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The Two-Phase Water/Silicon Oil Bioreactor Prospects in Off-Gas Treatment

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Abstract

Research was carried out to develop a biphasic biologic reactor able to clean the gas effluents polluted by volatile organic compounds. Initially, *Rhodococcus erythropolis* T 902.1 was selected on the basis of its capacity to degrade isopropylbenzene (IPB). The effect of gas flow and IPB concentration on the biodegadation of IPB was evaluated. The results show that the use of silicon oil allows large quantities of IPB to be absorbed within the medium of biologic abatement. On the other hand, the biodegradation rate was directly correlated to the inlet flow of IPB. Thus, the reactor presents interesting opportunities for the biologic treatment of gas effluents.

Index Entries: Two-phase bioreactor; silicon oil; volatile organic compounds; off-gas treatment.

Introduction

Research was carried out within the framework of gaseous treatment with the aim of developing a biphasic "water/silicon oil" reactor. Silicon oil was used to allow a better biologic abatement of the aromatic organic compounds by improving their solubility within the two-phase bioreactor. Initially, a bacterial strain (*Rhodococcus erythropolis* T 902.1) was selected on the basis of its good capacity to degrade isopropylbenzene (IPB), a compound selected as a model for the family of benzene. Various studies were performed in order to improve the degradation of volatile organic compounds (VOC) in gas effluents, particularly by improving the mass transfer of VOC within the reactor. Yeom and Daugulis (*1*) recommend the use of a biphasic biologic reactor whose organic phase (hexadecane) constitutes one-third of the reactional medium. Hexadecane presents the property of

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being slightly toxic for microorganisms. A similar process showed its effectiveness on the biodegradation of a mixture of organic pollutants (BTEX) by a *Pseudomonas* sp. strain (*2*,*3*).

Van Ede et al. (*4*) showed, by the theory called the Film Variable Holdup model, an enhancement in the gas transfer owing to the presence of a dispersed octene phase. Moreover, it was shown that the transfer rate and the biodegradation of apolar pollutants in biologic waste gas treatment, as well as the transfer rate of oxygen, can be enhanced by a dispersed organic solvent (FC40; 10% [v/v]). Theoretically, it could be shown that the addition of solvent has a more significant effect on the enhancement of transfer rate in the case of poorly water-soluble compounds compared with moderately water-soluble ones (*5*). On the other hand, Nielsen et al. (*6*) showed an increase in the oxygen's solubility within the system by the addition of hexadecane. This increase provides a potential for enhancement of oxygen's mass transfer rate.

Dumont and Delmas (*7*) reviewed the general concept of oil-in-water systems and demonstrated the ability of an immiscible oil phase to influence transfer from the gas phase to the aqueous phase. Another way to enhance oxygen gas transfer rate is proposed by using soybean oil-in-water dispersion (*8*).

In this context, silicon oil can also be used to reproduce the effect of a solvent. Thus, Budwill and Coleman (*9*) developed a biofilter with silicon oil addition that improves hexane biodegradation. Moreover, Gardin et al. improved the biodegradation of xylene and butyl acetate by using an aqueoussilicon oil two-phase system.

Finally, Aldric (*10*) showed the value of the use of silicon oil in a proportion of 10% in a two-phase bioreactor. Silicon oil allows an important improvement in gas retention into biphasic medium, i.e., the volume of gas retained within a volume of reactional medium liquid. Furthermore, the coefficient of oxygen mass transfer is not decreased compared to an aqueous medium. These findings demonstrate the possible application of silicon oil in the field of off-gas treatment. Indeed, silicon oil allows a significant solubilizaton of VOC. The object of the present study thus was to specify the influence of silicon oil on the biodegradation of IPB, selected as the model for the BTEX compounds.

