

POLLUTANT BIODEGRADATION IN BIOTRICKLING FILTERS REVISITED

Huub H.J. Cox, Tran N. Nguyen, and Marc A. Deshusses¹

ABSTRACT

The contribution of biodegradation in the liquid phase to the overall elimination capacity of toluene-degrading biotrickling filters was investigated. Replacement of the original culture liquid with fresh water in order to eliminate biodegradation by the suspended culture caused a decrease of toluene removal. Biodegradation in the original culture liquid was found to be 22% of the overall elimination capacity (37.3-47.0 g/m³.h), although the amount of suspended biomass was only 1% of the amount of immobilized biomass. Calculation of specific activities indicated that suspended cells were about 20 times more active than immobilized cells. Biodegradation in the liquid phase is generally neglected. However, it may significantly contribute to the overall elimination capacity when the nutrient supply and the pollutant concentration are high. Possible implications for the modeling of biotrickling filter performance are discussed.

INTRODUCTION

Pollutant degradation in biotrickling filters for waste air treatment is a complex process involving mass transfer from the gas phase to the liquid phase and the biofilm, degradation of the pollutant by primary degraders, and secondary processes like cryptic growth and predation of primary degraders by higher organisms (Cox and Deshusses, 1998^a). Predicting the performance of such a complex system is a difficult task. Several mathematical models have been developed to describe pollutant degradation in biotrickling filters, placing the emphasis e.g. on the kinetics of pollutant removal and the influence of operational parameters (Diks and Ottengraf, 1991^{a,b}; Hartmans and Tramper, 1991; Ockeloen *et al.*, 1996; Pedersen and Arvin, 1995), on oxygen limitation (Kirchner *et al.*, 1996) and on biomass accumulation (Alonso *et al.*, 1997; Diks *et al.*, 1994). Because of the complexity of the processes simultaneously taking place, mathematical models rely on many simplifications, some of which may be specific to the particular model in question and others being more generally applied.

In this contribution we question the following assumption commonly made in the mathematical modeling of biotrickling filter performance: the liquid recycle phase in biotrickling filters is an inert phase that serves for pollutant transfer from the gas phase to the biofilm, but does not contribute to pollutant degradation. During two years of investigation of toluene-degrading biotrickling filters we regularly observed high biomass concentrations with a concomitantly high biological activity in the liquid phase. It may therefore be postulated that pollutants are not only degraded in the biofilm, but also in the liquid phase. Here we show evidence that pollutant degradation in the liquid phase indeed takes place. The relevance of the findings are discussed with respect to mathematical modeling and the operation of biotrickling filters.

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MATERIALS AND METHODS

Biotrickling filter equipment

Two identical toluene-degrading biotrickling filters (diameter 0.152 m, bed height 1.3 m) were used, filled with 2.5 cm polypropylene Pall rings and inoculated with a toluene-degrading bacterium isolated from activated sludge. The biotrickling filters had been removing toluene for more than one year at the time of the experiments. A full description of the design, equipment, operation, medium composition and history of the reactors can be found elsewhere (Cox and Deshusses, 1998^{b,c}). Standard conditions of operation were the following: volumetric gas load of $64 \text{ m}^3/\text{m}^3 \cdot \text{h}$, toluene inlet concentration of 0, 1 or $2 \text{ g}/\text{m}^3$, superficial liquid velocity of 7.9 m/h, medium supply rate of approximately 250 mL/h, and a total liquid volume of approximately 3 L (effective volume in vessel for liquid circulation of 1.8 L, dynamic liquid hold-up during these experiments of 1.2 L).

Experiment 1

In this experiment, the performance of a biotrickling filter was compared before and after replacing the original culture liquid with the same volume (determined to be 3 L) of distilled water under otherwise identical standard conditions (recycle operation). The culture liquid was allowed to drain from the reactor for 30 min to ensure complete drainage. The experiment lasted for 6.5 h, during which period toluene in the outlet gas, total carbon in the liquid, and the biological activity in the liquid were regularly determined.

Experiment 2

This experiment was done two days after the first one, allowing the biotrickling filter to recover its normal performance at standard operation. The experimental set-up was similar to that of experiment 1, except that after removal of the original culture liquid the reactor continuously received fresh distilled water (one-pass operation) instead of recycle. The superficial liquid velocity was the same, i.e., 7.9 m/h. After about one hour of one-pass operation, a volume of 32 L of fresh water was circulated from a large reservoir at the bottom of the reactor (recycle operation).

