

# APPLICATION OF IMMOBILIZED CELLS FOR AIR POLLUTION CONTROL

*cleaning air naturally*

**MARC A. DESHUSSES**

*Department of Chemical and Environmental Engineering, University of California, Riverside, CA 92521, USA. Fax: 909-787-5696, Phone: 909-787-2477, Email: mdeshuss@engr.ucr.edu*

*Keywords: biofilter, biotrickling filter, air pollution control, biofilms, vapour-phase bioreactors, VOC control, odour control*

## **1. Introduction**

The use pollutant-degrading organisms for air pollution control is an important and emerging application of cell immobilization technology. The principle is relatively simple: a contaminated air stream is passed through a packed bed on which pollutant-degrading organisms are immobilized. Contaminants in the air are transferred to the microorganisms, and are degraded to harmless compounds. Air biotreatment is not a new concept, it has been proposed more than 40 years ago [1]. However, it is only in the past two decades that new environmental regulations have forced engineers to consider alternatives to convention air pollution control methods.

The most successful applications of biological techniques for air pollution control have been for the treatment of dilute, high flow waste gas streams containing odours or volatile organic compounds (VOCs) (Figure 1) [2,3,4]. Under optimum conditions, the contaminants are completely degraded to innocuous end-products. The major advantage over conventional treatment technologies is that air biotreatment is accomplished at low temperature, and has lower operating and maintenance costs (see Table 1).

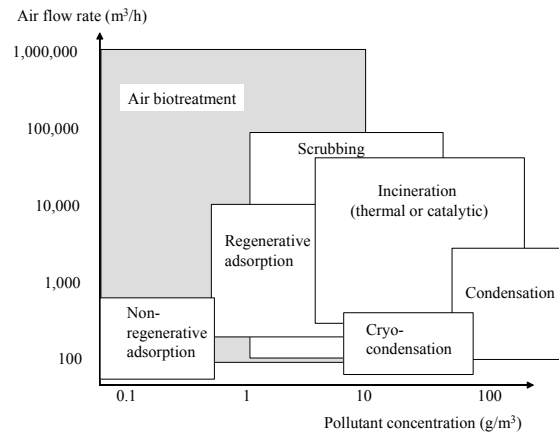


Figure 1. Applicability of various air pollution control technologies.

Table 1. Comparison of biotreatment and conventional air pollution control techniques.

Technology	Advantages	Disadvantages
Biotreatment	<ul style="list-style-type: none"> <li>• Simple and low cost technology</li> <li>• Low to medium capital costs, and low operating costs</li> <li>• Effective removal of odours and low concentrations of contaminants</li> <li>• No production of by-products</li> <li>• Low energy requirement, no fuel needed</li> <li>• Environmentally friendly</li> </ul>	<ul style="list-style-type: none"> <li>• Relatively large footprint requirement</li> <li>• Can not treat non-biodegradable pollutants</li> <li>• Medium replacement every 2-5 years (biofilters only)</li> <li>• Moisture and pH sometimes difficult to control (biofilters only)</li> <li>• Particulate matter may clog the bed</li> <li>• Clogging by growing biomass if too much nutrient is added and high concentrations of VOCs are treated</li> </ul>
Wet Scrubbing	<ul style="list-style-type: none"> <li>• Medium capital costs</li> <li>• Can operate with particulate in gas stream</li> <li>• Relatively small footprint</li> <li>• Ability to handle variable loads</li> <li>• Well proven technology</li> </ul>	<ul style="list-style-type: none"> <li>• Very high operating costs</li> <li>• Reduced performance by scale deposit</li> <li>• Need for complex chemical feed systems</li> <li>• Not effective for most VOCs</li> <li>• Requires toxic/dangerous chemicals</li> </ul>
Carbon Adsorption	<ul style="list-style-type: none"> <li>• Short retention time/small unit</li> <li>• Consistent, reliable operation</li> <li>• Moderate capital costs</li> </ul>	<ul style="list-style-type: none"> <li>• High to extremely operating costs</li> <li>• Carbon life reduced by moist gas</li> <li>• Creates secondary waste streams (spent carbon)</li> <li>• Medium pressure drop</li> </ul>
Incineration	<ul style="list-style-type: none"> <li>• Effective removal of compounds irrespective of nature and concentration</li> <li>• Suitable for very high loads</li> <li>• Performance is uniform and reliable</li> <li>• Small footprint</li> </ul>	<ul style="list-style-type: none"> <li>• High operating and capital costs</li> <li>• High flow/ low concentrations not cost-effective</li> <li>• Usually requires additional fuel</li> <li>• Creates a secondary waste (NO<sub>x</sub>)</li> <li>• Scrutinized by the public</li> </ul>

Biodegradation of the contaminants in bioreactors for air pollution control is usually mediated by mixed cultures or consortia. The primary pollutant-degraders are similar to the organisms found in wastewater treatment processes. They are thriving in a complex and stressful environment that include higher organisms, such as protozoa, rotifers, even larvae, worms, insects and other predators. The nature of the primary degraders and the fate of the pollutant depend on the main pollutant(s) being treated. Further discussion of the microbial ecology of bioreactors for air pollution control can be found in Section 2. Table 2 lists the most important applications and the class of organisms responsible for pollutant removal.

*Table 2. Type of air contaminants that can be treated in bioreactors and class of primary degrading organisms.*

Pollutant	Primary degraders	End product
VOCs, hydrocarbons	Heterotrophic, use hydrocarbon as carbon and energy source	CO <sub>2</sub> , biomass*
H <sub>2</sub> S	Autotrophic, use CO <sub>2</sub> as carbon source	SO <sub>4</sub> <sup>-</sup> , biomass
Reduced sulphur compounds (e.g., dimethyl sulphide)	Heterotrophic and autotrophic mixed cultures	CO <sub>2</sub> , SO <sub>4</sub> <sup>-</sup> , biomass
Ammonia	Nitrifying organisms, possibly associated with denitrifiers	NO <sub>3</sub> <sup>-</sup> (and NO <sub>2</sub> <sup>-</sup> ), possibly N <sub>2</sub> biomass
NO <sub>x</sub> (via denitrification)	Heterotrophic or autotrophic (denitrifying)	N <sub>2</sub> , oxidized electron donor (organic or inorganic), biomass
NO <sub>x</sub> (via nitrification)	Heterotrophic or autotrophic (nitrite oxidizers)	NO <sub>3</sub> <sup>-</sup> , biomass

\*Biodegradation of chlorinated VOC will result in chloride, brominated compounds will give bromide, sulphur containing VOC will result in sulphate, nitrogen containing compounds will result in nitrate.

The two most promising bioreactors for air pollution control are biofilters and biotrickling filters. Their principles are explained below and schematically shown in Figures 2-3. A picture of an actual biofilter system is shown in Section 3. Other bioreactor setups (e.g., airlift reactors, bioscrubbers, membrane bioreactors, etc.) have been used in air pollution control, but because they are less relevant for industrial application, this Chapter focuses on biofilters and biotrickling filters.

In biofilters, a humid stream of contaminated air is passed through a porous packed bed, usually made of a mixture of compost and wood chips or any other bulking agent [2,5,6]. On the packing, pollutant-degrading organisms form a biofilm and degrade the absorbed contaminants. Biofilters are very dry systems with no or little water trickling, hence any metabolite formed during biodegradation will stay in the damp material of the packing. Over time, this may cause inhibition of the process culture. Flushing of the bed is usually effective in removing accumulated metabolites, however it also leaches nutrients and often results in the compaction of the packed bed structure, hence, flushing should be exercised with caution. Biofilters are simple and cost effective. They require only low maintenance and are particularly effective for the treatment of odour and volatile compounds that are easy to biodegrade, and for compounds that do not generate acidic by-products. Biofilters are widely used in industrial applications for either VOC or odour control.