Materials and Methods

Strain and Chemicals

The *R. erythropolis* strain T 902.1 was obtained from the collection of the Walloon Center for Industrial Biology (Gembloux, Belgium) (*11*,*12*). All substrates and other chemicals were purchased from VWR (Leuven, Belgium) or Aldrich (Bornem, Belgium).

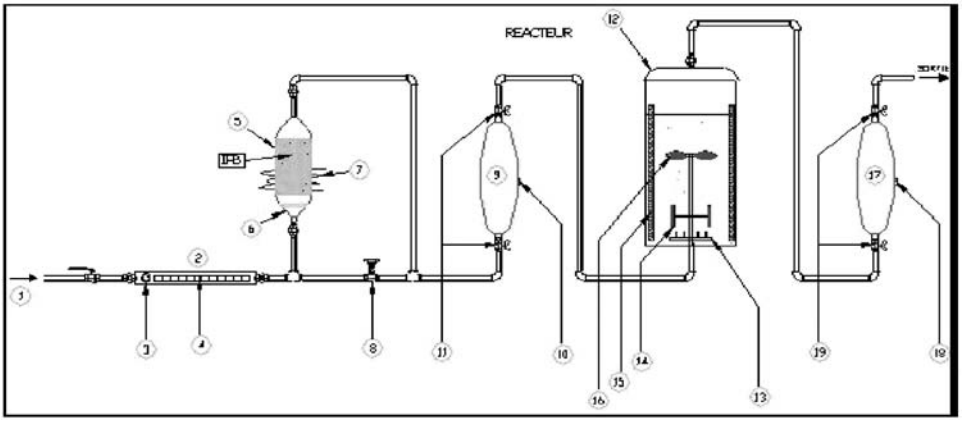

Fig. 1. Schematic representation of bioreactor assembly.

Bioreactor and Assembly

The stirred bioreactor used for biodegradation (LSL Biolafitte BL06.1; Saint Germain en Laye, France) was described by Aldric (10). Its reactional volume reaches 4.5 L and the stirring speed was maintained at 600 rpm. Figure 1 presents a schematic of the biorenctor's assembly.

The IPB-concentrated gas is generated by stripping within a thermostatized glass bottle. The gas flow is permanently controlled by a flow meter. The concentration of IPB in the gas coming into the bioreactor is controlled by an adjustable mixture between polluted gas and air.

Experimental Design

The Doehlert design (*13*) was selected to evaluate the effect of two factors on the biodegradation: gas flow and IPB concentration of the inlet gas. The values of concentration and flow corresponding to the experimental points are selected so that the pairs concentration-flow are located at the angles of a perfect hexagon in bidimensional space "Concentration/Flow." Nevertheless, adjustment of the parameters to their fixed values is difficult because of the technical constraints. Statistical analysis of the results thus requires taking into account the real average values of the two studied parameters. The shifted points obtained in experiments are indicated in Fig. 9. The area defined by the Doehlert design is a circle in a two-dimensional space. Five levels were retained (mg/Nm^3 i.e., m^3 under normal conditions of temperature and pressure [25°C, 101,325 Pa]) for the "concentration" factor and three levels for the "flow" factor (L/min).

The 10 performed experiments are included and characterized in Table 1. Shaded experiments represent the repetitions of the central experiment.

Tables 1
Experiments Utilizing the Doehlert Design

Experiments	Concentration (mg/N · m^3)	Gas flow (L/min)
1	3050	4.5
2	1575	1.47
3	3050	4.5
4	1575	7.53
5	4525	7.53
6	4525	1.47
7	3050	4.5
8	100	4.5
9	3050	4.5
10	6000	4.5

Statistical analysis of the results was carried out with the software SAS⟩. The Doehlert design *(13)* permits the establishment of a second-degree regression model with interactions of the second order.

