

Biofilter response to biomass reactivation for VOC treatment

ANA ELÍAS, ASTRID BARONA, GORKA GALLASTEGI, MIKEL LARRAÑAGA
AND MARÍA FERNÁNDEZ

*Department of Chemical and Environmental Engineering, Faculty of Engineering,
University of the Basque Country, Alda Urquijo s/n. 48013 Bilbao, Spain*

ABSTRACT

This research has undertaken a comparative study on using a fresh activated sludge or a refrigerated/reactivated sludge as active biomass source for biofiltration purposes. A sludge sample was initially selected based on the ratio between volatile solid content and total solid content before and after refrigeration at 6 °C for 90 days. The degradation rate of the activated sample for three addition doses of toluene was established before and after refrigeration. The same procedure was also carried out for ethylbenzene and p-xylene after refrigeration/reactivation. Surprisingly, the degradation rate for toluene was higher after refrigeration and the results were very similar for an addition of 2 and 8 µL. Subsequently, one biofilter was inoculated with the activated sample and another with the reactivated sample, and both were fed with toluene ranging from 2.6 to 26.2 g toluene m⁻³ h⁻¹. Concerning the elimination capacity of both biofilters, no relevant differences were found. It was concluded that the active biomass degrading toluene was not affected by refrigeration, in spite of the fact that the SV/ST ratio decreased after the storage period. The elimination capacity of the other two biofilters (ethylbenzene and p-xylene) was highly influenced by the gas flow rate.

1 INTRODUCTION

Environmental regulations for pollution control are frequently enacted before «suitable» (affordable, effective and environmentally friendly) technologies have been fully developed. Amongst biotechnologies, biofiltration is a seemingly simple system whose effectiveness relies on the optimization of several operating parameters and the selection of a suitable packing material and degrading biomass.

The biomass responsible for the degradation of the gaseous contaminant fed into the biofilter can be supplied by the support material itself (Barona *et al.*, 2004), purchased from trade catalogues (Christen *et al.*, 2002), isolated from other active bioreactors (Estévez *et al.*, 2004) or even collected from locations contaminated with the target pollutant (González-Sánchez and Revah, 2007).

Prior to the inoculation itself, the proper selection, storage, acclimation and activation of the microorganisms is crucial to ensure a long-lasting operation of the bioreactors. In fact, Prado *et al.* (2005) proved that the previous biomass concentration and biomass adaptation of the inoculum dramatically affected the start-up and performance of conventional biofilters treating methanol during the first stages of operation.

Likewise, simple analytical techniques for biomass growth detection are needed. Several methods, such as extra-cellular enzymatic activity (Laurent and Servais, 1995) or electron transportation activity (Fontvieille and Moul, 1985), were developed by microbiologists to estimate biomass activity. Among other methods, respirometry, optical density at 600 nm, plate counting methods and 4'6-diamidino-2-phenylindole (DAPI) staining have also been used in literature (Hwang *et al.*, 2003; Álvarez-Hornos *et al.*, 2005; Kim and Jaffé, 2007). Other simpler analyses, such as carbon balance and volatile suspended solid content, are also practical tools for assessing biomass concentration and adaptation, although they have obvious disadvantages, such as no discrimination between living or dead biomass or no accounting for changes in microorganism physiology.

Bearing in mind that biofilter controlling operators require simple and quick techniques for start-up and everyday operation, the objective of this study is to ascertain the influence of using a previously refrigerated and reactivated sample (a stored sample) as inoculum to set-up several biofilters. Likewise, the relevance of certain simple parameters for achieving biomass activity will be studied. The pollutants to be treated in the biofilters were toluene, ethylbenzene and p-xylene.

2. MATERIALS AND METHODS

2.1 SOURCE OF THE MICROORGANISMS AND MEDIA COMPOSITION

Three sludge samples were collected in a wastewater treatment plant (W), in a small river close to a petrochemical company (P) and near a synthetic resin-producing industry (F) in Bizkaia (Spain).

After sedimentation of the solid phase of each sample for 2 hours, the liquid phase was transferred into glass bottles for further experimentation. A nutrient medium (Barona *et al.*, 2007) was used for enriching and maintaining the cultures.