Analytical methods

Procedures for determining toluene gas phase concentrations, total carbon in the liquid phase and the wet weight of biomass in the reactor have been described previously (Cox and Deshusses, 1998^b). The biological activity of the liquid phase was determined in biological oxygen uptake rate (OUR) experiments. 2.5 mL sample was saturated with air at 25°C and the endogenous respiration was recorded for 5 min. Toluene-induced oxygen rates were determined after addition of 0.19 mM toluene and with correction for the endogenous oxygen uptake.

RESULTS

Experiment 1: recycling of 3 L water instead of 3 L of the original culture liquid

If pollutant degradation takes place in the liquid phase, then a reduction of the suspended biomass concentration should result in a decrease of the overall elimination capacity. Replacement of the original culture liquid with the same volume of water indeed caused an increase of the toluene gas outlet concentration (Figure 1), i.e., a decrease of the elimination

capacity. A constant outlet concentration was observed after about 1 h of recycling of water, presumably after the initial absorption of toluene in the liquid phase was completed. The total carbon concentration (indication of the suspended biomass concentration) was about five times lower when recycling water instead of the original culture liquid (Figure 1). Hence it is reasonable to conclude that the increase of the toluene outlet concentration and the decrease of total carbon or biomass in the liquid are correlated, as all other conditions were identical.

The toluene degradation rate in the original culture liquid can be estimated in two ways. First, the difference the overall elimination capacities while recycling the original liquid or water is a direct measure of toluene degradation in the original culture liquid (assumptions will be discussed later). Second, the toluene degradation rate in the original culture liquid can be estimated from toluene-induced OURs in the original culture liquid.

Table 1 presents an overview of the data of experiment 1. As judged from OURs, the toluene degradation rate in the liquid phase while circulating water was negligible compared to this of the original culture liquid (a reduction of 96%), although the liquid replacement resulted in a reduction of the biomass concentration as total carbon of only 79%. However, toluene degradation in the water was also reduced as result of a lower specific activity of the suspended culture (Table 1). Replacement of the original culture liquid with water caused a reduction of the overall elimination capacity of $9.7 \text{ g/m}^3 \cdot \text{h}$. Liquid phase toluene degradation while recycling water was $0.44 \text{ g/m}^3 \cdot \text{h}$, hence the toluene degradation rate in the original culture liquid was $10.1 \text{ g/m}^3 \cdot \text{h}$ as calculated from overall elimination capacities. It is interesting to note that the toluene degradation rate in the original culture liquid as estimated from OUR experiments was almost the same (Table 1). Apparently, the two methods to quantify the liquid phase toluene degradation rate, though very different, are in good agreement.

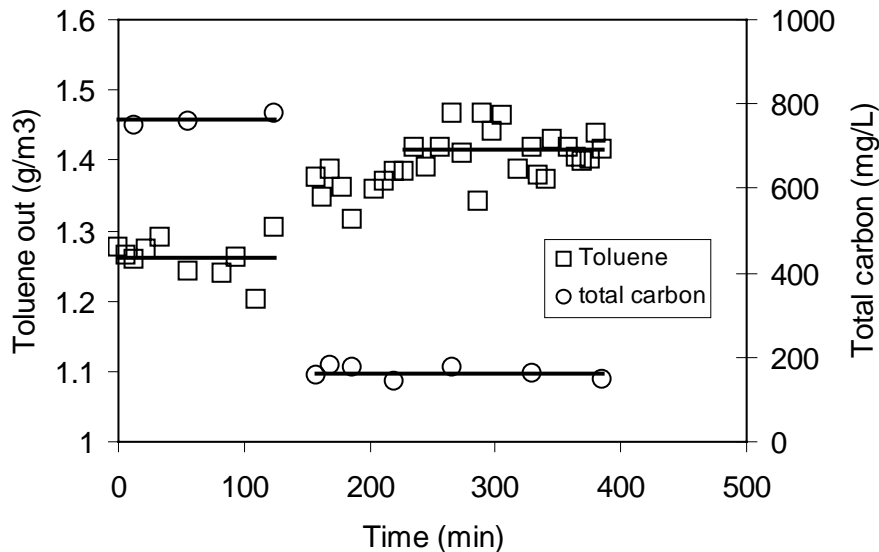


Figure 1. Effect of the replacement of original culture liquid with water on the toluene outlet gas concentration and the total carbon liquid concentration. Conditions: recycle of 3 L original culture liquid between $t = 0$ and 124 min, recycle of 3 L water between $t = 158$ and 386 min, volumetric load $64 \text{ m}^3/\text{m}^3 \cdot \text{h}$, toluene inlet concentration 2 g/m^3 , superficial liquid velocity 7.9 m/h . Solid lines show assumed steady state values.

Table 1. Overview of experimental data.