Sampling and Analytical Methods

Gas samples are regularly taken from the inlet and exit as well as samples of bubbles in the liquid reactional medium. IPB concentration was estimated using an HS 40 XL Perkin Elmer headspace sampler (for liquid samples) and a Hewlett Packard 5890 gas chromatograph equipped with an Alltech. Deerfield EC-WAX column and a flame ionization detector (for gas samples). Temperatures of the injector, column, and detector were, respectively, 153, 150, and 250°C.

Implementation of Biomass

The culture of *R. erythropolis* in 868 medium (20 g/L of glucose, 20 g/L of casein peptone, 10 g/L of yeast extract) was harvested after 64 h (optical density at 600 nm = 1.4). The inoculum for the biologic reactor was obtained by centrifuging 2.25 L of this culture. The pellet obtained was washed twice and diluted in 200 mL of saline water (6 g/L of NaCl). The inoculum was then introduced into the bioreactor, where the medium for biodegradation was composed of silicon oil (10% [v/v]) and aqueous medium M284 (90% [v/v]) whose composition was as follows: 6.06 g/L of Tris-HCl, 4.68 g/L of NaCl, 1.49 g/L of KCl, 1.07g/L of NH_4Cl, 0.43 g/L of Na_2SO_4, 0.20 g/L of $MgCl_2 \cdot 6H_2O$, 40 mg/L of $Na_2HPO_4 \cdot 2H_2O$, 30 mg/L of $CaCl_2 \cdot 2H_2O$, 4.8 mg/L of Fe(III)NH_4 citrate, 0.144 mg/L of $ZnSO_4 \cdot 7H_2O$, 0.1 mg/L of $MnCl_2 \cdot 4H_2O$, 0.062 mg/L of H_3BO_3, 0.19 mg/L of $CoCl_2 \cdot 6H_2O$, 0.017 mg/L of $CuCl_2 \cdot 2H_2O$, 0.024 mg/L of $NiCl_2 \cdot 6H_2O$, 0.036 mg/L of $Na_2MoO_40 \cdot 2H_2O$; 1 g/L of ethanol.

Determination of Concentrations and Flows

Only the data corresponding to a stabilization of the IPB concentration within the liquid medium were retained. The following relation is indeed correct because IPB consumption by the biomass (term to the left) is equal to the transferred quantity (term to the right):

$$\left(Q_{\text{in}} - Q_{\text{out}}\right) = K_L a\left(C_L^O - C_L\right)$$

where $K_l a$ is the total coefficient of mass transfer for IPB (min^{-1}); Q_{in} and Q_{out} are, respectively, the inlet flow and outlet flow of IPB (mg IPB/[min · L] of reactional medium); $C^0{}_L$ is the saturating concentration of IPB in the biphasic liquid medium (mg/Nm^3); and C_L is the equilibrium concentration of IPB in the biphasic liquid medium (mg/L).

Inlet flow and outlet flow of IPB are given by the following equations:

$$Q_{\text{in}} = \frac{\text{Conc.in}_{\text{mean}} \times \text{flow}}{\text{vol}}$$

$$Q_{\text{out}} = \frac{\text{Conc.out}_{\text{mean}} \times \text{flow}}{\text{vol}}$$

$$\text{Conc}_{\text{mean}} = \sum_{i-1\ n} \frac{\text{Conc}_i \times \left(t_i - t_{i-1}\right)}{t_{tot}}$$

in which $\text{Conc}_{\text{mean}}$ is the weighted average of concentrations during an equilibrium phase (mg/Nm^3), Conc_i is the specific concentration (mg/Nm^3) measured by gas injection at time $t = i$, flow is the flow of gas effluent loaded in IPB, t is the time (h), and vol is the volume of the reactional medium.

Determination of Biodegradation Rate

The biodegradation rate can be easily calculated using the following formula:

$$\text{Biodegradation rate } \% = 100 \times \left(Q_{\text{in}} - Q_{\text{out}}\right)$$

in which Q_{in} and Q_{out} are as described in the previous section.