2.2 BIOMASS STORAGE AND ACTIVATION

About 1 L of the supernatant phase of each sample was mixed with 1 L of the nutrient solution, and the total volume was transferred into 3 litre vessels that were continuously fed with toluene (inlet concentration ranging from 50 to 100 ppmv for 1000 hours at a gas flow rate of 2.5 L min⁻¹). When this procedure concluded, the activated samples were stored at 6 °C for 90 days. The samples were subsequently warmed to room temperature (20 °C) and the reactivation procedure was then repeated for a further 1000 hours.

2.3 BIOMASS RECOVERY IN BATCH ASSAYS

Two different parameters were used to compare biomass recovery before and after refrigeration and storage. The first parameter was the ratio between volatile solid content and total solid content (VS/TS), and it was basically used to select the best sample for subsequent experiments.

After the preliminary selection of one of the samples based on VS/TS data, the second step was to determine degradation rates in order to ascertain the influence of refrigeration and storage lag on the selected sample activity.

The originally activated sample was transferred into a hermetic vessel and 2 µL of toluene were added. The removal of the contaminant was monitored over time until the complete disappearance of the pollutant. Likewise, two consecutive doses of 4 and 8 µL of toluene were also added to determine the degradation rate of the biomass prior to inoculation.

The same procedure was repeated with the reactivated sample (after refrigeration), and in this case, toluene, ethylbenzene and xylene were individually added in the respective hermetic vessels. After the addition of 2, 4 and 8 µL of each pollutant, the degradation rate was monitored over time until the complete disappearance of the compound.

2.4 BIOFILTER SETUP

The packing material used for filling four biofilters was made up of composted pig manure and sawdust and has already been used in previous work (Elías *et al.*, 2002). The outline of the pilot plant for each of the four biofilters has already been described in detail (Moura *et al.*, 2006). The first biofilter (biofilter OT) was inoculated with the original activated sample (without refrigeration and for toluene degradation) and the other three were inoculated with the refrigerated/reactivated sample, although the contaminant to be degraded was toluene, ethylbenzene and p-xylene (biofilter T, biofilter E and biofilter X, respectively).

The inlet loading rate for the three contaminants ranged from 2.6 to 29.1 g m⁻³ h⁻¹. Two different gas flow rates were used during experimentation; the first one was 1 L min⁻¹ and the second one was 2 L min⁻¹, corresponding to a residence time of 180 and 90 s, respectively. Temperature was constant at 23 °C.

2.5 ANALYTICAL METHODS

Toluene, ethylbenzene and p-xylene were measured in a micro gas chromatograph (microGC CP 4900) equipped with auto-sampling injection mode and a TCD detector. Operating conditions were: injector temperature, 110 °C; column/transfer temperature 80 °C.

3. RESULTS AND DISCUSSION

The original three samples were activated for 1000 hours, and after a 90-day refrigeration and storage period, they were reactivated for a further 1000 hours. The evolution of the biomass during these activation assays was monitored by measuring the changes in the ratio between the volatile solid content and total solid content (VS/TS). The results, shown in Table 1, revealed that none of the three samples was able to fully recover initial values of the controlling parameter after tempering. In all cases, the VS/ST ratio decreased by about 20% after storage. Nevertheless, the sample from the wastewater facility (W) showed the highest VS/TS ratio before (85) and after refrigeration (65). Consequently, W sample was preliminary selected as the best inoculum for subsequent operation, although further analyses were carried out to ascertain the degradation rate before and after storage.

Table 1.

The VS/TS ratio for the three activated samples before and after refrigeration.

	100 x VS/TS		
	W sample	P sample	F sample
Original	28	21	20
After 1000 hours of activation	85	60	72
After refrigeration and 1000 hours of reactivation	65	48	58

The degradation rate before and after refrigeration for the toluene activated sample is shown in Table 2. The originally activated W sample achieved a relatively constant degradation rate for the addition of 4 µL of toluene (211 ppmv h⁻¹) and for 8 µL (222 ppmv h⁻¹). Surprisingly, the degradation rate was higher after refrigeration and very similar for an addition of 2 and 8 µL (291 and 280 ppmv h⁻¹, respectively). Consequently, the decrease in the VS/TS ratio after refrigeration for W sample was not considered to be a decisive result (Table 1). Thus, the ratio reached after storage was high enough for the biomass to recover activity.