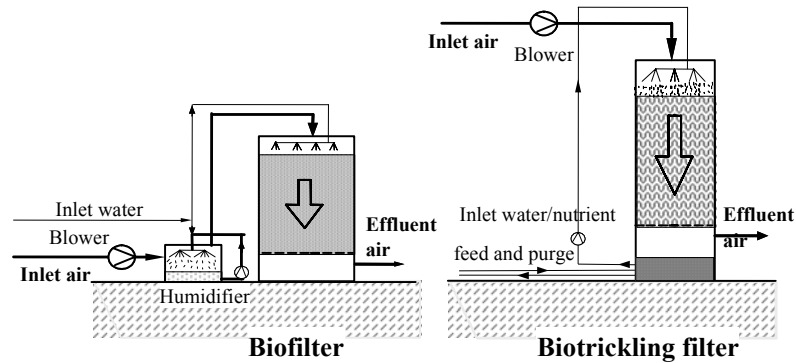


Figure 2. Schematic of biofilter and biotrickling filter setups. In-vessel systems are shown, but open bed design is common for biofilters. The air can be upflow or downflow. The biofilter shown includes sprinklers for additional moisture supply.

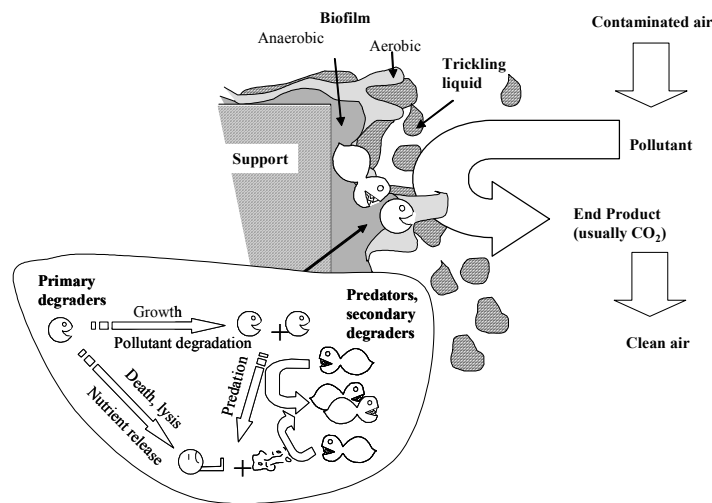


Figure 3. Simplified treatment mechanism in a biotrickling filter. Biofiltration mechanism is similar except that there is no free liquid trickling. Note that in both case, direct gas-biofilm contact exists.

Biotrickling filters work in a similar manner to biofilters, except that an aqueous phase is trickled over the packed bed, and that the packing is usually made of a synthetic or inert material, such as plastic rings, open pore foam, lava rock, etc. [7,8] The trickling solution contains essential inorganic nutrients such as nitrogen, phosphorous, potassium, etc. and is either slowly trickled and wasted (one refers often to a trickling bed biofilter in such case), or trickled at a higher rate and partly recycled. Biotrickling filters are more complex to build than biofilters but are usually more effective. They are specially well suited for the treatment of compounds that generate

acidic by-products, such as H<sub>2</sub>S or methylene chloride, because the free aqueous phase allows for a tight control of the conditions such as pH or ionic strength. Another advantage of biotrickling filters is that they can be built taller than biofilters (2-3 m bed height, vs. 1-1.5 m in biofilters) because the packing used for biotrickling filters is usually not subject to compaction. This reduces the required footprint. Biotrickling filters are more recent than biofilters, and have not yet been deployed for industrial applications to the same extent as biofilters.

## 2. Microbiology of Gas Phase Bioreactors

### 2.1 MICROFLORA

As shown in a simplified manner in Figure 3, pollutant elimination in biofilters or biotrickling filters is the result of many, interdependent processes that simultaneously take place inside the biofilm. The level of understanding of the detailed mechanisms of biofiltration and biotrickling filtration is still relatively limited, most probably because of the marked differences that exist between organisms in traditional bioprocesses and in biofilters or biotrickling filters (Table 3). However, significant progress has been made in the past decade, and a more detailed discussion of the process microbiology and biofilm architecture is warranted.

*Table 3. Differences between traditional biotechnology and biofiltration processes*

Traditional biotechnology process	Biofilter or biotrickling filter
Pure cultures, closed system	Mixed cultures, presence of higher organisms, open system
Plenty of nutrients	Often nutrient limited
Conditions optimized for cell growth, growing cells	Suboptimum growth conditions. Many cells in a resting state, predation, death and lysis
Homogeneous well mixed systems	Mass transfer limitations, presence of significant heterogeneities
Consistent well defined substrate feed	Continuously changing pollutant-substrate nature and concentration

Inside the biofilm, biodegradation of the pollutant is mediated by mixed cultures of bacteria and fungi, thriving in a complex ecosystem. Initial inoculation with competent pollutant-degrading organisms is often unnecessary for biofilters, as most biofilters use compost as packing, which contains a large and diverse microflora suitable for the degradation of common air pollutants [2]. Inoculation is always required for biotrickling filters which packing does not naturally contain microorganisms. The inoculum is usually from enrichment cultures, or simply from activated sludge or another convenient source of pollutant-degrading organisms. For research purposes, some investigators have worked with monocultures and have sterilized air and aqueous feeds [9,10]. However it is practically impossible to maintain such a system free of bacterial contamination for extended periods of time. Almost all biofilters and

biotrickling filters are open systems, in which the process culture naturally evolves over time depending on the operating conditions.

Webster et al [11] observed changes in phospholipids fatty acids (PLFA) biomarkers for as long as 6 months after the startup of their biofilters. A common observation by several investigators is that biodiversity decreases over time, as a result of natural enrichment of those organisms for which the conditions are favourable, and the slow disappearance of all other organisms by death, lysis and cryptic growth [12]. Sakano and Kerkhof [13] found that over a period of about 100 days, only 38% of the original species remained in their ammonia treating biofilter.

Usually, effective pollutant removal is observed within a few days after initiating treatment. However, a more careful selection of the inoculum source is required when the pollutant is either very toxic, difficult to degrade, or when the conditions are very stringent (e.g., low pH, or high temperature). Oh and Bartha [14] reported that a 4-week acclimation phase was required for effective treatment of nitrobenzene vapours. During the acclimation, the inlet concentration of nitrobenzene was incrementally increased to avoid toxic shocks. Another example illustrating the effect of the source inoculum is shown in Figure 4 for the treatment of methyl tert-butyl ether (MTBE), a compound which is only degraded by specialized organisms. Effective removal of MTBE was only observed after seven months of operation, when the biotrickling filter was inoculated with contaminated soil and groundwater. This is obviously an unacceptable delay for practical applications. The startup phase could be significantly shortened by inoculating the biotrickling filter with an adapted consortium. Finally, removal was observed only days after inoculating a pure culture capable of degrading MTBE.

Many of the organisms that have been identified in biofilters and biotrickling filters are similar to those found in wastewater treatment. They include organisms such as *Pseudomonads*, *Rhodococcus*, *Acinetobacter*, *Corynebacteria*, *Gordona*, *Xanthomonas* species, etc. [2,16,17,18]. Interestingly, the primary pollutant-degraders are not necessarily the dominant organism in biofiltration systems. Moller et al. [19] described the distribution of *Pseudomonas putida* in the biofilm of a toluene degrading biotrickling filter using scanning confocal laser microscopy, 16S rRNA probes, and various staining techniques. Interestingly, *P. putida* constituted only 4% of the total biofilm population, but was responsible for about 65% of the degradation of the toluene vapours. Further, the comparison of rRNA content of *P. putida* in the biofilm and growing under optimum conditions in suspension indicated that toluene degradation activity by *P. putida* in the biofilm was substantially lower than in suspension. This can most probably be attributed to various stresses and limitations imposed on the immobilized culture as outlined earlier in Table 3.

