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Toluene vapour removal in a laboratory-scale biofilter

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Abstract A bench-scale biofilter with a 0.5-m high filter bed, inoculated with a toluene-degrading strain of *Acinetobacter* sp. NCIMB 9689, was used to study toluene removal from a synthetic waste air stream. Different sets of continuous tests were conducted at influent toluene concentrations ranging over 0.1–4.0 g m^{-3} and at superficial gas velocities ranging over 17.8–255 m h^{-1}. The maximum volumetric toluene removal rate for the biofilter (242 g m^{-3} h^{-1}) was obtained at a superficial gas velocity of 127.5 m h^{-1} (corresponding to a residence time of 28 s) and a toluene inlet concentration of 4.0 g m^{-3}. Under these operating conditions, toluene removal efficiency was only 0.238, which suggested that effective operation required higher residence times. Removal efficiencies higher than 0.9 were achieved at organic loads less than 113.7 g m^{-3} h^{-1}. A macro-kinetic study, performed using concentration profiles along the bioreactor, revealed this process was limited by diffusion at organic loads less than 100 g m^{-3} h^{-1} and by biological reaction beyond this threshold.

Introduction

The release of large quantities of aromatic compounds into the environment has created serious problems. In particular, benzene, toluene and the isomers of xylene are among the 50 largest-volume industrial chemicals produced in the world, at a rate in the order of millions of tonnes per year (Smith 1990; Zilli and Converti 1999). Due to their high volatility and slight solubility in water, they are widespread environmental pollutants which are often present in soil, ground water and atmosphere. These compounds are quoted by the US Environmental Protection Agency as priority environmental toxic pollutants, due to their substantial toxicity and carcinogenic potential, even at low concentrations.

The control of volatile organic toxic emissions into the air from industrial facilities has become critical. It is expensive for the chemical industries, particularly the small-sized ones, to meet the increasingly severe quality standards. The biotechnological approach to air pollution is now a promising field of research which can supply reliable, simple and cheap technologies for the prevention of air contamination.

The reliability of biological processes and in particular biofiltration for the treatment of waste gas streams containing volatile organic compounds (VOCs) has been demonstrated by a very large number of experimental studies (Ottengraf and van den Oever 1983; Ottengraf 1986; Shareefdeen et al. 1993; Zilli et al. 1993; Mpanias and Baltzis 1998; Zilli and Converti 1999). Biofiltration is particularly suited and cost-effective for the treatment of high volumes of waste gases containing low VOC concentrations. Furthermore, it is environmentally friendly because the contaminants are completely converted at low temperature into non-hazardous final products.

Compared with other biological systems, biofilters have been shown to be more effective for treating some compounds poorly soluble in water, thanks to the high superficial area available for mass transfer. Optimal removal of aromatic air contaminants can best be achieved using biofilters rather than bioscrubbers or biotrickling filters (van Groenestijn and Hesselink 1993). Provided comparable performance can be obtained with similarly sized biofilters and biotrickling filters, both the simplicity and the ease of operation of biofilters favour their selection for the treatment of waste gases.

As is well known, toluene is highly volatile (with a Henry's Law coefficient of 0.26; Amoore and Hautala 1983) and is poorly soluble in water (water solubility range 515–627 g m^{-3}; Mackay and Shiu 1981). Moreover, toluene is a widely used solvent (in the production of paints and coatings, gums, resins and rubber) and

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reagent (in the production of drugs, dyes and perfumes), whose adverse effects on health are well documented (Dean 1985). As a consequence, the American Conference of Governmental Industrial Hygienists has set the following threshold limit values (TLVs) for the concentration of this compound in air: (1) the time weighed average (TWA) is 0.375 g m^{-3}, (2) the short time exposure level is 0.560 g m^{-3} and (3) the olfactory threshold value is 8.8×10^{-3} g m^{-3} (Guelfo et al. 1987).