Table 2.

Degradation rate for the originally activated and refrigerated/reactivated W sample.

Sample type	Compound	Degradation rate (ppmv h ⁻¹)		
		Dose		
		2 μL	4μL	8μL
Originally activated	Toluene	66	211	222
Refrigerated/reactivated	Toluene	291	213	280
Refrigerated/reactivated	Ethylbenzene	263	258	526
Refrigerated/reactivated	p-Xylene	194	233	204

The degradation rate for ethylbenzene and p-xylene is also shown in Table 2. The results for p-xylene revealed that the biomass in the refrigerated sample was able to degrade this pollutant at a rate ranging from 194 to 233 ppmv h⁻¹, in spite of the contaminant dose. The refrigerated sample also showed a high capacity to degrade ethylbenzene, above all for the highest dose. The degradation rate was fairly constant for the addition of 2 and 4 μL, but it doubled when 8 μL of ethylbenzene were added. As an example, the remaining amount of ethylbenzene after the addition of 4 μL is plotted in Figure 1.

Those preliminary results suggested that the refrigeration of W sample did not stop the biomass degrading the three simple alkylbenzenes. Furthermore, the highest degradation rate was reached for ethylbenzene with an addition dose of 8 μL.

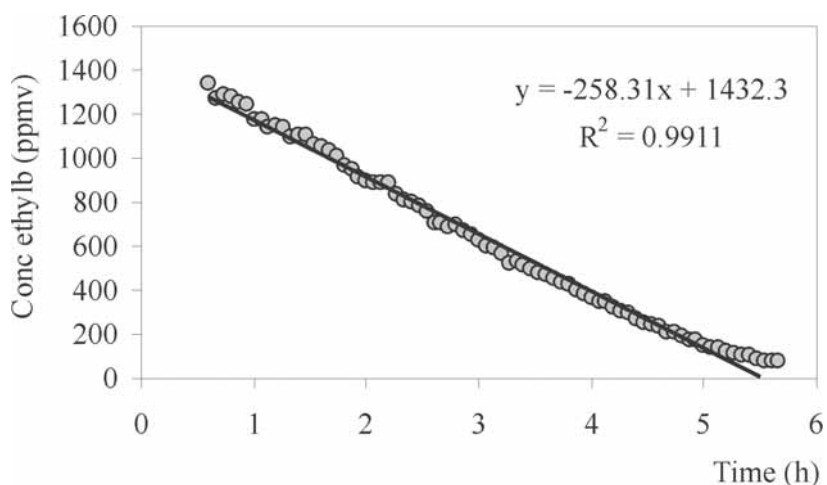


Figure 1. Evolution of the remaining concentration of ethylbenzene along time after an addition of 4 μL (refrigerated/reactivated sample).

In order to ascertain the behaviour of W sample (originally activated and refrigerated/reactivated) as inoculum for biofiltration purposes, four biofilters were started-up. Two of them were fed with toluene (biofilters 0T and T) and the other two (biofilters E and X) were fed with ethylbenzene and p-xylene, respectively. The inoculation of biofilter 0T was carried out with the originally activated W sample and the inoculation of the others involved the refrigerated and reactivated W sample. In all cases, two flow rates of 1 and 2 L min⁻¹ were tested.

The comparison of the response of biofilters 0T and T rendered similar results and, consequently, only the data for biofilter T have been plotted in Figure 2. It was concluded that the active biomass degrading toluene was not affected by the refrigeration, in spite of the fact that the VS/TS ratio reduced after the storage and reactivation period.