Fungi have also frequently been observed, especially in biofilters, either as primary pollutant-degraders or as secondary degraders living off metabolites or degrading the lignocellulosic material of compost biofilters. Interestingly, white-rot fungi which are very popular in xenobiotic biodegradation studies have not found widespread application in gas phase bioreactors. A possible reason is that they degrade pollutants by means of extracellular peroxidases, a process which results in the formation of intermediate by-products [20] for which the white-rot fungus may not compete effectively. Still, other fungi have been successfully deployed in biofilters. Their resistance to low pH and low water activities makes them particularly well suited for

gas-phase bioreactors. It has also been proposed that fungi may “harvest” pollutant via their airborne hyphae [21,22], therefore by-passing the mass transfer resistance posed by the liquid film. This may be particularly beneficial for the treatment of hydrophobic compounds. Indeed, effective removal of compounds such as styrene, toluene, alkylbenzenes, and nitric oxide has been reported in fungal based bioreactors [22,23,24,25,26].

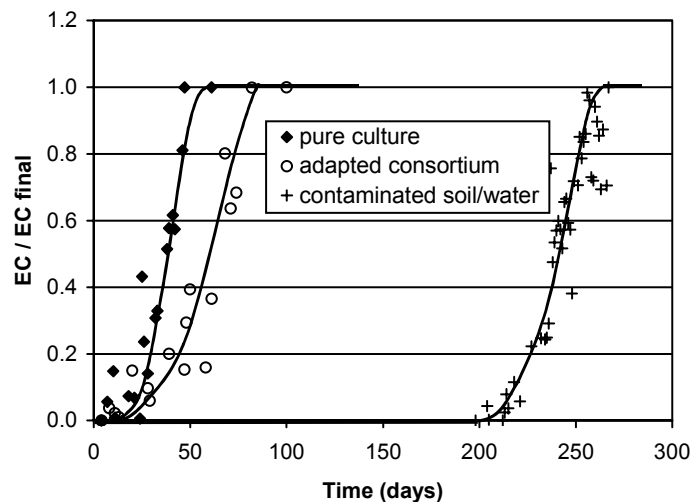


Figure 4. Start-up of MTBE-degrading biotrickling filters inoculated with different sources of microorganisms. EC = pollutant elimination capacity. (Source: unpublished data from Cox and Deshusses for the pure culture and adapted consortium, Fortin and Deshusses [15] for the contaminated soil/water inoculum data).

## 2.2 BIOFILM ARCHITECTURE

An important difference between the organisms present in biofilters and biotrickling filters and those discussed in many chapters of this book is that organisms in biofilters and biotrickling filters are present in biofilms rather than entrapped or attached. The biofilms consist mostly of cells and exopolymers and water [27]. The biofilms usually range from 20 micrometer to several millimetres thick, although it has been shown that in most cases, only the section that is close to the gas or liquid interface is biologically active, while the rest of the biofilm is subject to mass transfer limitations [19,27, 28].

Early representations of biofilms considered the biofilm to be a flat surface. It is now well known that biofilms are far from being planar but that they are very heterogeneous, with large channels extending from the gas or liquid interface of the biofilm deep into the biofilm, sometimes up to the substratum. Such channels and heterogeneities are thought to contribute to a possible enhancement of pollutant and oxygen mass transfer. de Beer et al. [28] evaluated that for submerged biofilms, the

supply of oxygen through such voids and channels was roughly 50% of the total oxygen transfer.

Detailed microscopical observation [29] of relatively thick biofilms (2-5 mm) taken from an active H<sub>2</sub>S degrading biotrickling filters revealed that they include three regions with approximately constant relative thicknesses. The external film (5% of total thickness) had a high bacterial and hyphal tip density and included numerous channels free of cells, as discussed above. While it was not measured, it is reasonable to speculate that this region was responsible for most of the biodegradative activity. The intermediate region (about 20% of total thickness) was characterized by a lower density of bacteria, an increased density of hyphae and an absence of channel-like structures. Also a number of nematodes were detected. Finally, the basal region (75% of total thickness) composed of tightly compacted hyphae against the substratum exhibited little staining with haemato-eosin. This indicates the absence of cytoplasmic material, i.e., the organisms were dead or completely inactive. The heavy presence of presumably heterotrophic fungi throughout the biofilm in a reactor used for hydrogen sulphide and carbon disulfide treatment suggests that the fungi were essentially living off the by-products or metabolites secreted by the primary degraders. A similar observation was made by Woertz et al. [26] who described the presence of heterotrophic denitrifying fungi in toluene degrading biofilters. It was shown that the fungi used metabolites of toluene biodegradation while denitrifying nitric oxide. In fact, heterotrophic denitrification of nitrate (while degrading organic contaminants) could be more widespread in gas phase biotrickling filtration than originally thought. The process would enable biodegradation of organic pollutants in anaerobic pockets resulting from oxygen diffusion limitation, and would be favoured in reactors where plenty of nitrate is available.

### 2.3 SECONDARY DEGRADERS AND PREDATORS

The current knowledge of the nature and role of secondary degraders and predators in biofilters and biotrickling filters is relatively limited. It is well known that protozoa and other higher organisms are always present in gas phase bioreactors. Their cryptic growth [12] and predation of the primary degraders are expected to play a major role in recycling essential nutrients such as nitrogen, phosphorous and potassium. In fact, applying conventional rules of thumb for carbon to nutrient ratio to operating biofilters and biotrickling filters reveals that those bioreactors are usually moderately to severely limited by the supply nutrients. While nutrient limitation is sometimes done on purpose for limiting the growth of biomass and avoid plugging of the bed [30,31,32], effective VOC biodegradation under severe nutrient limitation can only be explained by the recycling action of the predators and by the fact that the process culture degrades VOCs to satisfy its maintenance requirements.

Higher organisms also impact the rate of biomass accumulation by either physically detaching biofilms through their grazing and tunnelling activity, or by converting part of the biofilm to carbon dioxide as a result of their own metabolism. Cox and Deshusses [33] showed that biomass accumulation was slower in a toluene-degrading biotrickling filter in which selected protozoa were added, compared to a protozoa-free control. The presence of protozoa correlated with a higher rate of toluene degradation, a

higher CO<sub>2</sub> yield and lower pressure drop. Interestingly, the startup of the biotrickling filter with protozoa was faster than the control, possibly because of growth factors or recycled nutrients secreted by the protozoa. Cox and Deshusses also observed that the biofilms grown in protozoa-free environment were denser, and would not detach as easily from the substratum. Still, the predation by the protozoa was not sufficient to completely balance the growth of the process culture and the biotrickling filter eventually clogged. Other studies on predation include one by Woertz et al. [34] who observed that biofilters inoculated with the toluene-degrading fungus *Cladophialophora sp.* had a higher performance and a lower pressure drop when *Tyrophagus* mites were added to the system and allowed to graze on the fungus.