Although the biofiltration of toluene has been the subject of a great deal of research in the past decade and the microorganisms responsible for degradation (mainly Eubacteria belonging to the genus *Pseudomonas*) and their possible degradative pathways have been identified (Smith 1990; Choi et al. 1992; Chang et al. 1993; Duetz et al. 1994; Mirpuri et al. 1997), only in one case, at least to the authors' knowledge, has *Acinetobacter* sp. been used for the treatment of toluene-containing waste gases. This was in a consortium of five bacteria and two yeasts (Acuña et al. 1999). Therefore, the main objective of the present work was to investigate the ability of a biofiltration system inoculated with an *Acinetobacter* sp. strain to remove toluene vapour; and consequently to obtain information useful for applicative purposes. Toluene concentration profiles along the bioreactor were also studied in order to perform a macro-kinetic study on this removal process. Finally, the conditions necessary for the deodorisation of the gas were experimentally determined at various operating conditions.

Materials and methods

Bacterial strain and culture conditions

A toluene-degrading strain of *Acinetobacter* sp. NCIMB 9689, obtained from the National Collections of Industrial and Marine Bacteria, Aberdeen, Scotland, was used in this work. The strain was grown on a nutrient medium containing the following chemicals, per litre of tap water: 1,000 mg Lab-Lemco beef extract, 2,000 mg yeast extract, 5,000 mg peptone and 5,000 mg NaCl. The medium was sterilised by autoclaving at 121 °C for 15 min. Cultivation was performed in 250-ml Erlenmeyer flasks containing 150 ml medium at pH 6.8. The culture was then incubated at 25 °C with rotary shaking and was aerated with sterile air. After 24–48 h growth, the cells were harvested by centrifugation at 5,000 rpm for 20 min and re-suspended in fresh nutrient medium to be used as an inoculum for biodegradation experiments. Stock cultures of the strain were maintained by periodic subculture on the same nutrient medium and were stored at 4 °C.

Analytical procedures

Toluene concentration in air streams was analysed with a Carlo Erba Model HRGC 5160 gas chromatograph equipped with a capillary column (25 m × 0.32 mm, Mega Laboratory) and a flame ionisation detector (FID) connected to a computing integrator. The temperatures of injector and detector were 150 °C and 200 °C, respectively. Oven temperature was initially maintained at 50 °C for 2 min and then increased at a rate of 40 °C min^{-1} to 250 °C, where it remained constant for 5 min. Nitrogen was used as carrier gas. Air samples (0.5 ml) were injected into the FID gas chromatograph with a 1.0-ml gas-tight syringe and the toluene concentration was quantified by comparison with standards.

Cell mass concentration for inoculum preparation was determined by filtering 10 ml of a thick cell suspension through tared cellulose nitrate membrane filters (47 ± 3 mm diameter, with 0.45 μm pores; Sartorius, Goettingen, Germany). After washing with 10 ml 0.9% NaCl solution, the filters were then dried at 105 °C to constant weight and cooled in a desiccator prior to re-weighing.

Moisture content of the filter bed was determined by using the dried weight method (A.P.H.A. 1985).

Apparatus for continuous biofiltration experiments

The experimental apparatus, previously described in more detail (Zilli et al. 1996), consisted of a 0.65-m cylindrical column, made of glass, with an inner diameter of 0.05 m. It was provided with sampling ports, located at 0.12, 0.25, 0.37 and 0.52 m from the bottom of the filter, in order to follow the toluene concentration profile along the column. The height of the biofilter bed was 0.50 m.

The filter packing material was a mixture of sterilised peat (with a specific superficial area of 1.6 m^2 g^{-1}) and glass beads (diameter 5 mm) in a 4:1 volume ratio. It was supported by a finely perforated ceramic plate placed at the bottom of the column. The moisture content of the packing material was kept at 50–60%, either by bubbling the influent synthetic waste air through a humidification unit, or by periodically using a nozzle at the top of the packing material to distribute a mineral salts solution, flowing countercurrent with the gas rising through the column. The solution had the following composition: 5,800 mg KH_2PO_4, 4,500 mg K_2HPO_4, 2,000 mg $(NH_4)_2SO_4$, 340 mg $MgCl_2 \cdot 6H_2O$, 20 mg $CaCl_2 \cdot 6H_2O$, 2 mg $FeSO_4 \cdot 7H_2O$ and 1.6 mg $MnCl_2 \cdot 4H_2O$ in 1 l tap water.