Results and Discussion

Follow-Up of IPB Biodegradation in a Gas Effluent

Previously, *R. erythropolis* T 902.1 showed good potential for biodegradation of aromatic compounds (benzene, toluene, and xylene) in liquid medium. Moreover, this strain has very interesting properties to

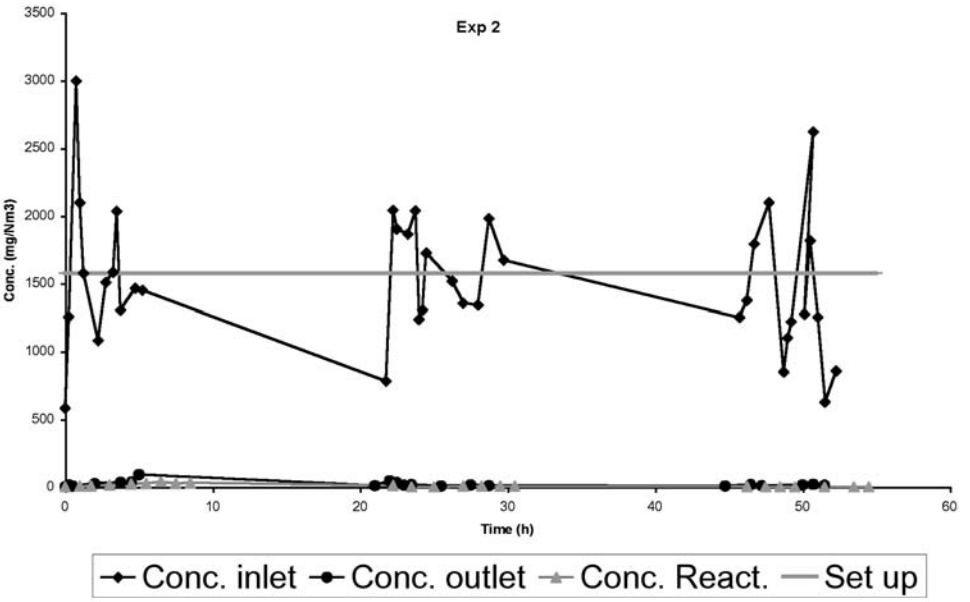

Fig. 2. Example of follow-up of experiment 2 during 3 d of experimentation.

produce starter cultures, specifically owing to the drying procedure. Industrial applications are thus possible *(11).*

More specifically, this strain contains a catabolic plasmid conferring it the ability to degrade toluene and other aromatic compounds such as IPB, selected as the model within the framework of the present research *(12).* In our research, this strain was implemented in a two-phase bioreactor that was developed to degrade IPB in gas effluents. The use of silicon oil is unique for the proposed two-phase reactor. This phase is used to improve the transfer and biodegradation of IPB from the off-gas.

A laboratory-scale two-phase reactor was developed to study the influence of IPB concentration and flow of gas (Fig. 1). The device permits generation of a polluted effluent with a flow and a specified IPB concentration. This device is followed by sample points within the reactional liquid medium, at the entry and the exit of the bioreactor (see Fig. 1). Follow-up makes it possible to determine the rate of IPB biodegradation and the loading capacity of the bioreactor for the various values of the studied parameters (flow and IPB concentration). Moreover, in order to follow the two studied parameters, a Doehlert design was implemented. The experiments included in the Doehlert design were monitored for 3 d. As described previously, the measurements taken into account to determine the parameters (biodegradation rate and loading capacity) are those corresponding to a stability of the concentration in the liquid medium (consumed quantity = transferred quantity).