In another study, Won et al. [35] described the effect of fly larvae on biotrickling filters for air pollution control. Flies have frequently been associated with biofilters and are usually considered as a nuisance. Here, a small fly identified as *Telmatoscopus albipunctatus*, a psychodid fly species thriving on decaying material, drain, and wastewater treatment activities invaded a lab-scale biotrickling filter and larvae rapidly spread throughout the reactor. This resulted in effective removal of biomass from the packing. The wet biomass content of the reactor was reduced from 455 to 28 kg m<sup>3</sup> reactor in 16 days with 80% of the biomass reduction occurring in 2-4 days. Analysis of the recycle liquid indicated that the major mechanism of biomass removal was detachment of biofilm, while an estimated 2-10% of the removal may have been by consumption by the larvae. It was speculated that larval activity loosened the biofilm structure, thus enhancing biofilm detachment by shear-stress from the trickling liquid.

Overall, these studies conducted with higher organisms illustrate the relatively unexplored potential for controlling biomass and possibly enhance the performance of biofilters and biotrickling filters. However, the complexity of these multispecies systems governed by interconnected relationships still represents a major research challenge.

## 2.4 BIODEGRADATION AND GROWTH KINETICS

The kinetics of pollutant elimination in the biofilm are influenced by numerous factors including the type and design of the bioreactor, the operating conditions, the pollutant(s) being treated, etc. Also, conditions along the height of the reactor are expected to be drastically different. At the microscale, biodegradation rates are influenced by the environmental conditions such as pH, substrate and nutrient concentrations, etc. which are themselves affected by mass transfer limitations. Clearly, the situation is complex. In a first approximation, neglecting heterogeneities and mass transfer effects, Deshusses and Cox [36] proposed that, one could write that the pollutant elimination capacity (EC) expressed in g of pollutant degraded per unit bed volume per hour, depended on the intrinsic growth rate of the active fraction of the primary degraders ( $X_1$ ) and its maintenance requirement, as in Equation 1.

$$EC = \left( \frac{\mu}{Y_{X/S}} + m \right) \times X_{1(\text{active fraction})} \quad (1)$$

where  $\mu$  is the specific growth rate of the primary degraders,  $Y_{X/S}$  is the biomass yield,  $m$  the maintenance energy requirement, and  $X_{1(\text{active fraction})}$  is the biomass content of active primary degraders per volume of reactor. Note that the maintenance energy is included in the kinetic relationship, as it is suspected to be an important mechanism of pollutant removal compared to growth associated pollutant degradation. This is because pollutant-substrate concentrations can be very low (see Table 4, below), and because the primary degraders are subject to significant stresses that may result in high maintenance requirements. The shift from a growing to an essentially non-growing metabolism over time was extensively discussed by Cherry and Thomsson [37] and experimentally proven later by Fuerer and Deshusses [38].

The specific growth rate of the active fraction of the primary degraders can be expressed using a modified Monod equation that takes into account all possible limitations.

$$\mu = \frac{\mu_{\max} \times S}{K_S + S} \times \frac{N}{K_{S_N} + N} \times \frac{O}{K_{S_O} + O} \times \frac{I}{1 + \frac{I}{K_I}} \quad (2)$$

where  $S$  is the pollutant-substrate concentration,  $N$  is a lumped parameter representing the concentration of limiting nutrients,  $O$  is the dissolved oxygen concentration,  $I$  the concentration of any inhibitor, and  $K_S$ ,  $K_{S_N}$ ,  $K_{S_O}$ , and  $K_I$  are the respective half-saturation and inhibition constants.

Interestingly, biofilters and biotrickling filters have been applied for pollutants with an extremely broad spectrum of physico-chemical properties. Of prime importance is Henry's law coefficient of the pollutant undergoing treatment. Table 4 illustrates that the actual concentration seen by the process culture may vary by several orders of magnitude, depending on the application. This will of course affect the biodegradation kinetics (see Equation 2). Pollutants with low Henry coefficients will favourably partition into the biofilm and concentrations will often be above the substrate half-saturation constant  $K_S$ . These pollutants may cause oxygen depletion in the biofilm if present at high concentration in the air undergoing treatment. Pollutants with higher Henry coefficients will have concentrations in the biofilm that are comparable or below the  $K_S$  value, and pollutant removal will be subject to kinetic limitations. Certainly, the lowest biofilm concentrations indicated in Table 4 raise interesting fundamental questions on the induction of key enzymes in the process culture. This remains a relatively unexplored area.

Table 4. Henry's law coefficient (H) and typical gaseous inlet concentration of selected pollutant and corresponding liquid equilibrium concentration. Note that the actual concentration in the biofilm may be orders of magnitude lower because of mass transfer limitations and/or axial position in the reactor.

Pollutant	Typical application	Typical inlet gas concentration (g m <sup>-3</sup> )	H (-)	Corresponding pollutant equilibrium liquid concentration (mg L <sup>-1</sup> )
Methanol	Wood industry	0.2-1.0	0.0002	1000-5000
Methyl ethyl ketone	Paint spray booth	0.1-0.5	0.0024	40-200
Toluene	Various processing	0.1-0.5	0.275	0.4-2
Toluene	Air exhaust, wastewater treatment plants	0.002	0.275	0.007
H <sub>2</sub> S	Air exhaust, wastewater treatment plants	0.003-0.15	0.385	0.008-0.1
NO	Combustion gases	0.5	21	0.024
Hexane	Various processing	0.1-0.5	53	0.002-0.01

As far as secondary degraders as concerned, an equation similar to Equation 2 can be written for all the species (or group of species) present in the system. Each will have one or several specific substrates (such as a metabolite, exopolymer, primary degrading organism, etc.), specific kinetic constants, and thus a different specific growth rate. The overall rate of biomass accumulation in the biofilter or biotrickling filter system is the sum for all the different species (designated by the index *i* in Equation 3) of the growth rate minus death and lysis (*d<sub>i</sub>* term), the predation by other species and the wash-out via leaching. This is expressed in Equation 3.

$$\text{Rate of biomass accumulation} = \sum_i ((\mu_i - d_i) \times X_i - \text{Pr edation}_i - \text{Wash out}_i) \quad (3)$$

As mentioned, Equations 1-3 are highly simplified since they do not take local heterogeneities into account. Still, they define a number of parameters that are impossible to determine. A possible solution is to conceptually split the process culture into large classes of organisms, such as primary degraders, secondary degraders, predators, etc. and use lumped kinetic parameters. This is an area of current research. Even so, Equations 1-3 reflect the fact that the pollutant elimination, secondary processes and the observed biomass growth are interconnected in a complex manner. Equation 3 can be further used to support the development of biomass control strategies by minimizing or zeroing out the biomass accumulation term. Examination of Equation 3 shows that meaningful biomass control strategies should consider means to reduce the specific growth rate [30,31,32,39], accelerating death and lysis, stimulating predation [33,34], and mechanically washing out the biomass [40,41,42]. The challenge is to conduct any of the above biomass control measures in a cost effective manner, without negatively affecting the rate of pollutant elimination by the primary degrading organisms (Equation 1).

