the biofilter with wood bark, the filter was fed with air without toluene, for about 90 days, in order to aerate the packing material and to check air permeability. Subsequently, the biofilter was fed with air–toluene mixtures. Nutrients were never added to the bed. After a preliminary adaptation phase, lasting about 20 days, in which toluene was present in the air-stream at a low concentration (3 mg m^{-3}) to avoid poisoning of microflora, toluene concentration was gradually increased to about 40 mg m^{-3}.

The details of the experimental conditions are summarised in Table 1.

Sampling and analytical methods

Sampling of the air stream was periodically carried out at three points of the biofilter: the inlet air-stream was sampled at three ports located in the air distribution system at a distance of a few centimetres from the bottom of the filter bed; the outlet air-stream was sampled at the external surface of the filter bed. For each sampling 10 l of air-stream was adsorbed, by a portable air pump, on Dräger tubes containing 300 mg of activated carbon and subsequently eluted with 3 ml of carbon sulphide and sonicated for 15 min. One µl of the solution was then injected into a Hewlett-Packard 5890 series II gas chromatograph and toluene was determined by a microbore DB-5 capillary column, 90 m length, 0.53 mm diameter, 1.5 µm thin film (J & W), maintained isothermically at 150°C (carrier gas He at a flow rate of 8 ml/min).

The amount of adsorbed toluene to biofiltering material was determined on 5 g of triturated packing material plugged in a 20 ml air-tight flask and placed into a stove at 90°C for 2 hours. 250 µl of head space air was then gas chromatographically analysed.

For measurements of pH, dry weight (d.w.), number of microorganisms and microbial ATP content, packing material was sampled at three different sites longitudinally located, at a 50 cm depth, corresponding

to half of its thickness. Five hundred grams of this material was drawn from each site and was triturated to obtain large lumps of about 5 cm and the required measurements were carried out. For pH measurements, 10 g of triturated material was homogenised with 90 ml of sterile water for 15 min, and after 30 min of gravitational settling the pH value was measured using a pH meter. The pressure drops across the filter bed were measured with a water manometer. The temperature was measured at 30 and 60 cm below the surface of the biofilter. For the determination of dry weight , 20 g of triturated material was placed in a stove at 105°C for 24 hours to reach constant weight. The dried material was then cooled in a dessicator and weighed.

Enumeration of bacteria

One hundred grams of triturated material (w.w.) was mixed with 900 ml of physiological solution (9 g/l NaCl) and incubated on a rotary shaker for 2 hours. Duplicates of 10-fold dilutions of this suspension were plated on the solid medium or inoculated in the appropriate liquid medium. The count of heterotrophic bacteria was carried out using Plate Count Agar (PCA), Difco (Detroit, Mich. USA) as medium after incubation for 5 days at 30°C. To determine the number of toluene, benzene and isomers of xylene degrading bacteria, duplicates of dilutions were inoculated into minimal medium M9 (Kunz & Chapman 1981) supplemented separately with 250 mg/l of each aromatic compound, as sole carbon and energy source, and incubated at 30° C in an atmosphere enriched with the appropriate compound. After 14 days, visible turbidity and microscopic observations indicating bacterial growth were recorded and the number of bacteria was expressed as MPN/d.w. of sample. Controls were prepared with sterile medium inoculated with the microbial dilutions and without aromatic compound. To determine the number of nitrifiers (NH_4^+ oxidisers) duplicates of dilutions were inoculated into liquid Stanier medium (Pochon & Tardieux 1962). The presence of nitrite was evidenced colorimetrically with Griess-Ilosvay's reactive after 26 days of incubation at 30°C in the dark using a test kit Aquaquant 14424, (Merck, Darmstadt, Germany). The tubes in which nitrite was detected were counted for enumeration and the number of bacteria was expressed as MPN/d.w. of sample.