	Experiment 1		Experiment 2	
	Original culture liquid	Water	Original culture liquid	Water
<i>Parameters</i>				
Toluene in gas inlet (g/m ³)	2	2	2	2
Toluene in gas outlet (g/m ³)	1.261	1.414	1.437	1.563
Volumetric load (m ³ /m ³ .h)	64	64	64	64
Volume liquid (L)	3	3	3	32
Liquid velocity (m/h)	7.9	7.9	7.9	7.9
Suspended total carbon (mg/L)	762	162	n.d	n.d
Endogenous OUR (mg O ₂ /L.min)	0.68	0.17	0.59	0.07
Toluene-induced OUR (mg O ₂ /L.min)	2.88	0.13	1.11	0
Immobilized wet biomass (kg/m ³)	383	383	392	392
<i>Elimination capacities</i>				
Overall toluene elimination capacity (g/m ³ .h) ^a	47.0	37.3	35.8	27.8
Toluene degradation rate in liquid (g/m ³ .h) ^b	10.2	0.44	3.9	0
Toluene degradation rate in biofilm (g/m ³ .h) ^c	36.8	36.9	31.9	27.8
<i>Specific activities</i>				
Suspended biomass (mg toluene/g carbon.h) ^d	105	21.4	n.d.	n.d.
Immobilized biomass (mg toluene/g carbon.h) ^e	4.8	4.8	4.0	3.5

^a Overall elimination capacity of the reactor as determined by GC gas analysis.

^b Toluene degradation in the liquid per m³ reactor. Values were estimated from toluene-induced oxygen uptake rates, assuming 69% carbon mineralization (Cox and Deshusses, 1988^c) and a correspondingly lower stoichiometric oxygen demand.

^c By difference from overall elimination capacity and degradation in liquid.

^d Toluene degradation rate in the liquid divided by the amount of total carbon in the liquid.

^e Toluene degradation rate in the biofilm divided by the amount of immobilized C-biomass. Conversion factors for immobilized biomass of 0.046 g dry biomass/g wet biomass and 0.44 g carbon/g dry biomass were used (Cox and Deshusses, 1988^b).

Experiment 2: one-pass operation with fresh water followed by recycling of 32 L water

This experiment consisted of a first phase with fresh water trickling as one-pass operation followed by closed loop recycle of a large volume of water. The total carbon concentration in the original recycle liquid on the day of this experiment was not determined. The toluene-induced OUR was 1.1 mg O₂/L.min, which is 38% of the activity of the original culture liquid in the previous experiment, indicating a lower activity in the liquid at the start.

As compared to recycle operation of the original culture liquid, one-pass operation with fresh water resulted in a decrease of the toluene outlet concentration (Figure 2). This can be explained by continuous absorption of toluene. The toluene degradation rate in the original culture liquid can not be estimated from this one-pass experiment as it would also require quantification of the amount of toluene absorbed and purged via the liquid. After 70 minutes, it was decided to stop the one-pass operation and recycle the water. This is similar to experiment 1, but the volume of recycled water was increased to further reduce the biomass concentration and the biological activity in the liquid phase. Recycling of 32 L water caused a rise of the toluene gas outlet concentration until a constant value was obtained after 1 h (Figure 2). No biological activity was detected in the liquid phase as measured by OUR experiments. Therefore, the overall elimination capacity while recycling 32 L water can entirely be attributed to the activity of immobilized cells. The rate of toluene degradation by the original suspended culture estimated from the difference in overall elimination capacities was 8.0 g/m³.h, which is slightly lower than found in the previous experiment (Table 1). However, in experiment 2 the toluene degradation rate in the original liquid determined from OUR experiments was only 3.9 g/m³.h. We have no good explanation for this low value. It might be due to a lower biomass concentration in the