The biofilter column was operated at room temperature (20–21 °C). The pH of the packing material was kept around neutrality, as previously described (Zilli et al. 1996). The biofilter was inoculated with a cell suspension of *Acinetobacter* sp., mixed with dry packing material, which had previously been sterilised by autoclaving at 120 °C for 20 min. The proportion was 25% by volume and the cell density in the inoculum was 6.0 g l^{-1}.

The synthetic waste gas was generated by injecting a low-flow laboratory-scale air stream into a liquid toluene reservoir, the stream becoming polluted by toluene evaporation. The air containing toluene vapour was then adequately mixed with a high-flow clean air stream, previously humidified by bubbling it through a water container. In this way, the desired concentration of toluene in the air stream was obtained. The system was provided with two flow-meters which allowed the measurement of both the total air flow and the separate streams flowing in the vessels. Toluene concentration in the influent gas was varied by regulating, via the flow-meters, the flows of both contaminated and clean air streams in a mixing chamber.

Results

Continuous biofiltration of toluene

Acinetobacter sp. NCIMB 9689 was selected for continuous tests of toluene removal in the biofilter mainly because of the lack of information about its application and its peculiar ability to survive in consortia and to dominate other ubiquitous strains in activated sludge (Converti et al. 1993).

In order to test the elimination capacity of the system, different sets of continuous experiments were made over a period of 10 months, by changing the superficial gas velocity and the toluene concentration in the influent air stream. In particular, four series of experiments were performed at 17.8, 35.7, 127.5 and 255 m h^{-1} and, for each given superficial gas velocity, testing five different inlet toluene concentrations, namely 0.1–0.2, 0.4–0.5,

1.0, 2.0 and 4.0 g m^{-3}. Sampling at different dimensionless filter heights (0.25, 0.50, 0.75 and 1.00) allowed the determination of the concentration profiles of toluene along the biofilter, under all tested conditions. All experiments were performed under non-sterile conditions. During the start-up phase, a control column was used in order to determine the initial abiotic removal due to physical adsorption by the packing material. Toluene breakthrough occurred after 3 h operation.

Figure 1 shows the experimental results of continuous tests carried out during the whole experimental schedule. Each test, under a given set of operating conditions, lasted about 10–12 days. In order to prevent any shock to the microorganisms, the first set of experiments was performed at the lowest superficial gas velocity (17.8 m h^{-1}) and at the lowest range of inlet toluene concentration (0.1–0.2 g m^{-3}). In this period, a removal efficiency close to 100% was initially observed, which was followed by a gradual decrease and a final restoration of the high starting values. This behaviour is the result of the preliminary adsorption of the pollutant by the filter bed and of the subsequent biological action of the microorganisms.

Nearly constant values of degradation efficiency were reached in about 3 days continuous operation at the lowest concentration range and the highest residence time. This short start-up period was probably due to the inoculation of the biofilter with a specific and adapted microorganism. A final average degree of conversion close to 100% was assured, indicating this biofiltration system was a really efficient technique for the control of waste gases containing toluene at low concentrations.

After the efficiency of the system had been tested at low pollutant levels, the inlet toluene concentration was progressively increased to 0.4–0.5, 1.0, 2.0 and 4.0 g m^{-3}, while the residence time was decreased from 202 s to 14 s, with the aim of evaluating the actual possibility of employing this system in the presence of variable and high toluene loads in waste gases.

The average degree of conversion calculated in these subsequent phases was quite high and the system proved very stable during the whole continuous experimentation. This also indicated the high capacity of the microorganism to adapt itself to large variations in pollutant concentration.

Deodorisation tests

In the last part of this study, gas deodorisation was considered, i.e. finding the operating conditions necessary to obtain an outlet toluene concentration lower than 8.8×10^{-3} g m^{-3}. Tests were performed by changing either the toluene concentration in the inlet waste gas, up to 2.0 g m^{-3}, or the superficial gas velocity in the range 17.8–255 m h^{-1}.

Discussion

Continuous biofiltration tests

The experimental results presented in the previous section are here utilised to study the macro-kinetics of the toluene biofiltration process, which are the consequence of the complex interplay between physical factors, such as mass transfer phenomena and residence time distribution of the gas flow, and biological reactions.