Determination of cellular ATP content

Cellular ATP content of all microbial species present in the biofilter material was enzymatically assayed. Thirty to fourty grams of triturated material (w.w.) was left to dry overnight under laminar flow and then was finely ground. One gram of ground material was added with 10 ml of a sulphuric acid phosphate solution (0.5 M H_2SO_4, 0.25 M Na_2HPO_3) and was vigorously shaken for 15 min. An aliquot of 50 μl of the suspension was neutralised with 1.5 ml of a 25 mM Tris −0.4 mM EDTA solution, pH 7.5. 100 μl of the solution was mixed with 100 μl of an ATP releasing reagent (NRM) and shaken gently for 30 s. Then 100 μl of luciferin-luciferase was added to the mixture and the ATP content was measured after integration for 15 s in a Lumac M 1500 Biocounter (Lumac). The ATP content was read in Arbitrary Luminous Units (A.L.U.) and then converted in μg ATP/g d.w. The results were read from an internal standard curve, which was used to compensate for interfering factors. The internal standard curve was prepared by adding different ATP concentrations to the wood bark suspensions and then treating them in the same way as the samples.

Isolation of toluene and xylene degraders

One ml of cultural broth where growth occurred was serially diluted and seeded on PCA. After 5 days of incubation at 28°C the colonies appearing on the plate were isolated. The strains were maintained both on PCA agar and minimal medium M9 supplemented with 250 mg/l of the appropiate compound. The strains were characterised on the basis of their morphological and physiological characteristics using an API 20 NE system (Bio Merieux, Montalieu, Vercieu, France).

The capability of these strains to grow on various aromatic compounds was tested by inoculating the strains in liquid minimal medium M9 added with 250 mg/l of appropriate aromatic compound as sole carbon and energy source. For growth experiments on toluene and benzene, eighteen 30 ml vials, each containing 10 ml of liquid minimal medium M9 added with 250 mg/l either of toluene or benzene were inoculated with 1 ml of culture of a strain isolated and pregrown on the same compound. The vials were sealed with teflon- rubber stoppers and incubated at 30°C for 9 days. Growth was tested at intervals, by observing turbidity and by measuring the disapearance and degradation of the compound. Duplicate vials were analysed at sampling point. Controls were performed to assess

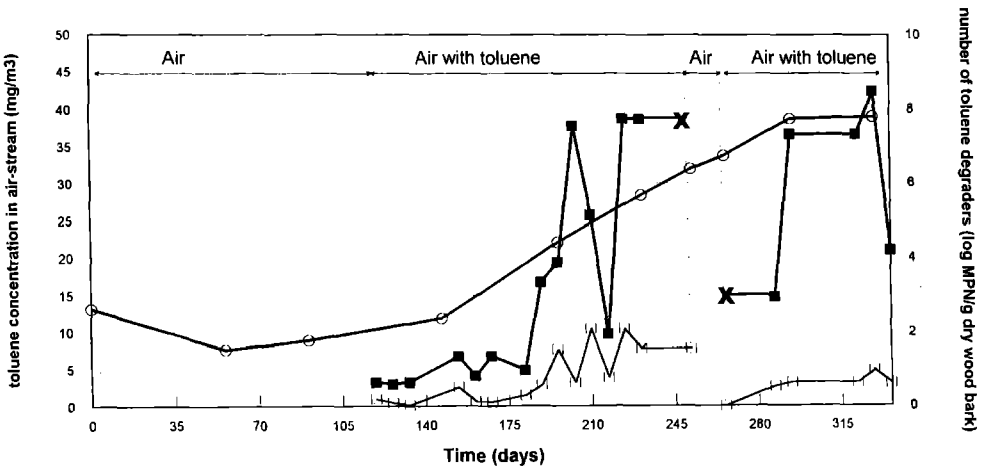

Figure 2. Time-dependence of toluene removal: ■ Inlet air–toluene concentration; □ outlet air–toluene concentration; ○ number of toluene degrading bacteria. The points (X) indicate a toluene concentration not determined but presumed as the inlet concentration was kept at the level fixed.

abiotic degradation and vials tightness. Benzene and toluene concentrations were gas chromatographically determined by analysing head-space samples of the vials.

Determination of cis,cis-muconic acid

The determination of *cis, cis*-muconic acid, intermediate in the *ortho*-clevage of benzene by bacteria was performed by HPLC after solid-phase anion exchange of cultural medium (disposable SAX column 3 ml capacity; packed with 500 mg of strong anion-exchange sorbent, quaternary anion). The acid eluate was injected (Octadecyl dimethyl sylil column 5 × 0.40 cm I.D., 3 μm particle size) and eluted isocratically using a mobile phase consisting of formic acid-tetrahydrofuran-water (14:17:969; v/v) at a flow rate of 3 ml/min. *Cis-cis* muconic determination was performed recording UV response of eluate at 263 nm and by comparison with *t-t* muconic acid aqueous solution as reference.