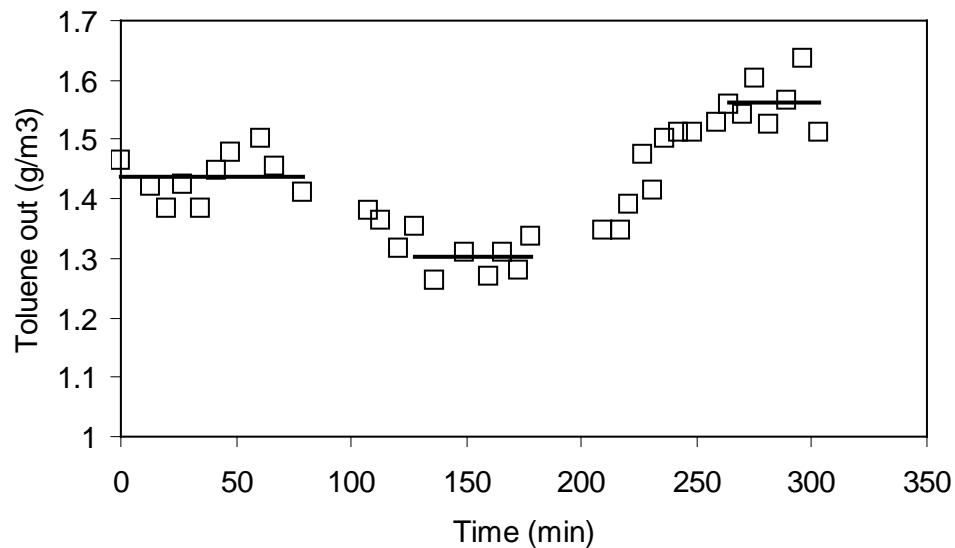


Figure 2. Effect of one-pass operation with water followed by recycle of 32 L of water on the toluene outlet gas concentration. Conditions: recycle of 3 L original culture liquid between $t = 0$ and 79 min, one-pass operation of fresh water between $t = 108$ and 178, recycle of 32 L water between $t = 204$ and 303 min, volumetric load 64 m³/m³.h, toluene inlet concentration 2 g/m³, superficial liquid velocity 7.9 m/h. Solid lines show assumed steady state values.

liquid at the beginning of the experiment.

The suspended biomass contributes to a small but significant part of the total toluene degradation rate. In both experiments, toluene degradation in the liquid phase was 22% of the overall toluene elimination capacity.

DISCUSSION

The results clearly show that a significant part of the pollutant is degraded by the suspended culture, i.e., 22% of the overall elimination capacity. To our knowledge, liquid phase pollutant degradation has only been determined by Hartmans and Tramper (1991), who observed a pollutant degradation rate in the liquid of up to $9.5 \text{ g/m}^3 \cdot \text{h}$ in a dichloromethane-degrading biotrickling filter with a maximal elimination capacity of $200 \text{ g/m}^3 \cdot \text{h}$. In the following sections, we discuss the methodology used and the implications of the findings for modeling and practical biotrickling filter operation.

As outlined in the previous section, two approaches were used to estimate toluene degradation rate in the liquid. The difference between the overall elimination capacities while recycling the original culture liquid and recycling water is the easiest method. Two requirements must be met though. First, toluene degradation in the liquid while recycling water should be zero. OUR experiments indicate that replacement of the original culture liquid with water resulted in a (near) complete loss of suspended biological activity in both experiments (Table 1). The second requirement is that the toluene degradation rate in the biofilm is the same when recycling either the original culture liquid or water. Recycle of water may cause nutrients to be limited in the biofilm, however, the time-span of these experiments was short and nutrient limitation is unlikely to be significant. More important is that after replacing the original culture liquid with water the toluene concentration in the liquid phase will be higher because of an increase of the average toluene gas phase concentration. Hence, the toluene flux into the biofilm will be higher while

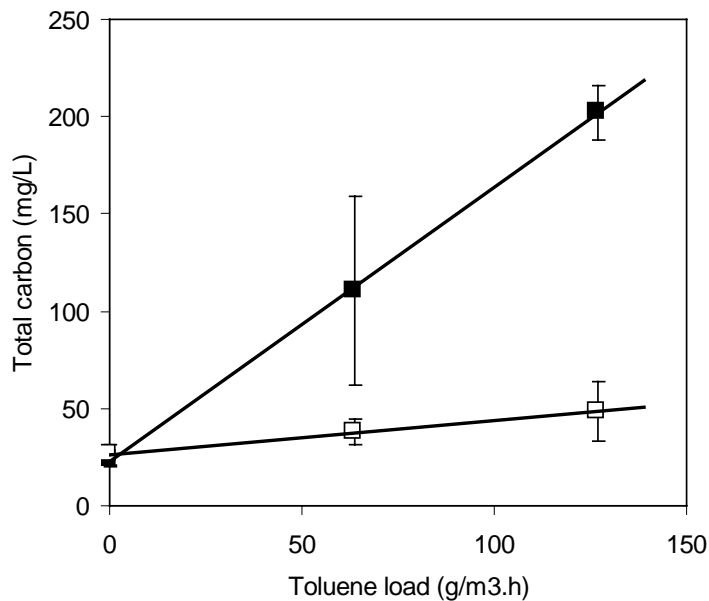


Figure 3. Influence of nutrient supply and toluene load on the biomass concentration in the liquid measured as total carbon. Conditions: volumetric load $64 \text{ m}^3/\text{m}^3 \cdot \text{h}$, toluene inlet concentration 0, 1 or 2 g/m^3 , superficial liquid velocity 7.9 m/h , continuous supply of mineral medium (closed squares) or phosphate buffer (open squares) at an average rate of $11.6 \text{ L/m}^3 \cdot \text{h}$.