### 3. Biofilter and Biotrickling Filter Applications

#### 3.1 DEFINITIONS AND PERFORMANCE REPORTING

The performance of bioreactors for air pollution control is generally expressed as pollutant removal efficiency, or pollutant elimination capacity and reported as a function of the pollutant inlet concentration, pollutant loading, or gas empty bed retention time (EBRT). These terms are defined below (Equations 4-7).

$$\text{Removal} = \text{RE} = \frac{C_{\text{in}} - C_{\text{out}}}{C_{\text{in}}} \times 100 \quad (\%) \quad (4)$$

$$\text{Pollutant Elimination Capacity} = \text{EC} = \frac{(C_{\text{in}} - C_{\text{out}})}{V} \times Q \quad (\text{g m}^{-3} \text{ h}^{-1}) \quad (5)$$

$$\text{Empty Bed Retention Time} = \text{EBRT} = \frac{V}{Q} \quad (\text{s or min}) \quad (6)$$

$$\text{Pollutant loading} = L = \frac{C_{\text{in}}}{V} \times Q \quad (\text{g m}^{-3} \text{ h}^{-1}) \quad (7)$$

where  $C_{\text{in}}$  and  $C_{\text{out}}$  are the inlet and outlet pollutant concentrations (usually in  $\text{g m}^{-3}$  or in dilution to threshold for odour removal), respectively,  $V$  is the volume of the packed bed ( $\text{m}^3$ ) and  $Q$  is the air flow rate ( $\text{m}^3 \text{ h}^{-1}$ ). It should be stressed that the elimination capacity and the loading are calculated using the volume of the packed bed and not the total volume of the reactor. Such normalization enables comparison of systems of different sizes operated under different conditions. Depending on the reactor design, the volume of the packed bed will be about 40-90% of the total reactor volume [2]. Also, EBRT is calculated on the basis of the total volume of packed bed (Equation 6) and not the void space in the packing. This is because the porosity of the bed is usually unknown and it may change over time as a result of bed compaction (in biofilters) or biomass growth. Typical porosities range from 40 to 60% of the bed volume, hence the actual gas residence time will be much lower than the EBRT. Typical EBRTs in biofilters and biotrickling filters range from 15-60 seconds. Obviously, compounds that are more difficult to biodegrade require longer contact times.

In the case of a typical VOC, reporting pollutant elimination capacity vs. its loading usually results in a curve similar to the one shown in Figure 5. One underlying assumption is that the performance depends only on the pollutant load, hence, that low concentrations-high flowrates conditions lead to similar elimination capacities than high concentrations-low flowrates. This assumption is often valid in laboratory studies because the pollutant concentrations treated in biofilters and biotrickling filters are

often high enough for the micro-kinetics to be of zero order. As schematically shown in Figure 5, the assumption is no longer true at low pollutant concentrations, in particular for pollutants with high Henry's law coefficients, because first order kinetics will prevail in the biofilm resulting in a reduction of the maximum elimination capacity.

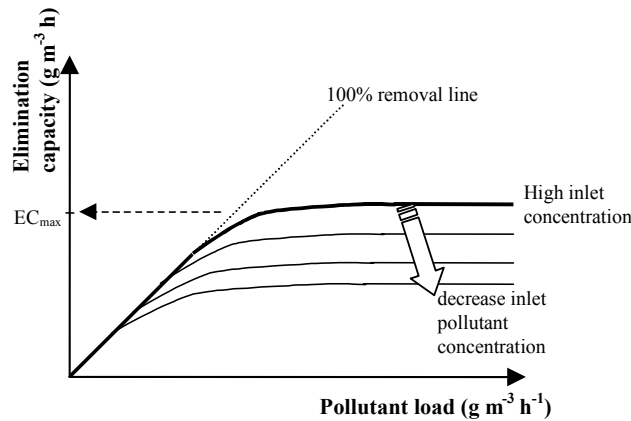


Figure 5. Schematic of a typical elimination capacity vs. load curve for a biofilter or a biotrickling filter.

Examination of Figure 5 reveals that there are essentially three operating regimes:

1. Low loading, also called first order regime. The elimination capacity and the loading are identical and the pollutant is completely removed. The bioreactor is operated well below its maximum elimination capacity. The performance increases proportionally with the loading.
2. Intermediate range. Breakthrough of the pollutant occurs. With higher inlet concentrations or higher air flow rates, the elimination capacity increases, but to a lesser extent than the loading.
3. High loading, also called zero order regime. The biofilter or biotrickling filter is operated at its maximum elimination capacity. Increases in loading do not result in further increases in elimination capacity, the removal efficiency decreases. As indicated on the figure, if the concentration is very low, the maximum elimination capacity will be reduced.

For the evaluation of biofilter or biotrickling filter performance, one should consider both the maximum elimination capacity and the removal efficiency. For practical reasons, academic research is mainly concerned with the maximum elimination capacity or with high performance, which occur at relatively high pollutant concentration and often less than ~90% removal efficiency. On the other hand, reactor design for industrial application often needs to meet a certain discharge requirement, or achieve a high removal percentage. Also, lab-scale studies are usually conducted with single pollutant under steady conditions. On the other hand, industrial exhausts usually consist of mixtures of pollutant, with high variability in concentrations over time. Thus there are significant challenges in extrapolating research data for reactor design.

### 3.2 EXAMPLES OF APPLICATIONS

In the present section, selected examples of application are briefly listed. It should be stressed that research in the area of gas-phase bioreactors has increased exponentially over the past decades, and that numerous papers exist on lab-scale applications. Comparison of the performance between different studies is made difficult by the differences in the reactor types, packing, operating conditions, etc. As a result, the reported performance for a given pollutant often greatly vary depending on the studies, which makes design of large scale biofilters and biotrickling filters a difficult task in absence of pilot testing. Table 5 also lists selected field applications. It should be mentioned that there is a relatively large number of full-scale biofilters operating successfully in the field. Because of environmental regulations, a majority of applications in the USA has been for odour control, whereas in Europe, there is a greater proportion of applications for VOCs control. A picture of an actual biofilter is shown in Figure 6.



*Figure 6. Picture of an actual biofilter. The in-vessel system treats  $H_2S$  and other odours at a wastewater treatment facility. The EBRT is 20 s. (Courtesy of Biorem Technologies, Inc.)*

Table 5. Examples of biofilter and biotrickling filter applications (laboratory and full-scale). Bf = biofilter; Btf = biotrickling filter.

Pollutant	Type of reactor	Packing	EBRT (s)	EC <sub>max</sub> (g m <sup>-3</sup> h <sup>-1</sup> )	Remarks	Ref.
Ethanol	Lab-scale bf	Granular activated carbon	186	200	Acetic acid was formed probably because of oxygen limitation	[43]
Styrene	Lab-scale bf	Perlite	25-90	60	Fungal based system	[22]
Hexane	Lab-scale bf	Compost + perlite	60	21	System performance correlated with nutrient supply	[44]
Diethyl ether	Lab-scale btf	Celite (inert silicate pellets)	25	60	Low trickling rate, one pass	[45]
Methyl ethyl ketone	Lab-scale btf	Wood bars	88	30 (wood)	Performance correlated with packing area	[46]
		Polyprop. Spheres	88	40 (PP)		
Toluene	Lab-scale btf	2.5 cm Polyprop. Pall rings	56	71-83	CO <sub>2</sub> balances are shown	[33]
Toluene	Lab-scale btf	Steel Pall rings	32-160	35	Partial removal at high EC	[47]
Toluene	Lab-scale bf	Celite (inert silicate pellets)	60	270	Fungal based system	[26]
Methanol	Lab-scale btf	Polypropylene	60	85-90	Thermophilic operation at 70 C.	[48]
Alpha-pinene	Lab-scale btf	Polypropylene	60	30	At 55 C, no removal at 70 C.	[48]
Gasoline vapours	Full-scale bf	Compost + perlite	96-180	Not reached	60-90% removal of 0.4-0.8 g m <sup>-3</sup>	[49]
CS <sub>2</sub>	Full-scale btf	Structured packing, plastic	33-40	220	Simultaneous 99% removal of trace H <sub>2</sub> S	[50]
H <sub>2</sub> S	Full-scale btf	Open pore polyurethane foam	1.6-2.3	105	Secondary effluent used as nutrient source. pH of btf = 1.8.	[51]
Odours	Full-scale bf	Soil	210	NA	99% odour removal	[2]
Odours	Full-scale btf	Open pore polyurethane foam	11	NA	>90% odour removal	[2]

#### **4. Current Research, Emerging Topics**

Bioreactors for air pollution control are a major progress in environmental protection. Currently, factors slowing down the deployment of air biotreatment techniques include both regulatory issues, technical issues, and the inherent resistance of the market to new technologies. The latter aspect is also significantly influenced by the preconceived idea that biological systems are unreliable and can not consistently sustain effective treatment over time. With the increasing number of success stories, this element should decrease in the near future. As far as regulations are concerned, there is still a bias towards less environmental friendly techniques such as incineration or carbon adsorption. While, these techniques may be able to achieve higher pollutant removal percentages, they usually do not compare well with biological techniques when the global environmental impact is considered. For example, incineration of low VOC concentrations requires burning additional fossil fuel and generate nitrogen oxides and CO<sub>2</sub>. The global impact of these is usually not considered, although it can be significant. Further studies in the area of lifecycle assessment are required to adjust the metrics used in air pollution control regulations, and to inform regulators about the benefits of biotechniques for air pollution control.