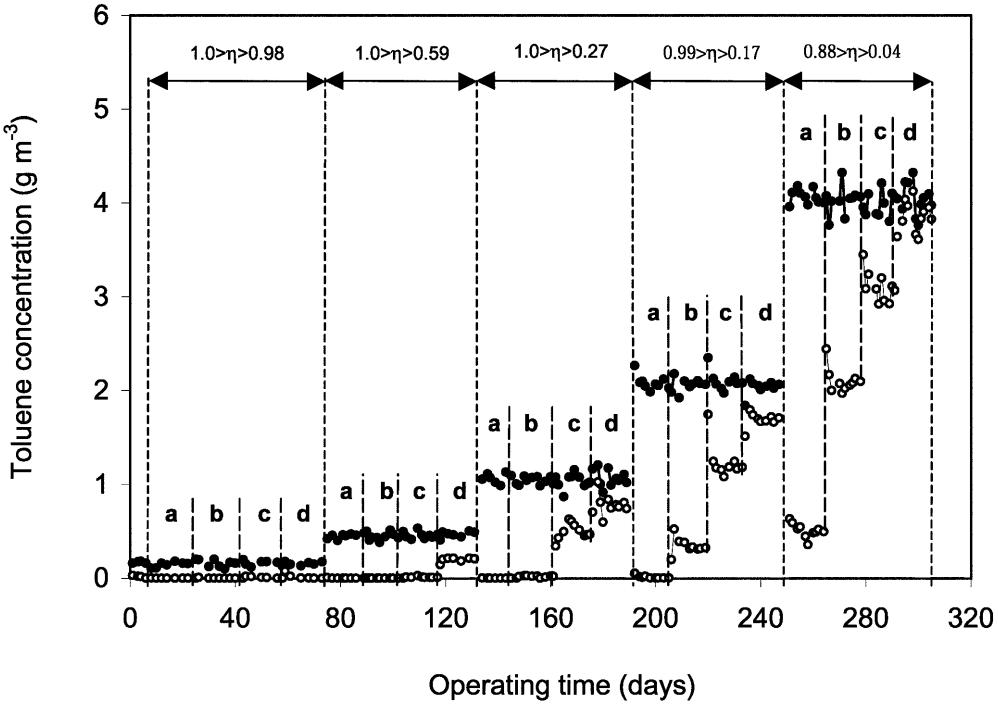

Fig. 1 Biofilter behavior during continuous experiments of toluene removal. Toluene concentration (g m^{-3}): ● inlet, ○ outlet. Superficial gas velocity, U_{go} (m h^{-1}): *a* 17.8, *b* 35.7, *c* 127.5, *d* 255.0

Effects of superficial gas velocity and toluene inlet concentration

The combined effects of the superficial gas velocity and of the inlet toluene concentration on the biofilter removal capacity are evident in Fig. 2. The ratio between the outlet and inlet toluene concentrations is reported (for five different ranges of inlet concentrations) as a function of the ratio between the minimum superficial gas velocity (17.8 m h^{-1}) and the one considered in each experiment ($U_{g,min}\ U_g^{-1}$); i.e. a dimensionless inverse residence time. Each experimental value corresponds to the average of about ten experimental runs with inlet toluene concentration equal to the reported nominal value $\pm 5\%$. As expected, these results on the whole clearly show that the biofilter removal efficiency ($1 - C_{ge}/C_{go}$) decreases with increasing either U_g or C_{go}.

Toluene concentration profiles along the column

A second series of tests was carried out to evaluate the concentration profiles along the column. Two different situations were possible, according to the most common models, in the case where K_s was negligible in Monod's equation (zero-order kinetics): (1) no toluene diffusion limitation in the wet biofilm, implying a fully active biofilm and a reaction limitation, and (2) diffusion limitation in the wet biofilm, indicating a non-fully active biofilm. A different situation would occur when the concentration of pollutant in the liquid film was negligible with respect to K_s (first-order kinetics). In this case, biofiltration would be controlled by reaction. The equation proposed by Ottengraf (1986) for first-order macro-kinetics could then be used:

$$\ln(C_{ge}/C_{go}) = -HK_1/m_i U_g$$

where K_1 is an apparent first-order parameter, m_i is the distribution coefficient of toluene and H is the filter bed height.