recycling water, and the toluene degradation rate in the biofilm probably as well. We neglected this possible effect as the toluene gas phase concentration only slightly increased (Figure 1). It should further be noted that the toluene degradation rate in the liquid phase would have been higher if these mass transfer considerations were indeed accounted for. OUR experiments are an indirect approach to determine the toluene degradation rate in the original culture liquid. The limitations are that oxygen consumption is measured instead of actual toluene degradation and that conditions during OUR measurements may be quite different from the biotrickling filter environment. This may explain why in experiment 2 the toluene degradation rate in the original culture liquid as estimated from OUR experiments was only 49% from as estimated from overall toluene elimination capacities, although a good agreement was observed in experiment 1.

Liquid phase pollutant degradation clearly requires the presence of suspended biomass. Suspended biomass may originate from detachment of immobilized cells. Alternatively, a suspended culture may develop independently from the biofilm when conditions in the liquid are favorable for microbial growth. Over a seven-month period, we investigated steady-state performance and carbon balances of two toluene-degrading biotrickling filters at different loads of nutrients and toluene (Cox and Deshusses, 1998^c). Total carbon in the liquid was used as a measure of the suspended biomass concentration. Figure 3 shows that a high total carbon concentration in the liquid phase requires both the addition of nutrients and a high toluene inlet concentration. These results clearly indicate that in these biotrickling filters high suspended biomass concentrations are caused by intrinsic growth of the suspended culture rather than detachment of immobilized cells. This also implies that the liquid phase pollutant degradation is more likely to be significant in biotrickling filters operated with a high supply rate of nutrients.

Liquid phase pollutant degradation is generally neglected because suspended biomass amounts are considered too low to be significant. Indeed, in experiment 1 the total amount of suspended biomass was only 1% of the amount of immobilized biomass. However, the specific activity of the cells should be considered instead. In our biotrickling filters, the suspended cells were about 20 times more active than the immobilized cells (Table 1). A likely explanation is that conditions for growth and toluene degradation are more optimal in the liquid (high nutrient and toluene concentrations, no oxygen limitation) than in the biofilm. The presence of inactive zones in the biofilm is generally accepted (Cox and Deshusses, 1998^a). In the case of toluene as pollutant, toxicity of the substrate is an additional aspect to consider. Mirpuri *et al.* (1997) and Villaverde *et al.* (1997) have shown that toluene injured bacteria (loss of toluene degradation activity) and that the effects increased both with time and with increasing toluene concentration. As the suspended culture in our biotrickling filters was continuously diluted (mean cell residence time of 12 h), cell injury may have been more pronounced in the immobilized culture than in the suspended culture.

The contribution of biodegradation in the liquid phase to the overall elimination capacity depends on many factors, such as the nutrient availability and toluene gas phase concentration (Figure 3), the recycle liquid dilution rate, the ratio of total liquid volume to reactor volume, and biokinetic parameters of the suspended and immobilized culture. Because most of these factors will be different in each experimental set-up, only general remarks can be made on the effect of liquid phase pollutant degradation on modeling and operation of biotrickling filters. Obviously, neglecting liquid phase degradation will result in an overestimation of pollutant degradation in the biofilm and the actual specific activity of immobilized cells will be lower than estimated from the overall elimination capacity. Depending on the specifics of mass transfer, biodegradation by suspended cells will also decrease the substrate concentration in the liquid layer. This will cause a lower driving force for mass transfer into the biofilm than would be expected when no degradation takes place in the liquid. Finally, intermittent trickling (Wolff, 1992) and operation of a 'dry' biotrickling filter (De Heyder *et al.*, 1994) have been suggested as a means to improve the performance of biotrickling filters degrading hydrophobic compounds. Reduction of the thickness of the water layer improves the mass transfer of these compounds and hence overall performance.

However, depending on the contribution of the liquid to the overall pollutant degradation, this effect could be partially counterbalanced by reduction of liquid phase pollutant degradation.