Chemicals

Toluene, benzene and xylene (purity > 99%) were obtained from Merck (Dasrmstadt, Germany). All other chemicals were of reagent grade.

Results and discussion

Biofilter efficiency in toluene removal

The time dependence of the toluene concentration determined at the inlet and outlet air-stream and of the number of toluene degrading bacteria are reported in Figure 2. Toluene removal was always high through the operation period resulting in low levels of toluene in the off gas. The removal was performed by toluene degraders as documented by the significant increase of their cell number that ranged from 2.4×10^2 to 6.4×10^7 MPN/g dry wood bark when the toluene concentration supplied to the biofilter passed from 7 to 43 mg m $^{-3}$. The amount of toluene removed by absorption processes was always negligible.

Fluctuations in air–toluene concentrations did not influence the activity of the microorganisms which were effective in biodegradation even when toluene concentrations in air supplied increased from 10 to 40 mg m^{-3} in one day. The elimination capacity of the biofilter (the amount of toluene degraded per volume of filter bed per hour) versus toluene load is plotted in Figure 3. The relationship seems to be approximately linear (R^2 = 0.96), indicating a first order reaction rate. The biofilter immediately achieved a good elimination efficiency of approximately 85% which remained constant for a seven-month period. At a toluene concentrations in air up to 45 mg m^{-3}, the elimination capacity of the biofilter is a function of the inlet gas concentration; with a toluene load of 6.7 g m^{-3} h^{-1},

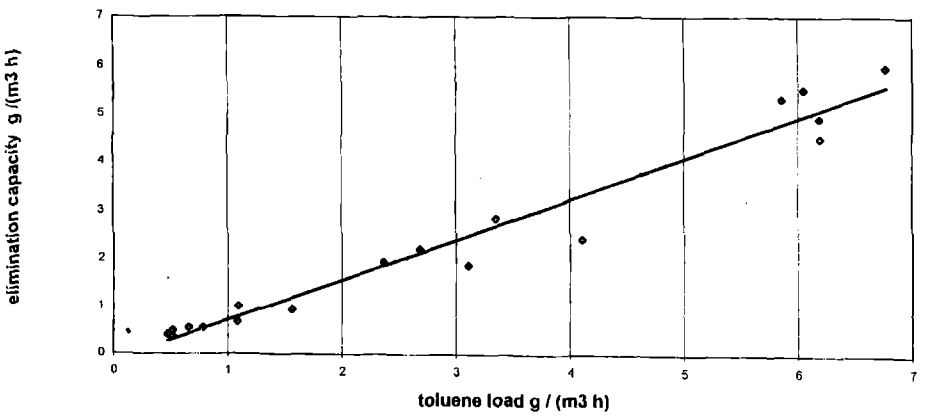

Figure 3. Elimination capacity of biofilter versus toluene load: Slope β = 0.85; intercept = -0.14.

corresponding to inlet gas concentration of 43 mg m^{-3}, it was 6.0 g m^{-3} h^{-1}.

Higher toluene loads were never supplied because our aim was to study, with an industrial biofilter, the removal of toluene in concentrations that are frequently encountered in soil and groundwater. Higher concentrations of contaminant can be treated more effectively and cheaply by other systems (Dragt 1992). Consequently, with the concentrations tested, it was not possible to determine the maximum removal capacity of the industrial biofilter differently from what has been reported by Ottengraf and Van den Oever (1983) and Liu et al. (1994). The former authors, when treating in laboratory scale columns mixtures of ethylacetate, butylacetate, butanol and toluene at varied concentrations, found a linear relationship between degradation rate versus toluene concentration up to 1 g m^{-3} and a zero order kinetics for concentrations above 3 g m^{-3} and observed a removal capacity ranging from 20 to 30 g m^{-3} h^{-1} at a whole load of 90 g m^{-3} h^{-1}. The latter found an elimination capacity of around 20 g m^{-3} h^{-1} with 90% removal efficiency using laboratory scale activated carbon biofilters to treat an air stream having a toluene concentration ranging from 0.05 to 0.1 g m^{-3}. If a linear relationship between removal capacity versus toluene concentration is assumed up to 1 g m^{-3}, the performance of the industrial biofilter would be higher than that reported in the other two studies mentioned.