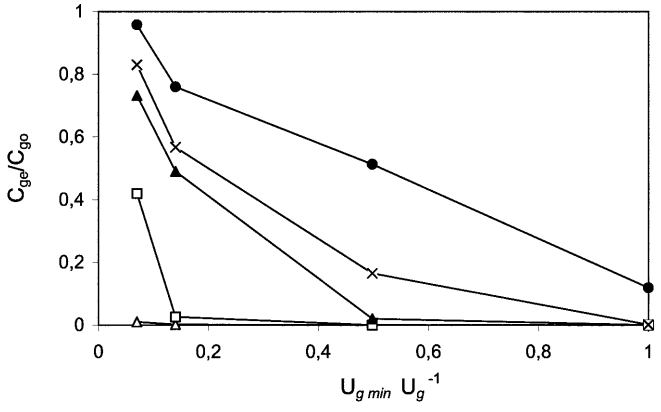

Fig. 2 Toluene gas concentration profiles as a function of the superficial gas velocity, obtained at different inlet toluene concentrations, C_{go} (g m^{-3}): △ 0.1–0.2, □ 0.4–0.5, ▲ 1.0, × 2.0, ● 4.0

Using the experimental results from our continuous tests, this equation offers a good correlation along the entire filter height, over the whole range of concentrations investigated (Fig. 3), however this is not a sufficient