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Pollutant mass transfer and biodegradation in biotrickling filters revisited

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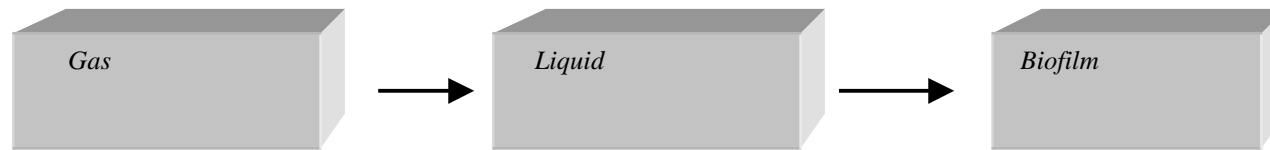
Outline:

- Pollutant mass transfer revisited
- Biodegradation in biotrickling filters revisited
- Conclusions



Myth 1

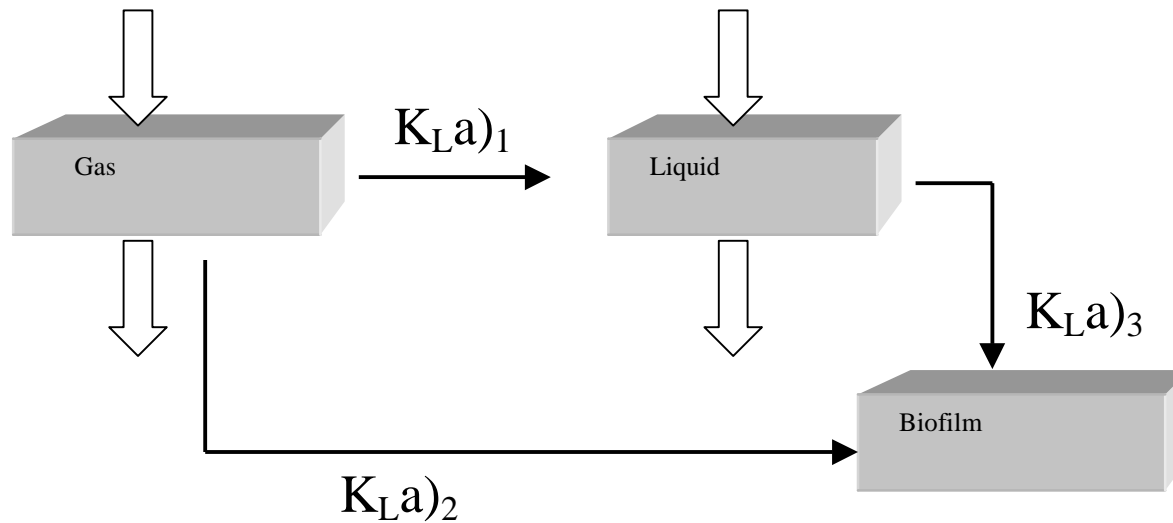
Pollutant mass transfer is:



When the pollutant was added to the liquid instead of the gas, a lower elimination rate was observed.



Conceptual model for pollutant transfer in biotrickling filters

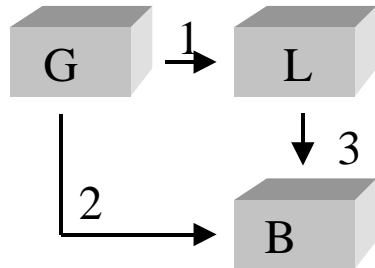


$$\text{Pollutant transfer rate} = K_L a)_i \times \Delta C$$



Determination of the mass transfer coefficients

Absorption/desorption studies with an “inert” VOC: MTBE



- $K_L a)_1$ Steady state: absorption of gaseous MTBE in water (one pass)
- $K_L a)_3$ Transient: biofilm saturated with MTBE, desorption into water (one pass), no air
- $K_L a)_2$ Transient: biofilm saturated with MTBE, desorption into air (one pass) and water (recycle); $K_L a)_1$, $K_L a)_3$ are determined above



Solutions for $K_L a$ determinations

- Approximative, using mean log averages of concentrations
- Mathematical model (transient) with curve fitting

Biotrickling filter

H = 1.3 m

ID = 0.15 m

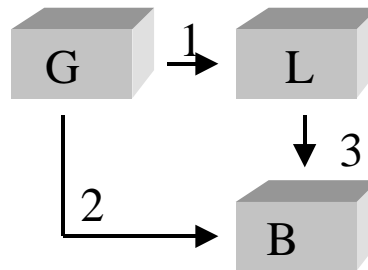
Pall rings, 2.5 cm

Packing area = $220 \text{ m}^2 \text{ m}^{-3}$

EBRT = 113 s

Liquid velocity = 7.9 m h^{-1}

Biomass content = 14 % vol.