The evolution of IPB concentration in inlet gas, in outlet gas, and within the liquid medium is given as an example in Figs. 2 and 3 for the

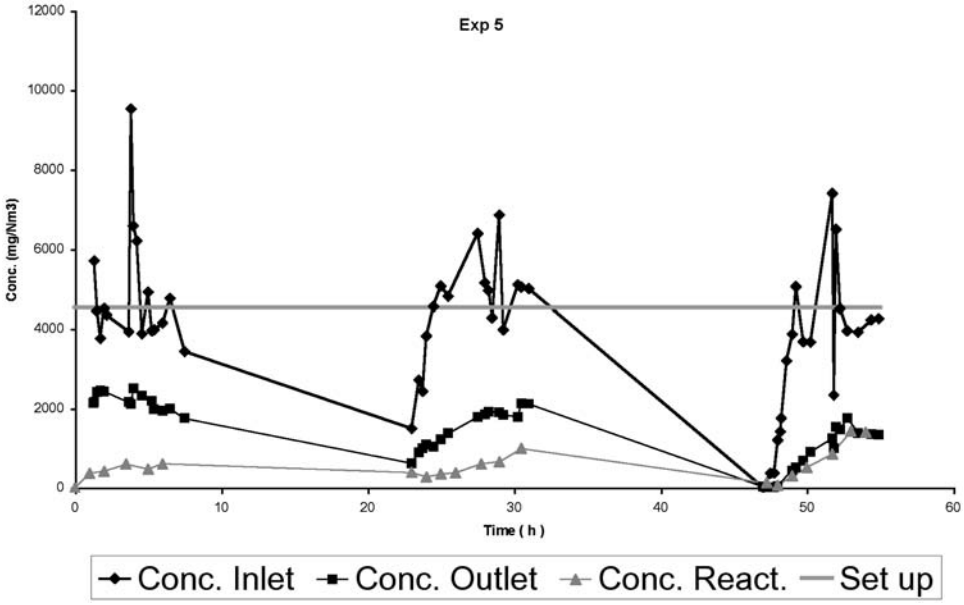

Fig. 3. Example of follow-up of experiment 5 during 3 d of experimentation.

two most significant experiments corresponding, respectively, to a low flow and a high flow of IPB (mg/[min · L])

The experiment presented in Fig. 2 corresponds to an IPB flow of reactional medium of 0.60 mg/(min · L). Both concentration within the liquid and concentration in the outlet gas of the bioreactor were very low, representing a very good biodegradation yield.

The experiment presented in Fig. 3 corresponds to the highest loading of IPB (9.47 mg/[min · L]) and shows the evolution of the IPB concentration for this experiment. It is obvious that the concentration of outlet gas was high (1727 mg/Nm^3 on average), but compared with the concentration in the inlet gas and to the gas flow, the biodegradation yield remained quite acceptable.

These two examples show that the biodegradation of IPB by the selected strain was important, including for high loads of IPB (>9 mg/[min · L]; 540 g/[m^3 · h]).

To define the potentialities of the proposed bioreactor, the biodegradation rate (percentage of IPB degraded by the biomass) and the loading capacity (mg/[min · L]) were given for each experiment.

Biodegradation Rate Functions of Flow and Concentration

Biodegradation rate is the relationship between the quantity of pollutant degraded by the biomass (mg/[min · L]) and the flow of pollutant entering the bioreactor (mg/[min · L]). It expresses the efficiency of the bioreactor for each pair (concentration-flow).

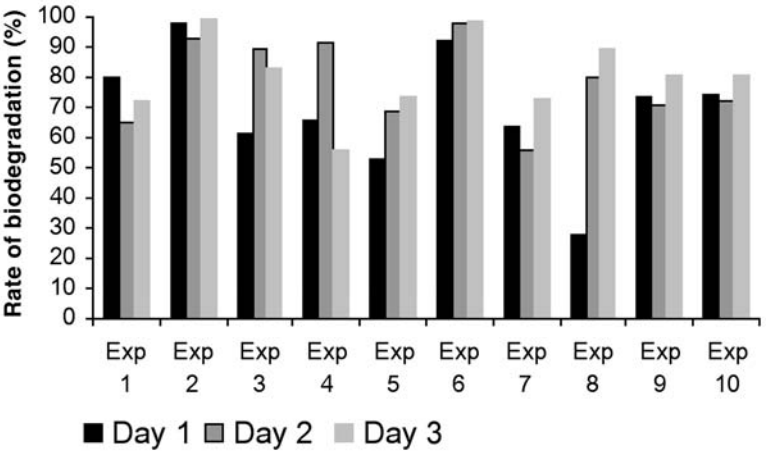

Fig. 4. Average rate of biodegradation.