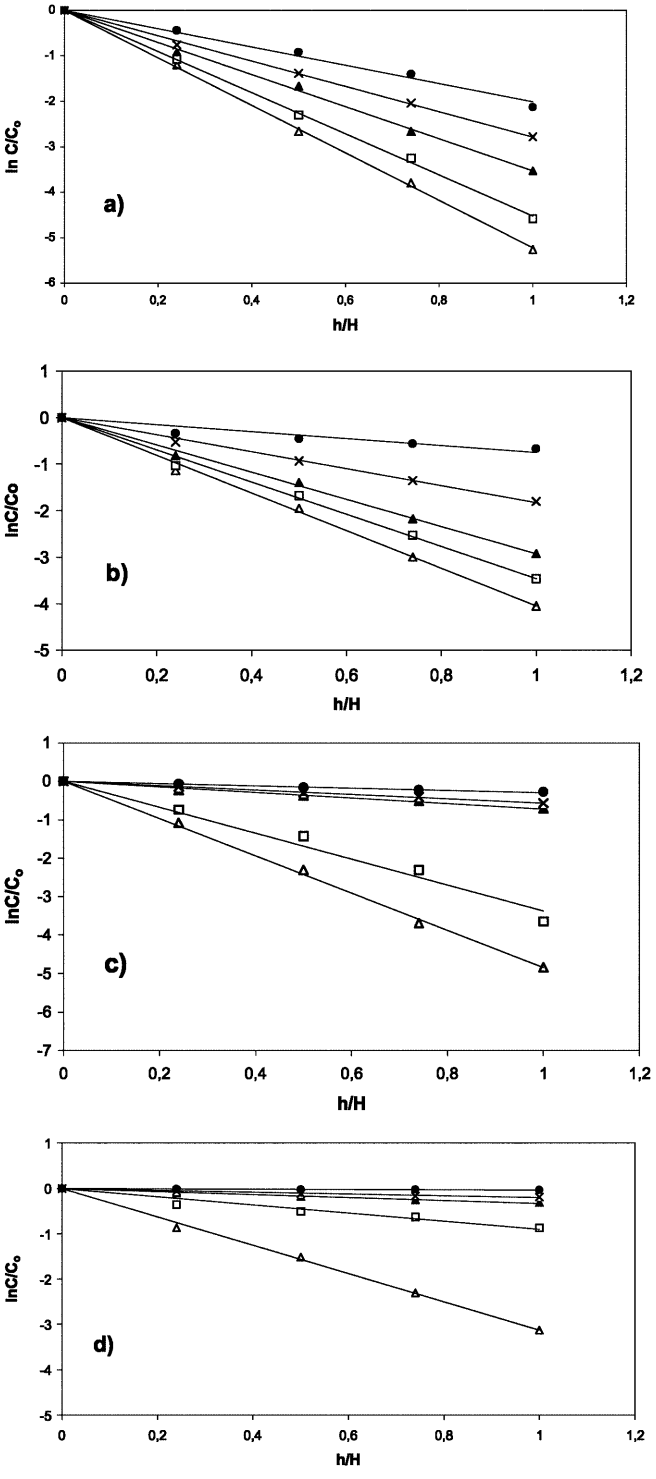

Fig. 3 Toluene concentration profiles along the column at different superficial gas velocities and inlet toluene concentrations, U_{go} (m h^{-1}): **a** 17.8, **b** 35.7, **c** 127.5, **d** 255.0; C_{go} (g m^{-3}): △ 0.1–0.2, □ 0.4–0.5, ▲ 1.0, × 2.0, ● 4.0

proof that first-order kinetics actually applied. It could be observed that, at a fixed height of the filter bed, the higher the superficial gas velocity, the higher the outlet dimensionless concentration, or in other words, the lower the degree of conversion.

Under steady-state conditions, the degree of conversion at a residence time of about 200 s (Fig. 3, case a) was always higher than 0.85 over the entire range of inlet toluene concentrations tested in this study. But at the lowest residence time (14 s; Fig. 3, case d), a satisfactory removal efficiency was only ensured at the lowest range of inlet toluene concentration (0.1–0.2 g m^{-3}). Similar trends for concentration profiles along the column were detected in the biofiltration of other VOCs, dichloromethane and phenol (Bohn 1992; Ergas et al. 1994; Zilli et al. 1996).

Linear kinetics were more evident at higher superficial gas velocities, which corresponded to lower degrees of conversion. In fact, at the lowest superficial gas velocity, a relevant column section was practically inactive in toluene elimination. In other words, the column was oversized.

From the resulting linear trends evident in Fig. 3, it was also possible to use the above equation to estimate the values of the pseudo first-order parameter, K_1, which are listed in Table 1.

In theory, up to a critical value of the superficial gas velocity, the macro-kinetics of the process should be controlled by the feed velocity or by the inlet pollutant concentration (diffusion limitation). At higher values of the superficial gas velocity, the macro-kinetics of the process should be reaction-limited. Therefore, no influence on K_1 could be ascribed to the substrate feed rate, unless inhibition phenomena related to excess substrate could take place. Taking these considerations into account, an empirical linear dependence of K_1 on the superficial gas velocity was assessed using a previous work for phenol degradation (Zilli et al. 1996), through which a critical value of 80 ± 4 m h^{-1} was estimated for U_g. However, the behaviour of K_1 versus load depicted in Fig. 4 shows that K_1 not only increases with the specific gas velocity, but is also a decreasing function of toluene concentration. For this reason a critical U_g value should be estimated for each value of C_{go}.

However in the case of bioreaction limitation ($U_g > U_{g,crit}$), K_1, which corresponds to a sort of specific degradation rate, should reach maximum threshold values, depending on the influent toluene concentration.

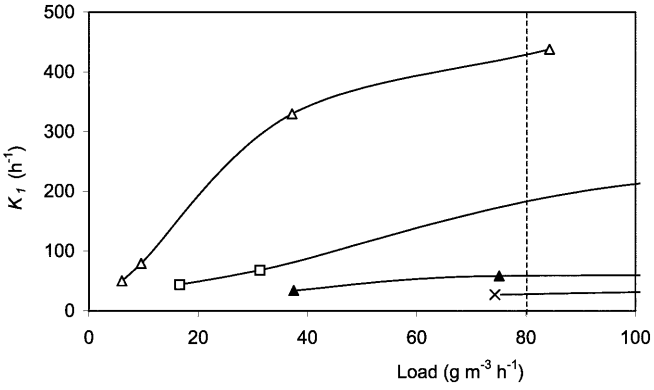

Fig. 4 Dependence of the pseudo first-order kinetic parameter, K_1, on the inlet toluene concentration, at a given organic load, C_{go} (g m^{-3}): △ 0.1–0.2, □ 0.4–0.5, ▲ 1.0, × 2.0