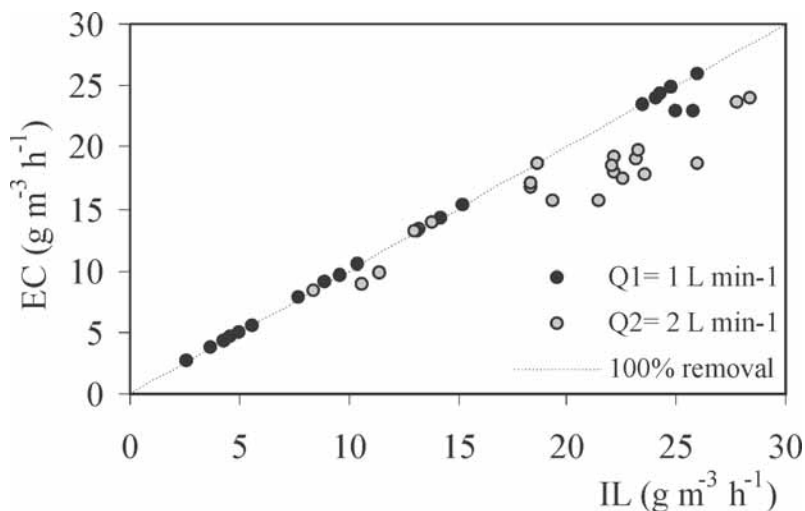


Figure 2. Response of Biofilter T treating toluene (inoculation with the refrigerated/reactivated sample).

As shown in Figure 2, when the inlet loading rate (IL) ranged from 2.6 to 26.2 g toluene m⁻³ h⁻¹ for a residence time of 180 s (gas flow rate of 1 L min⁻¹), the removal efficiency was close to 100%. When the residence time was reduced by half (80 s) as a consequence of increasing the gas flow rate to 2 L min⁻¹, inlet load higher than 18 g m⁻³ h⁻¹ rendered lower results for elimination capacity (data plotted below the dashed line).

When the gas flow rate was 1 L min⁻¹ and the inlet load of ethylbenzene fed into biofilter E ranged from 4.8 to 28.5 g m⁻³ h⁻¹, the biomass was able to completely

degrade the contaminant. In contrast, the change to the higher gas flow rate dramatically reduced elimination capacity and biofilter response was very variable. It is remarkable that the latter result is not consistent with the data shown in Table 2, where ethylbenzene recorded the highest degradation rate. Nevertheless, this behaviour may be explained by the random attachment of biomass to the support material, which is a phenomenon for further research.

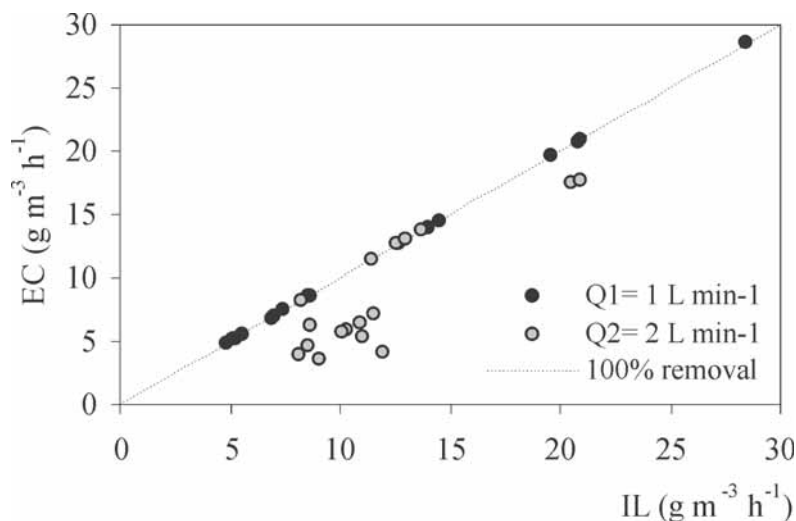


Figure 3. Response of Biofilter E treating ethylbenzene (inoculation with the refrigerated/reactivated sample).

The results obtained for the p-xylene biofilter were similar to those obtained for biofilter E, and, for brevity, are not shown.