Results

$$K_L a)_1 = 1.91 \text{ h}^{-1}$$

$$K_L a)_2 = 4.7-7.1 \text{ h}^{-1}$$

$$K_L a)_3 = 6.8 \text{ h}^{-1}$$



Interim conclusions

- $K_L a$ values are in a common range for bioreactors: $1-7 \text{ h}^{-1}$
- Gas/liquid mass transfer may be a possible rate limiting factor
- Significant pollutant transfer is directly from the gas to the biofilm



Myth 2

Biodegradation only takes place in the biofilm

The recycle liquid in biotrickling filters is often turbid, which indicates a high concentration of suspended microorganisms.

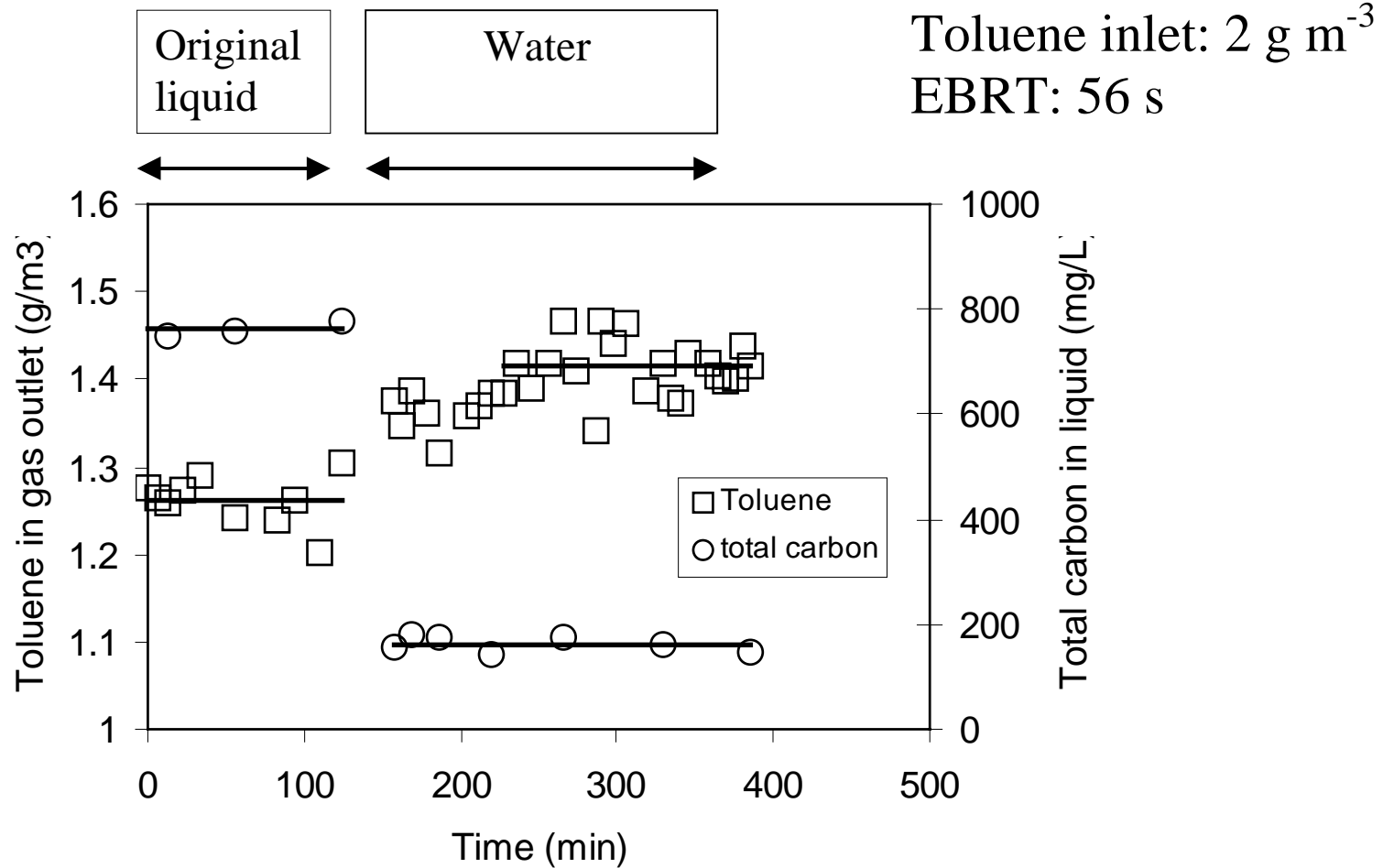


Determination of biodegradation in the liquid

Two approaches:

- 1 Replace the original recycle liquid with the same volume of water and determine the elimination capacity of the biotrickling filter
- 2 Measure the toluene degradation rate in the original recycle liquid (OUR experiments) and extrapolate to the entire filter