Removal capacities of the same magnitude were found by other researchers studing toluene removal from waste gases in laboratory trickling filters (Wolff 1992; Schindler et al. 1994 and Pedersen & Arvin 1995). Shareefdeen & Baltzis (1994) recently proved that a laboratory biofilter, modelled both for steady-state and transient operations, was capable of achieving 100% removal of toluene, given enough residence time.

Changes of pH, temperature and pressure in the biofilter

The pH in the biofilter declined very slowly by passing from about 9.5 to about 8.5 in five months and then remained constant. This decrease was probably due to the oxidation of NH_4^+, released by the wood bark, by nitrifiers, whose cell number increased (Figure 4) and to the production of acids by all the other microorganisms of the biofilter. The temperature inside the biofilter was always almost equal to that outside and followed the seasonal trend, ranging between 5 and 25°C. The pressure drop was only of 5 mm of water during the operation period.

Enumeration of bacteria

The microflora present in the packing material was periodically determined during the operation period. Figure 4 shows the number of toluene degrading bacteria as well as *o-*, *m-*, and *p*-xylene degrading bacteria and nitrifier bacteria. Wood bark appeared to own a high number of heterotrophic bacteria (3.4×10^9 CFU/g dry wood bark) and among these toluene degraders, *m-*, *o-*, and *p*-xylene degraders were present. The number of indigenous toluene degraders (4.4×10^2 MPN/g dry wood bark), of *m-* and *p-* xylene degraders and

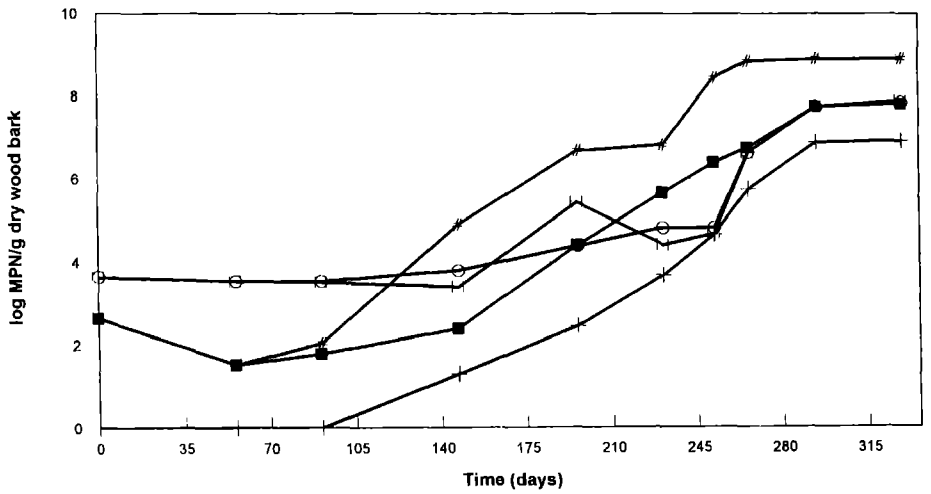

Figure 4. Time dependence of microorganism number monitored during the operation period: ■ toluene degraders; □ *m*-xylene degraders; o *p*-xylene; + *o*-xylene; # nitrifiers. Data points represent the average of three determinations.

of nitrifiers was low but their presence was sufficient for a rapid formation of a microflora capable of biodegrading toluene and consequently the biofilter did not require any inoculation.

With the beginning of toluene supply, the colonisation by toluene degraders occurred and cell number increased by seven orders of magnitude, ranging from 6.2×10 to 6.4×10^7 MPN/g dry wood bark. Their substantial increase occurred concomitantly with the highest toluene concentrations supplied (30–40 mg m^{-3}). Toluene degraders survived for a fairly long period when the filter bed was not loaded with toluene and there was no loss of microbial activity for 15 days (Figure 2). The number of microorganisms capable of degrading isomers of xylene, and of nitrifiers also increased and followed the trend of toluene degraders. These aromatic compounds are easily degraded by soil microorganisms and several strains of *Pseudomonas* are capable of degrading both toluene, *m*- and *p*-xylene (Smith 1990). Nitrifiers, besides being capable of oxidising ammonia, can oxidise benzene to phenol and hydroxylate toluene by ammonia monooxygenase (Hyman et al. 1985).