For each experiment of Doehlert design, the biodegradation rate of the IPB was calculated as follows:

$$\tau = \frac{100 \times Q_{in} - Q_{out}}{Q_{in}}$$

In which τ is the biodegradation rate of IPB (%); and Q_{in} and Q_{out} are, respectively, the flows of IPB entering and leaving the bioreactor (mg/[min · L]). The biodegradation rate of IPB for each experiment and each day is shown in Fig. 4. The biodegradation rates were high in all experiments, except experiments 5 and 7. In the implemented range of flow and concentration, the biodegradation rate was never >53% except for the first day of experiment 8. After comparing the biodegradation rate according to the days of experimentation, it was observed that the biodegradation rate was often the highest at the end of the experiment (third day). This can be explained by an adaptation phase of the biomass to the presence of IPB in addition to an increase in biomass.

As described previously, the objective of this research is to evaluate the effect of the gas flow and the IPB concentration on the effectiveness of the proposed bioreactor, i.e. on the biodegradation rate. It is why a statistical analysis of the results was carried out.

Statistical Analysis of Results

Statistical analysis of the results, according to the response surface method, was carried out in order to establish a second-degree regression model between the biodegradation rate and the two chosen parameters: gas flow (L/min) and IPB concentration (mg/Nm^3). Considering that the cells must be adapted, only the values of biodegadation rate corresponding to the third day were taken into account for the statistical analysis.

Table 2
Significance of Coefficients

Variable	*P* value ($\alpha = 0.05$)
Initial *Y*-axis	< 0.0001
$Conc_{in}$	0.5682
Q	0.0017
Interaction $Conc_{in} \cdot Q$	0.1811
Q^2	0.1218
$Conc_{in}^2$	0.1411

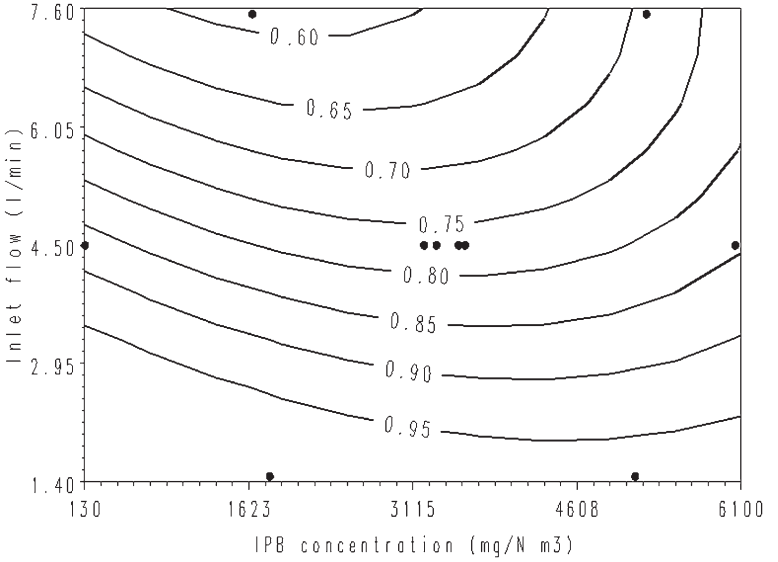

Fig. 5. Diagram of iso-response presenting the evolution of biodegradation rate according to inlet flow (L/min) and IPB concentration (mg/N·m^3).