Replacement of original recycle liquid with water





Toluene degradation rates

Rate ($\text{g m}^{-3} \text{ h}^{-1}$)	Toluene elimination during circulation of	
	Original recycle liquid	Water
Overall EC	47.0	37.3
In liquid (OUR experiments)	10.2	0.4
In biofilm (overall EC – OUR _{liq})	36.8	36.9
% of EC in liquid	22%	-

Good agreement between the two methods for estimation of the toluene degradation in the original recycle liquid:

$$\text{Method 1: } 47.0 - 37.3 = 9.7 \text{ g m}^{-3} \text{ h}^{-1}$$

$$\text{Method 2: } 10.2 - 0.4 = 9.8 \text{ g m}^{-3} \text{ h}^{-1}$$



Why is biodegradation in the liquid often neglected?

Biotrickling filter (typical data)

Packed bed volume = 23.6 L

Immobilized wet biomass = 9.0 kg

Liquid volume = 3 L

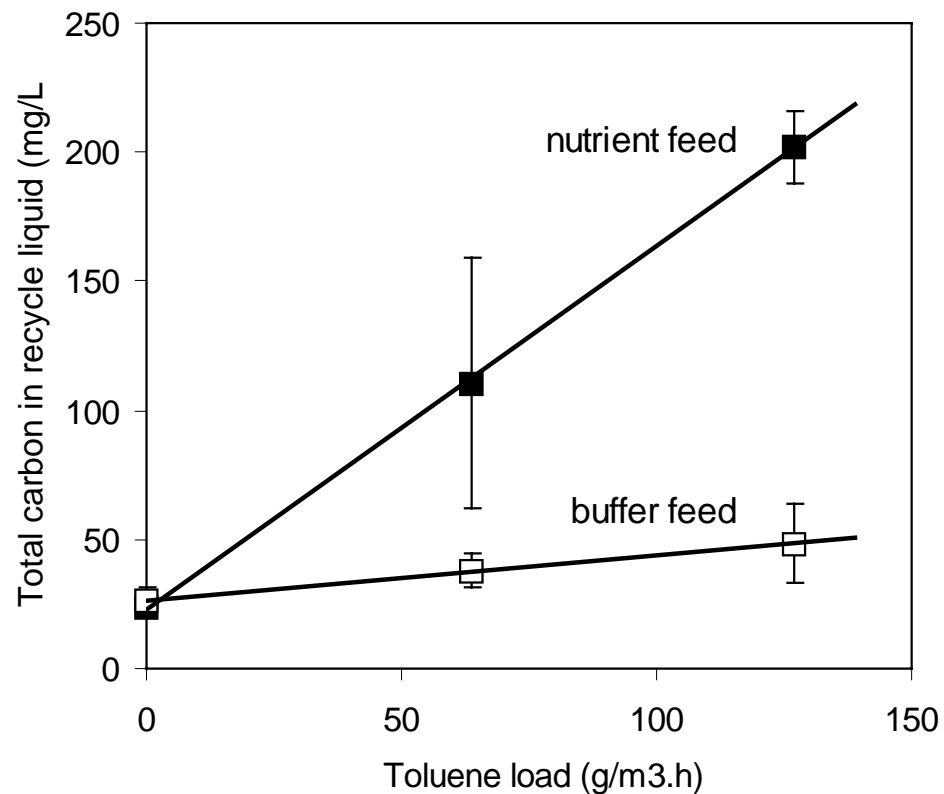
Parameter	Biomass	
	Immobilized	Suspended
Amount of biomass (g C-biomass/m ³ reactor)	7752	97
Specific activity (mg toluene/g C-biomass.h)	4.8	105



When is biodegradation in the liquid significant?

Monitoring total carbon (\approx suspended C-biomass) in the liquid at:

- 0, 1 and 2 g/m³ toluene in the gas
- continuous supply of buffer or nutrient medium





Interim conclusions

- Biodegradation in the liquid can significantly contribute to the overall elimination capacity (up to 20%).
- Biodegradation in the liquid is generally neglected because the amount of suspended biomass is small. However, the activity of the suspended biomass is very high.
- Biodegradation in the liquid is stimulated by high pollutant concentrations and supply of nutrients.



Overall Conclusions

Biodegradation in the recycle liquid can be an important part of the overall pollutant elimination

- Optimizing biotrickling filter performance should consider both the biofilm and the suspended culture
- The health of the suspended culture might be an indication of the reactor well being

Significant pollutant mass transfer occurs directly from the gas phase to the biofilm

- There is room for improvement of mass transfer in biotrickling filters

Mathematical models should consider both biodegradation in the liquid and gas/biofilm mass transfer