The present know-how in air biotreatment is sufficient to deal with a large number of cases, in particular in VOC and odour control. Still, a variety of challenges remain before the complex phenomena that occur during biotreatment in vapour-phase bioreactors are fully understood. Whether such a detailed understanding is absolutely required is debatable. In fact, the situation is not very different from the current situation with wastewater treatment, where biotreatment is widely applied, but fundamental aspects of the treatment are not fully understood. Fundamental research challenges in air biotreatment include understanding the complex microbiology of the process, and mass transfer aspects specific to gas-phase bioreactors. Current research efforts are directed towards developing new vapour-phase bioreactors for air pollution control, new packing supports for microorganisms, optimization of current applications and finding new ones. Selected exciting recent developments or areas of growing research are briefly summarized below.

As discussed in Section 2, the use of fungi in vapour-phase bioreactors appears to be extremely promising. It has already been shown that fungal based system can often degrade VOCs at faster rate than conventional bacterial based systems. Fungal based systems seem to be less sensitive to pH and relative humidity changes. Still several issues such as the control of the growth of fungi (especially if dimorphic fungi are used), or the possible emissions of spores, remain before fungal systems can be widely applied in the field.

In the recent years, biotreatment has been proposed for the control of trace indoor air contaminants [52,53]. Indoor air is characterized by very low contaminant concentrations and application of biofiltration should consider issues such as the release of microorganisms and of unwanted odours, as well as excessive humidification of the treated air. The biofilter that has been reported for indoor application is somewhat different from those discussed in this chapter. It includes plants, living mosses and is

more like a small ecosystem [52]. Presumably, photosynthesis and the resulting plant residues may provide the system with the extra energy it requires to support active bacterial life, so that trace contaminant removal can be accomplished. The reported pollutant removal performance appears to be promising, and if this application matures to a commercial development, the impact could be very large, as indoor air quality is a major concern of industrialized countries.

Recently, effective treatment of low H<sub>2</sub>S concentrations (2-50 ppm<sub>v</sub>) has been shown in a high performance biotrickling filter [51]. The originality of the work lies in the fact that an existing chemical scrubber was converted to a biotrickling filter, keeping the original gas contact time of 1.6-2.3 seconds, i.e., a much shorter time than any previous biotrickling filtration studies. The scrubber vessel was reused and the conversion procedure was simple and relatively inexpensive. Reclaimed water was used as nutrient source for the process and for maintaining the pH in the biotrickling filter between 1.5 and 2.2. Under these conditions, successful treatment of H<sub>2</sub>S at rates comparable to those of chemical scrubbers was observed. H<sub>2</sub>S removal was in excess of 98% for inlet H<sub>2</sub>S concentrations as high as 30 to 50 ppm<sub>v</sub>. This corresponds to volumetric elimination rates of H<sub>2</sub>S of 95 to 105 g H<sub>2</sub>S m<sup>-3</sup> h<sup>-1</sup>. Such performance is exceptionally high compared with other biofilters or biotrickling filters removing low concentration of H<sub>2</sub>S, even at higher gas contact times. The authors speculate that a combination of high pollutant mass transfer rate due to the special polyurethane packing support that was used and optimum operating conditions (nutrient, pH, CO<sub>2</sub>) was responsible for the unprecedented performance. Significant removal of reduced sulphur compounds, ammonia and volatile organic compounds present in traces in the air was also simultaneously observed. The demonstration that H<sub>2</sub>S can be effectively treated biologically at contact times comparable to those of chemical scrubbers has substantial implications for odour control, as a large number of chemical scrubbers could be similarly converted to biotrickling filters, with substantial cost and environment health and safety benefits.

Highly chlorinated solvents remain some of the most challenging compounds as far as air biotreatment is concerned. This is because of their recalcitrance under aerobic conditions or the requirements of co-substrates for inducing cometabolism. Recently, Schwartz et al. [54] treated simulated landfill gas, consisting of an equimolar mixture of carbon dioxide and methane with traces of containing tetrachloroethylene (PCE) in a biofilter under anaerobic conditions. Sucrose was added as an additional energy and carbon source. Tetrachloroethylene was dechlorinated completely. The final end-product of biodegradation could not be identified, but the usual PCE biodegradation metabolites (vinyl chloride, dichloroethylene and trichloroethylene) were excluded. After an acclimation period the removal efficiency was higher than 98%. Such development could be applied to the remediation of contaminated groundwater, by sparging the aquifer with nitrogen, and treating chlorinated solvent vapours in a biofilter. In another study by Sun and Wood [55] a different approach was used for the treatment of trichloroethylene (TCE) vapours. A pure culture of *Burkholderia cepacia* PR<sub>123</sub> (TOM<sub>23C</sub>) that constitutively expresses toluene ortho-monooxygenase (TOM) was used to cometabolize TCE vapours in a biotrickling filter. Usually, aerobic biodegradation of TCE only occurs through cometabolism, and a growth substrate (e.g., toluene, methane, propane, phenol, or ammonia) is required to induce the expression of

the appropriate TCE-degrading enzyme. *B. cepacia* PR<sub>123</sub>, however, expresses TOM constitutively, which avoids the competitive inhibition by the inducer. Sun and Wood used a glucose solution as a carbon and energy source and observed TCE eliminations up to 200 times higher than previously reported. However, rapid inactivation of the TCE-degrading enzyme by TCE breakdown products (TCE epoxide) remained a problem. In both these applications, economical and technological challenges exist before deployment at the industrial scale.

The recent advances in molecular methods for studying microbial communities in complex environments has started to impact the research in vapour-phase bioreactors. These tools include DNA microarrays, 16S rDNA based analyses, fluorescence in-situ hybridization (FISH), 2D gel electrophoresis, specific gene probes, etc. [56] While these new tools have only recently been applied to vapour-phase bioreactors, they appear to be extremely promising in describing the complex ecology of biofilters and biotrickling filters [13,18,57,58,59]. A challenge for the application of these tools is to relate the observations made with these advanced tools to the performance of the reactor systems, so that the advanced tools are used to develop a better understanding of the process, or as a diagnostic for troubleshooting, or to support process optimization.

There are many other research issues in air biotreatment. One important one that has received little attention is this of the biodegradation kinetics of very low concentrations of pollutants in vapour-phase bioreactors. Clearly, a better understanding of the physiology of the process culture, of the induction of key enzymes and of the pollutant metabolism under the complex environmental conditions experienced in vapour-phase bioreactors is desirable. This should enable scientists and engineers to design better reactors that are more effective over a broader range of conditions, and a wider spectrum of pollutants. Another issue is this of pollutant mass transfer. As improvements in the biological kinetics are made, pollutant removal in gas-phase bioreactors may become limited by the rate at which pollutant is transferred to the microorganism. Hence, a better understanding of mass transfer in biofilters and biotrickling filters is required. Advances in this area will support the development of new packings and possible means to improve mass transfer. Undoubtedly, kinetic and mass transfer aspects are closely linked, and an integrated approach will be required.