The values in Table 1 also show that the threshold value at the lowest inlet toluene concentration could be around 450 h^{-1}, corresponding to $U_{g,crit} > 255$ m h^{-1}. But at higher C_{go} values, an evident decrease in K_1 takes place at $U_g > U_{g,crit}$, thus showing a clear substrate inhibition phenomenon which is responsible for a completely different behaviour with respect to phenol. Threshold K_1 values estimated for the different inlet toluene concentrations tested in this study were about 450, 223, 54, 37 and 19 h^{-1}, respectively.

These K_1 threshold values decreasing with C_{go} suggest that, over a certain value of inlet toluene concentration, the biological reaction becomes the rate-limiting step instead of diffusion. In fact, since the diffusivity should be nearly independent of inlet toluene concentration, the observed variability of K_1 threshold values can only be ascribed to the biological reaction rate variations. The rate of diffusion should become limiting only at very low gas phase concentrations, which are not of interest for this work. Similar decreases of maximum specific degradation rate with increasing inlet substrate concentration (C_{go}) were experimentally observed for other pollutants, like phenol (Zilli et al. 1996) and in other biological processes, e.g. alcohol fermentation (Ciftci et al. 1983).

The good removal capacity of the biofilter is demonstrated in Fig. 5, where the elimination capacity is plotted versus the organic load at different inlet toluene concentrations. From these results, it is evident this parameter increases regularly with the organic load, with little relevance whether such a dependence is the result of

Table 1 Calculated values (and r^2) of the pseudo first-order kinetic parameter, K_1 (h^{-1}) for different levels of inlet toluene concentration (C_{go}) and superficial gas velocities (U_g)

C_{go} (g m^{-3})	U_g (m h^{-1})			
	17.8	35.7	127.5	255.0
0.1–0.2	48.36 (0.999)	75.17 (0.996)	320.90 (0.998)	413.84 (0.997)
0.4–0.5	41.90 (0.999)	64.21 (0.994)	223.09 (0.977)	119.92 (0.939)
1.0	32.66 (0.998)	54.28 (0.996)	47.65 (0.984)	43.81 (0.980)
2.0	25.80 (0.998)	33.95 (0.995)	37.45 (0.998)	27.22 (0.962)
4.0	18.63 (0.989)	13.94 (0.849)	19.22 (0.985)	5.86 (0.981)

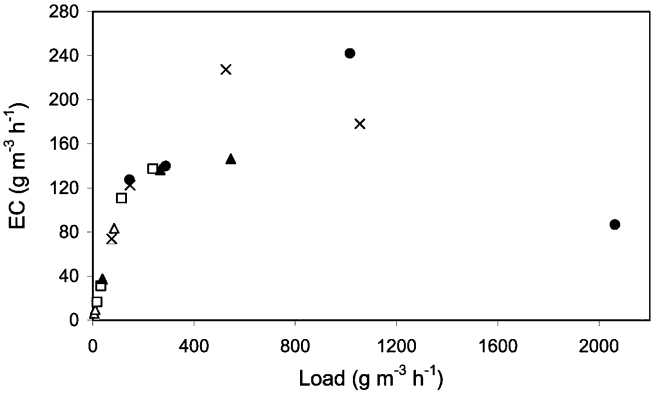

Fig. 5 Toluene elimination capacity (*EC*) of the biofilter versus the organic load, C_{go} (g m^{-3}): △ 0.1–0.2, □ 0.4–0.5, ▲ 1.0, × 2.0, ● 4.0

an increase in C_{go} or a decrease in residence time. In particular, a gradual and linear increase of the elimination capacity occurs, up to a value of the organic load corresponding to about 100 g m^{-3} h^{-1}. Beyond this value, the elimination capacity increases more slowly and reaches a maximum threshold value around 230–240 g m^{-3} h^{-1} at inlet toluene concentrations ranging over 2.0–4.0 g m^{-3} and a superficial gas velocity of 127.5 m h^{-1}. Such a behaviour of the elimination capacity indicates that, at low loading rates, there is a linear relationship between the removal rate and the inlet load, and that toluene is nearly completely removed. In this range of the organic load, the system performance is limited only by pollutant availability, i.e. by the diffusion. But with further increase of the load, the elimination rate increases more slowly up to a critical load at which it keeps constant, indicating that the maximum elimination capacity of the biofilter is achieved. Under these last conditions, the limiting step of the process is the biological reaction, the elimination rate being lower than the toluene feed rate. At loads higher than 1,000 g m^{-3} h^{-1}, toluene becomes inhibitory and the elimination capacity falls down.