The number of CFUs of viable heterotrophic bacteria was always high in wood bark unexposed and exposed to toluene and, compared with the initial ones, their cell number remained constant, 10^8 CFU/g. Therefore the increase of toluene degraders may be significant enough to indicate that a selection of microorganisms which superseded the heterotrophic population initially present took place.

Table 2. ATP biomass concentration

Running time (days)	ATP (μg/g d.w)	Recovery[1] (%)
0	0.17	40
	0.24	47
92	1.43	89
	1.84	82
145	1.53	79
	3.50	85
	2.17	62
194	1.84	80
	2.70	85
	2.56	61
231	1.95	82
	3.06	56
	2.01	70

[1] % recovery of ATP, calculated as (sample + standard ATP) - (sample) × 100 / (standard ATP measured in the extract.)

ATP biomass concentration

Table 2 shows the ATP biomass content values. The ATP content of wood bark biomass used to fill the filter and kept without aeration was very low (0.17 μg/g dry wood bark) and this value was not considered significant because of the low level of recovery calculated.

Table 3. Growth characteristics of strains isolated

Strain	Growth with given compound as sole carbon source				
	Toluene	*m*-xylene	*p*-xylene	*o*-xylene	Benzene
B1 (*P. fluorescens*)	+	-	-	-	-
B2 (*Micrococcus sp.*)	+	-	-	-	-
B3	-	-	-	+	-
B4 (*B. cepacia*)	+	-	-	-	+
B5	-	-	+	-	+
B6 (*P. pudita*)	+	+	-	-	-
B7 (*P. putida*)	+	+	-	-	-
B8 (*P. putida*)	+	+	-	-	-
B9 (*P. putida*)	+	+	-	-	-

Growth was monitored qualitatively (increase in absorbance of >0.5 compared with controls at 600 nm) in liquid culture after 10 days incubation at 30°C.
+, good growth.
-, absence of growth.

With the beginning of aeration of the biofilter, the ATP biomass content determined was found to be higher. The ATP content increased when the biofilter was fed with air–toluene mixtures, then it remained constant for the period monitored, suggesting that the microorganisms could be in or close to the stationary phase with respect to their metabolism. ATP biomass content was never determined during the 15 days in which no toluene was supplied and therefore ATP content could not be utilised as bioindicator of toluene degrading activity.

Toluene and xylene degraders and their substrate ranges

Different strains were isolated from bacterial cultures and some of these are reported in Table 3. The strains were oxidase-positive and Gram-negative rods with the exception of B2 and B5, which were oxidase-negative and Gram-positive cocci. Most strains isolated were identified as *Pseudomonas* (Bergey 1984), one as *Burkholderia cepacia*, formerly classified in the Bergey's Manual as *Pseudomanas* and one as *Micrococcus* (Bergey 1986). The *B. cepacia* strain isolated was chosen to demonstrate the effectiveness of microbial removal of toluene. The degrading capability of this microorganism towards benzene was also investigated. This microorganism degraded toluene and benzene when separately supplied as the only carbon and energy source in one and five days, respectively. The degradation of benzene was accompanied by the formation of *cis,cis*- muconate which was detected in low amounts in cultural broths (data not shown), suggesting that after an initial attack by catechol 1,2-dioxygenase, the benzene ring was completely degraded. Toluene degradation, instead, was not accompanied by any metabolite formation.

Conclusions

Experimental data gathered for almost one year clearly demonstrate that the full-scale biofilter can operate successfully and steadily in removing low levels of toluene from an air-stream over a seven-month period. No nutrients were supplied to the biofilter, which remained active and exhibited a low pressure drop, around 5 mm of water column over the period, confirming the good air permeability charateristic of the wood bark. This packing material also proved to be useful for the contribution of microorganisms capable of degrading toluene. Our results demonstrate that the selected toluene degrading bacteria removed this pollutant with high efficiency and seem to suggest that the biofilter does not lose microbial activity during the 15 days in which no toluene was supplied. This is based on the MPN's of toluene degraders which actually increased slightly during this period. Unfortunately, ATP biomass levels were not determined during this period and also toluene removal was not determined directly after the toluene supply was restored. The absence of any loss of microbial activity observed for these 15 days means that the biofilter can be active in a short

time after periods of inactivity and that this system can be used for discontinuous operations.

Finally, the adaptation of the microbial population to structurally related aromatic chemicals plays an important role in the application of this biofilter for the elimination of toluene, benzene and xylene polluted air streams.