## 5. References

- [1] Pomeroy, R.D. (1957) Deodorizing gas streams by the use of microbiological growths. US Patent 2.793.096.
- [2] Devlinny, J.S.; Deshusses, M.A. and Webster, T.S. (1999) Biofiltration for air pollution control. Lewis Publishers, Boca Raton, FL, USA, 300 pp.
- [3] van Groenestijn, J.W. and Hesselink, P.G.M. (1993) Biotechniques for air pollution control. Biodegradation. 4: 283-301.
- [4] Leson, G. and Winer, A.M. (1991) Biofiltration - An innovative air pollution control technology for VOC emissions. J. Air Waste Manage. Assoc. 41: 1045-1054.
- [5] Kennes, C. and Veiga, M.C. (2001). Conventional Biofilters. in Kennes C. and Veiga M.C. (Eds.) Bioreactors for Waste Gas Treatment, Kluwer Academic Publisher, The Netherlands, 47-98.
- [6] Deshusses, M.A. (1997) Biological waste air treatment in biofilters. Current Opinion in Biotechnology 8(3): 335-339.

- [7] Cox, H.H.J. and Deshusses, M.A. (1998) Biological waste air treatment in biotrickling filters. *Current Opinion in Biotechnology* 9(3): 256-262.
- [8] Cox, H.H.J and Deshusses, M.A. (2001) Biotrickling Filters. in Kennes C. and Veiga M.C. (Eds.) *Bioreactors for Waste Gas Treatment*, Kluwer Academic Publisher, The Netherlands, 99-131.
- [9] Kirchner, K.; Wagner, S. and Rehm, H.J. (1992) Exhaust gas purification using biocatalysts (fixed bacteria monocultures) - the influence of biofilm diffusion rate (O<sub>2</sub>) on the overall reaction rate. *Appl. Microbiol. Biotechnol.* 37: 277-279.
- [10] Zilli, M.; Converti, A.; Lodi, A.; Del Borghi, M. and Ferraiolo, G. (1993) Phenol removal from waste gases with a biological filter by *Pseudomonas putida*. *Biotechnol. Bioeng.* 41: 693-699.
- [11] Webster, T.S.; Deviny, J.S.; Torres, E.M. and Basrai, S.S. (1997) Microbial ecosystems in compost and granular activated carbon biofilters. *Biotechnol. Bioeng.* 53: 296-303.
- [12] Mason C.A.; Hamer, G. and Bryers, J.D. (1986) The death and lysis of microorganisms in environmental processes. *FEMS Microbiol. Rev.* 39: 373-401.
- [13] Sakano, Y. and Kerkhof, L. (1998) Assessment of changes in microbial community structure during operation of an ammonia biofilter with molecular tools. *Appl. Environ. Microbiol.* 64: 4877-4882.
- [14] Oh, Y.S. and Bartha, R. (1997) Removal of nitrobenzene vapors by a trickling air biofilter. *J. Ind. Microbiol. Biotechnol.* 18: 293-296.
- [15] Fortin, N.Y. and Deshusses, M.A. (1999) Treatment of methyl tert-butyl ether vapors in biotrickling filters. 1. Reactor startup, steady-state performance, and culture characteristics. *Environ. Sci. Technol.* 33: 2980-2986.
- [16] Bendinger, B.; Kroppenstedt, R.M.; Klatt, S. and Altendorf, K. (1992) Chemotaxonomic differentiation of coryneform bacteria isolated from biofilters. *Int. J. Syst. Bacteriol.* 42: 474-486
- [17] Juteau, P.; Larocque, R.; Rho, D. and Le Duy, A. (1999) Analysis of the relative abundance of different types of bacteria capable of toluene degradation in a compost biofilter. *Appl. Microbiol. Biotechnol.* 52(6): 863-868.
- [18] Friedrich, U.; Naismith, M.M.; Altendorf, K. and Lipski, A. (1999). Community analysis of biofilters using fluorescence in situ hybridization including a new probe for the Xanthomonas branch of the class Proteobacteria. *Appl. Environ. Microbiol.* 65: 3547-3554.
- [19] Moller, S.; Pedersen, A.R.; Poulsen, L.K.; Arvin, E. and Molin, S. (1996) Activity and three-dimensional distribution of toluene-degrading *Pseudomonas putida* in a multispecies biofilm assessed by quantitative in situ hybridization and scanning confocal laser microscopy. *Appl. Environ. Microbiol.* 12: 4632-4640.
- [20] Yadav, J.S. and Reddy, C.A. (1993) Degradation of benzene, toluene, ethylbenzene and xylenes (BTEX) by the lignin-degrading basidiomycete *Phanerochaete chrysosporium*. *Appl. Environ. Microbiol.* 59: 756-762.
- [21] Braun-Lüllemann, A.; Majcherzyk, A.; Tebbe, N. and Hüttermann, A. (1992) Bioluftfilter auf der Basis von Weißfäulepilzen. in *Biotechniques for Air Pollution Abatement and Odour Control Policies*. Dragt A.J. and van Ham J. (Eds.) Elsevier, Amsterdam, The Netherlands, 91-95.
- [22] Cox, H.H.J.; Moerman, R.E.; van Baalen, S.; van Heiningen, W.N.M.; Doddema, H.J. and Harder, W. (1997) Performance of a styrene-degrading biofilter containing the yeast *Exophiala jeanselmei*. *Biotechnol. Bioeng.* 53: 259-266.
- [23] Garcia-Pena, E. I.; Hernandez, S.; Favela-Torres, E.; Auria, R. and Revah, S. (2001) Toluene biofiltration by the fungus *Scedosporium apiospermum* TB1. *Biotechnol. Bioeng.* 76(1): 61-69.
- [24] Phae, C.G. and Shoda, M. (1991) A new fungus which degrades hydrogen sulphide, methanethiol, dimethyl sulphide and dimethyl disulfide. *Biotechnol. Lett.* 13: 375-380.
- [25] Woertz, J.R.; Kinney, K.A.; McIntosh, N.D.P. and Szaniszló, P.J. (2001) Removal of toluene in a vapor-phase bioreactor containing a strain of the dimorphic black yeast *Exophiala lecanii-corni*. *Biotechnol. Bioeng.* 75(5): 550-558.
- [26] Woertz, J.R.; Kinney, K.A. and Szaniszló, P.J. (2001) A fungal vapor-phase bioreactor for the removal of nitric oxide from waste gas streams. *J. Air Waste Manage. Assoc.* 51(6): 895-902.
- [27] Characklis, W.G. and Marshall, K.C. (1990) *Biofilms*. Wiley & Sons, New York, NY, USA, 796 pp.
- [28] de Beer, D.; Stoodley, P.; Roe, F. and Lewandowski, Z. (1994) Effects of biofilm structures on oxygen distribution and mass transport. *Biotechnol. Bioeng.* 43: 1131-1138.
- [29] Hugler, W.C.; Cantu-De la Garza J.G.; and Villa-Garcia, M. (1996) Biofilm analysis from an odor-removing trickling filter. in *Proc. Annual Meeting and Exhibition of the Air & Waste Management Association*, Air & Waste Management Association (Ed), Pittsburgh, PA, USA, paper 96-RA87A.04: 20 pp.