Many experimental studies on the removal of toluene from air streams were performed with conventional biofilters and biotrickling filters. A comparison of our data with those available in the literature for laboratory-scale peat biofilters shows that the maximum elimination capacity obtained in the present work is about 10–21% higher than the best values reported by Morales et al. (1998): 190 g m^{-3} h^{-1} and Acuña et al. (1999): 215 g m^{-3} h^{-1}. Nevertheless, extreme care should be taken when modifying real-world plans on the basis of these findings. Further studies on biofiltration of toluene vapours were performed by Ottengraf and van den Oever (1983), Morales et al. (1994) and Zarook et al. (1998), which reported maximum elimination capacities of 57.1, 20 and 25 g m^{-3} h^{-1} with laboratory-scale biofilters packed with peat.

Experimental results for toluene degradation with biotrickling filters were also reported by various investigators. Pedersen and Arvin (1997) compared various studies and found that toluene elimination capacities ranged over 12–84 g m^{-3} h^{-1} at loadings ranging over 12–220 g m^{-3} h^{-1} and residence times from 1.2 s to 4 min. Weber and Hartmans (1996) obtained an average elimination capacity of 27 g m^{-3} h^{-1} over a period of 375 days with a trickle-bed reactor inoculated with a fungal culture. Sorial et al. (1995) and Arcangeli and Arvin (1992) presented maximum elimination capacities between 68–112 g m^{-3} h^{-1} and 25–45 g m^{-3} h^{-1}, respectively. Cox and Deshusses (1999) presented a maximum elimination capacity of 83 g m^{-3} h^{-1} for a biotrickling filter enriched with protozoa. The best results with this reactor configuration were obtained by Laurenzis et al. (1998), who obtained a maximum elimination capacity of 275 g m^{-3} h^{-1} (higher than that obtained in this study) with a trickle-bed reactor with discontinuous movement of the packed bed and intermittent percolation.

Finally, it must be stressed that outlet toluene concentrations below the TLV-TWA (0.375 g m^{-3}; Guelfo et al. 1987) were achieved at a loading rate of toluene up to 71 g m^{-3} h^{-1}, corresponding to an inlet toluene concentration of 2.0 g m^{-3} and a superficial gas velocity of 35.7 m h^{-1}.

Deodorisation tests

The average results of deodorisation obtained under different conditions are summarised in Table 2, where the effect of superficial gas velocity on the maximum inlet pollutant concentration (at which the gas deodorisation is obtained) is shown together with the toluene removal rates, calculated at the different superficial gas velocities. It can be observed that the value of the maximum inlet toluene concentration at which the deodorisation of the gas is ensured, decreases sharply with increasing superficial gas velocity. This is a consequence of the decrease in the removal efficiency of the biofilter with decreasing residence time. The maximum toluene concentration at which the outlet pollutant concentration was reduced below the olfactory threshold value (8.8×10^{-3} g m^{-3}) was 1.99 g m^{-3}; and this was obtained at the lowest superficial gas velocity. This seems to be a very interesting result, if one considers that biofiltration is usually considered a particularly effective technique for treating gaseous emissions containing contaminants in relatively low concentrations (< 1.0 g m^{-3}).

Table 2 Operating conditions under which the deodorisation of the waste gas was achieved

C_{go} (g m^{-3})	U_g (m h^{-1})	Loading rate (g m^{-3} h^{-1})	Removal rate (g m^{-3} h^{-1})
1.99	17.8	70.8	70.5
1.02	35.7	72.8	72.2
0.31	127.5	79.0	76.8
0.25	255.0	127.5	123.0

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