4 CONCLUSIONS

The storage of active biomass under refrigeration (not in freezing conditions) is necessary for reproducing biofilters. In this study, the influence of a 90-day refrigeration lag was studied. Initially, the volatile solid content/total solid content (VS/TS) ratio of three activated sludge samples was measured before and after refrigeration. None of the three samples was able to fully recover initial values of the controlling parameter after tempering. Nevertheless, the sample with the largest VS/ST ratio was selected for determining the degradation rate of toluene before and after refrigeration.

Surprisingly, the degradation rate for toluene was higher after refrigeration. Subsequently, one biofilter was inoculated with the activated sample and another one with the refrigerated/reactivated sample, with both of them being fed with a toluene loading rate ranging from 2.6 to 26.2 g m⁻³ h⁻¹. It was concluded that the active biomass degrading toluene was not affected by the refrigeration, in spite of the fact the SV/ST ratio decreased after the storage and reactivation period. The elimination capacity of the biofilters for ethylbenzene and p-xylene was highly influenced by the gas flow rate, which is possibly related to the uneven attachment of biomass to the support material in these cases.

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REFERENCES

- Alvarez-Hornos, F.J., Gabaldón, C., Martínez-Soria, V., Marzal, P. and Peña-Roja, J.M. (2005) Biodegradation of ethyl acetate and toluene mixtures by a peat biofilter: Respirometry monitoring and dynamics of living and dead bacterial cells. Proceedings of the International Congress on Biotechniques for Air Pollution Control, La Coruña, Spain, pp. 413-420.
- Barona, A., Elías, A., Arias, R., Cano, I. and González, R. (2004) Biofilter response to gradual and sudden variations in operating conditions. *Biochem. Eng. J.* 22: 25-31.
- Barona, A., Elías, A., Cano, I., Uriarte, A. and Artetxe, J. (2007) Additional determinations in a biofiltration system for toluene: Adsorption and partition in the nutrient solution. *Chem. and Biochem. Eng. Q. CABEQ* 763 (in press).
- Christen, P., Domenech, F., Michelena, G., Auria, R. and Revah, S. (2002) Biofiltration of volatile ethanol using sugar cane bagasse inoculated with *Candida utilis*. *J. Hazard. Mat.* B89: 253-265.
- Elías, A., Barona, A., Arreguy, A., Ríos, J., Aranguiz, I. and Peñas, J. (2002) Evaluation of a packing material for the biodegradation of H₂S and product analysis. *Proc. Biochem.* 37(8): 812-820.
- Estévez, E., Veiga, M.C. and Kennes, C. (2004) Fungal biodegradation of toluene in gas-phase biofilters. In: Verstraete, W. (Ed.), Proceedings of the 5th European Symposium on Environmental Biotechnology. Oostende, Belgium, 25-28 April, pp. 337-340.

- Fontvieille, D. and Moul, A. (1985) Dénombrement et mesure d'activité, *Bactériologie des milieux aquatiques*, Point sur l'épuration et le traitement des effluents (eau, air): Lavoisier Ed.: 79-131.
- González-Sánchez, A. and Revah, S. (2007) The effect of chemical oxidation on the biological sulphide oxidation by an alkaliphilic sulfoxidizing bacterial consortium. *Enz. Microb. Technol.* 40: 292-298.
- Hwang, S.C.J., Lee, C.M., Lee, H.C. and Fang Pua, H. (2003) Biofiltration of waste gases containing both ethyl acetate and toluene using different combinations of bacterial cultures. *J. Biotechnol.* 105: 83-94.
- Kim, H.S. and Jaffé, P.R. (2007) Spatial distribution and physiological state of bacteria in a sand column experiment during the biodegradation of toluene. *Wat. Res.* 41: 2089-2100.
- Laurent, P. and Servais, P. (1995) Fixed bacterial biomass estimated by potential exoproteolytic activity. *Can. J. Microbiol.* 41: 749-752.
- Moura, M. (2006) Sistemática de crecimiento y seguimiento de biomasa para sistemas de biofiltración. Project 2006. School of Engineering, University of the Basque Country.
- Prado, O.J., Veiga, M.C. and Kennes, C. (2005) Treatment of gas-phase methanol in conventional biofilters packed with lava rock. *Wat. Res.* 39: 2385-2393.