- [30] Holubar, P.; Andorfer, C. and Braun, R. (1999) Effects of nitrogen limitation on biofilm formation in a hydrocarbon-degrading trickle-bed filter. *Appl. Microbiol. Biotechnol.* 51: 536-540.
- [31] Wübker, S.M. and Friedrich, C. (1996) Reduction of biomass in a bioscrubber for waste gas treatment by limited supply of phosphate and potassium ions. *Appl. Microbiol. Biotechnol.* 46: 475-480.
- [32] Weber, F.J. and Hartmans, S. (1996) Prevention of clogging in a biological trickle-bed reactor removing toluene from contaminated air. *Biotechnol. Bioeng.* 50: 91-97.
- [33] Cox, H.H.J. and Deshusses, M.A. (1999) Biomass control in waste air biotrickling filters by protozoan predation. *Biotechnol. Bioeng.* 62: 216-224.
- [34] Woertz, J.R.; van Heiningen, W.N.M.; van Eekert, M.H.A.; Kraakman, N.J.R.; Kinney, K.A. and van Groenestijn, J.W. (2002) Dynamic bioreactor operation: Effects of packing material and mite predation on toluene removal from off-gas. *Appl. Microbiol. Biotechnol.* 58(5): 690-694.
- [35] Won, Y.S.; Cox, H.H.J.; Walton, W.E. and Deshusses, M.A. (2002) An environmentally friendly method for controlling biomass in biotrickling filters for air pollution control. in Proc. of the Annual Meeting and Exhibition of the Air & Waste Management Association, Air & Waste Management Association (Ed), Pittsburgh, PA, USA, paper #43554, 12 pp.
- [36] Deshusses, M.A. and Cox, H.H.J (2002) Biotrickling Filters for Air Pollution Control. In The Encyclopedia of Environmental Microbiology, Vol. 2. G. Bitton (Ed.), Wiley & Sons, New York, NY, USA, 782-795.
- [37] Cherry, R.S. and Thompson, D.N. (1997) Shift from growth to nutrient-limited maintenance kinetics during biofilter acclimation. *Biotechnol. Bioeng.* 56(3): 330-339.
- [38] Fürer, C. and Deshusses M.A. (2000) Biodegradation in biofilters: Did the microbe inhale the VOC? in Proc. of the Annual Meeting and Exhibition of the Air & Waste Management Association, Air & Waste Management Association (Ed), Pittsburgh, PA, USA, paper #799, 13 pp.
- [39] Schönduvel, P.; Sára, M. and Friedl, A. (1996) Influence of physiologically relevant parameters on biomass formation in a trickle-bed bioreactor used for waste gas cleaning. *Appl. Microbiol. Biotechnol.* 45: 286-292.
- [40] Laurenzis, A.; Heits, H.; Wübker, S.M.; Heinze, U.; Friedrich, C. and Werner, U. (1998) Continuous biological waste gas treatment in stirred trickle-bed reactor with discontinuous removal of biomass. *Biotechnol. Bioeng.* 57: 497-503.
- [41] Smith, F.L.; Sorial, G.A.; Suidan, M.T.; Breen, A.W.; Biswas, P. and Brenner, R.C. (1996) Development of two biomass control strategies for extended, stable operation of highly efficient biofilters with high toluene loadings. *Environ. Sci. Technol.* 30: 1744-1751.
- [42] Cox, H.H.J. and Deshusses, M.A. (1999) Chemical removal of biomass from waste air biotrickling filters: screening of chemicals of potential interest. *Wat. Res.* 33: 2383-2391.
- [43] Devinny, J.S. and Hodge, D.S. (1995) Formation of acidic and toxic intermediates in overloaded ethanol biofilters. *J. Air Waste Manage. Assoc.* 45: 125-131.
- [44] Morgenroth, E.; Schroeder, E.D.; Chang, D.P.Y. and Scow, K.W. (1996) Nutrient limitation in a compost biofilter degrading hexane. *J. Air Waste Manage. Assoc.* 46: 300-308.
- [45] Zhu, X.; Rihn, M.J.; Suidan, M.T.; Kim, B.J. and Kim, B.R. (1996) The effect of nitrate on VOC removal in trickle bed biofilters. *Wat. Sci. Technol.* 34: 573-581.
- [46] Chou, M.S. and Huang, J.J. (1997) Treatment of methyl ethyl ketone in air stream by biotrickling filters. *J. Environ. Eng.* 123(6): 569-576.
- [47] Pedersen, A.R. and Arvin, E. (1995) Removal of toluene in waste gases using a biological trickling filter. *Biodegradation* 6: 109-118.
- [48] Kong, Z.; Farhana, L.; Fulthorpe, R.R. and Allen, D.G. (2001) Treatment of volatile organic compounds in a biotrickling filter under thermophilic conditions. *Environ. Sci. Technol.* 35(21): 4347-4352.
- [49] Wright, W.F.; Schroeder, E.D.; Chang, D.P.Y. and Romstad, K. (1997) Performance of a pilot-scale compost biofilter treating gasoline vapor. *J. Environ. Eng.* 123(6): 547-555.
- [50] Hugler, W.; Acosta, C. and Revah, S. (1999) Biological removal of carbon disulfide from waste air streams. *Environ. Prog.* 18(3): 173-177.
- [51] Gabriel, D.; Cox, H.H.J.; Brown, J.; Torres, E. and Deshusses, M.A. (2002) Biotrickling filters for POTWs air treatment: Full-scale experience with a converted scrubber. in Proc. Odors and Toxic Air Emissions 2002. Water Environment Federation (Ed.), Alexandria, VA, USA, 13 pp.
- [52] Darlington, A.B.; Dat, J.F. and Dixon, M.A.(2001) The biofiltration of indoor air: Air flux and temperature influences the removal of toluene, ethylbenzene, and xylene. *Environ. Sci. Technol.* 35(1): 240-246.

- [53] Darlington, A.; Chan, M.; Malloch, D.; Pilger, C. and Dixon, M.A. (2000) The biofiltration of indoor air: Implications for air quality. *Indoor Air-Int. J. Indoor Air Qual. Climate*, 10(1): 39-46.
- [54] Schwarz, B.C.E.; Devinny, J.S. and Tsotsis, T.T. (1999) Degradation of PCE in an anaerobic waste gas by biofiltration. *Chem. Eng. Sci.* 54(15-16): 3187-3195.
- [55] Sun, A.K. and Wood, T.K. (1997) Trichloroethylene mineralization in a fixed-film bioreactor using a pure culture expressing constitutively toluene ortho-monooxygenase. *Biotechnol. Bioeng.* 55: 674-685.
- [56] Wilderer, P.A.; Bungartz, H.J.; Lemmer, H.; Wagner, M.; Keller, J. and Wuertz, S. (2002) Modern scientific methods and their potential in wastewater science and technology. *Wat. Res.* 36(2): 370-393.
- [57] Tresse, O.; Lorrain, M.J. and Rho, D. (2002) Population dynamics of free-floating and attached bacteria in a styrene-degrading biotrickling filter analyzed by denaturing gradient gel electrophoresis. *Appl. Microbiol. Biotechnol.* 59(4-5): 585-590.
- [58] Alexandrino, M.; Knief, C. and Lipski, A. (2001) Stable-isotope-based labeling of styrene-degrading microorganisms in biofilters. *Appl. Environ. Microbiol.* 67: 4796-4804.
- [59] Malhautier, L.; Degrange, V.; Guay, R.; Degorce-Dumar, J.R.; Bardin, R. and Le Cloirec, P. (1998) Estimation size and diversity of nitrifying communities in deodorizing filters using PCR and immunofluorescence. *J. Appl. Microbiol.* 85: 255-262.