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References

Arcangeli JP & Arvin E (1992) Toluene biodegradation and biofilm growth in a aerobic fixed-film reactor. Appl. Microbiol. Biotechnol., 37: 510–517

Bergey's Manual of Systematic Bacteriology (1984) Section 4. NR Krieg, JG Holt (Eds). Baltimore, The Williams & Wilkins Co.

Bergey's Manual of Systematic Bacteriology (1986) Sections 12. Snearth PHA, Mair NS, Sharpe ME, Holt JG (Eds). Baltimore, The Williams & Wilkins Co.

Douglass RH, Armstrong JM & Korreck WM (1991) Design of a packed column bioreactor for on-site treatment of air stripper off-gas. In: Hinchee RE & Olfenbuttel RF (Eds) On-site Bioreclamation, Process for Xenobiotic and Hydrocarbon Treatment (pp 209–225). Batelle Memorial Institute, Columbus, Ohio, Butterworth-Heinemann.

Dragt AJ (1992). In: Dragt AJ, Van Ham J (Eds) Biotechniques for Air Pollution Abatement and Odour control Policies (pp 3–9) Elsevier, Amsterdam.

Fukuyama J & Honda A (1976) Removal of malodorous gases containing aromatic hydrocarbons by activated sludge. Taiki Osen Kenkyu. 11: 386

Groenestijn Jw & Hesselink GM (1993) Biotechniques for air pollution control. Biodegradation 4: 283–301

Hirai M, Ohtake M & Shoda M (1990) Removal kinetics of hydrogen sulfide, methanethiol and dimethyl sulfide by peat biofilter. J. Ferment. Bioeng. 70: 334–339

Hyman MR, Sansome-Smith AW, Shears JH & Wood PM (1985) A kinetic study of benzene oxidation to phenol by whole cells of *Nitrosomonas europea* and evidence for further oxidation of phenol to hydroquinone. Arch. Microbiol. 143: 302-306

Kunz DA & Chapman PJ (1981) Catabolism of pseudocumene and 3-ethyltoluene by *Pseudomonas putida (arvilla)* mt-2: evidence for new function of the TOL (pWWO) plasmid. J. Bacteriol. 146: 179–191

Leson G & Winer AM (1991) Biofiltration: an innovative air pollution control technology for VOC emissions. J. Air Waste Manage. Assoc. 41: 1045–1054

Liu PKT, Gregg RL, Sabol HK & Barkley N (1994) Engineered biofilter for removing organic contaminants in air. Air & Waste 44: 299–303

Ottengraf SPP & van den Oever AHC (1983) Kinetics of organic compound removal from waste gases with a biological filter. Biotechnol. Bioeng. 25: 3089–3102

Pedersen AR & Arvin E (1995) Removal of toluene in waste gases using a biological trickling filter. Biodegradation 6: 109–118

Pochon J & Tardieux P (1962) Techniques d'Analyse en Microbiologie du Sol. In: De la Tourelle, St. Mandè (Eds) Collection 'Techniques de Base' (pp 59-62)

Pomeroy RD (1982) Biological treatment of odorous air. J. Wat. Pollut. Control Fed. 54: 1541–1545

Schindler I & Friedl A (1995) Degradation of toluene/heptane mixtures in a trickling-bed bioreactor. Appl. Microbiol. Biotechnol. 44: 230–233

Shareefdeen Z & Baltzis BC (1994) Biofiltration of toluene vapor under steady-state and transient conditions: theory and experimental results. Chem. Engng. Science. 49: 4347–4360

Smith MR (1990) The biodegradation of aromatic hydrocarbons by bacteria. Biodegradation 1: 191–206

Van Langenhove H, Wutys E and Schamp N (1986) Elimation of hydrogen sulphide from odorous air by a wood bark biofiltration. Wat. Res. 20: 1471–1476

Weber FJ & Hartmans S (1995) Use of activated carbon as a buffer in biofiltration of waste gases with fluctuating concentrations of toluene. Appl. Microbiol. Biotechnol. 43: 365–369

Wolff F (1992) Biologische abluftrenigung mit einem intermittierend befeuchteten tropfkörper. In: Dragt AJ & van Ham J (Eds) Biotecniques for Air Pollution Abatement and Odour Control Policies (pp 49-62). Elsevier Science Publishers B.V.