For this purpose, the values of biodegradation rate measured were transformed by the arcsine square root transformation. This traditional transformation permits observation of the conditions for application for analysis of the results according to Doehlert *(13).* On the basis of the aforementioned considerations, a second-degree regression model can be proposed:

$$\arcsin^{-1}\Gamma = 1.987 - 0.216Q - 1.4 \times 10^{-4}Conc_{in} + 1.12 \times 10^{-4}Q \times Conc_{in} + 9.97 \times 10^{-3}Q^2\ 1.29 \times 10^{-8}Conc_{in}^2,\ R^2 = 0.9399$$

in which *T* is the IPB biodegradation rate (%); *Q* is the inlet flow of gas (L/min), first factor; and $Conc_{in}$ is the IPB concentration of inlet flow (mg/Nm^3), second factor.

For each estimated coefficient, Table 2 presents the *p* value. As shown in Table 2, the first-degree coefficient of the flow (0.0017) was highly significant. The factor flow (*Q*) was thus most influential on the biodegradation rate. In addition, Fig. 5 shows the combined effect of flow and concentration of the

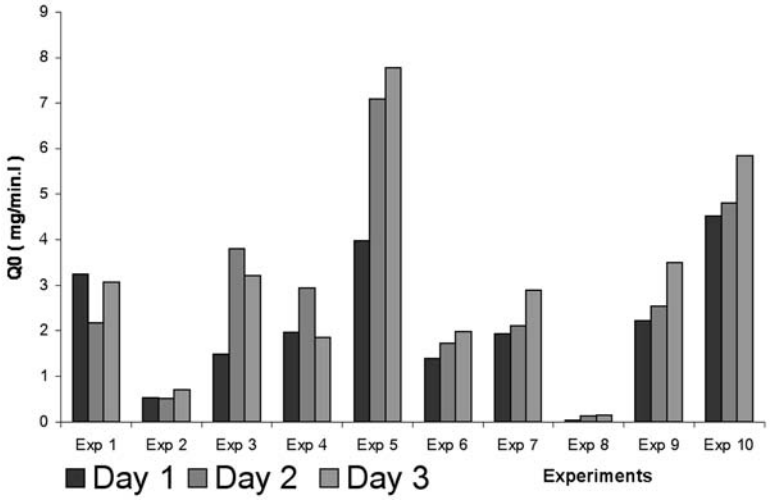

Fig. 6. Loading capacity of bioreactor.

effluent on the biodegradation of IPB. The contours of the second-order equation indicate the "concentration-flow" pairs, for which the same biodegradation rate was observed. By means of this diagram, it is possible to estimate the biodegradation rate for a flow-concentration pair. The shape of the diagram indicates that the variation of the flow induces a strong variation of the biodegradation rate. On the other hand, for the same flow, the variation of effluent concentration slightly influences the biodegradation rate. These observations can be explained by the fact that the two-phase bioreactor is able to transfer and absorb great quantities of pollutants only if the superficial gas velocity, generated by a high flow, is not too high into the bioreactor.

Loading Capacity for Experiments

The biodegradation rate characterizes the efficiency of a process. However, it is also important to determine the limits of the bioreactor. Therefore, the loading capacity can be defined as the absolute quantity of IPB degraded by the biomass per unit of time and unit of reactional volume: $Q_0 = Q_{in} - Q_{out}$ (mg/[min · L]). Figure 6 presents the described loading capacity. The loading capacity is logically correlated with the flow of IPB from the inlet gas (mg/[min · L]). However, the limit seems to be reached in experiment 5, with a loading capacity of 7.5 mg/(min · L) (450 g/[m^3 · h]). This limit can be explained by Fig. 7, which shows the IPB concentration in the biphasic liquid medium. Experiments 5, 7, and 9 (Fig. 7) show that the concentration in the liquid medium reached approx 1200 mg/L. This concentration within the liquid medium (C_L) seems to be a factor limiting the mass transfer of IPB. Indeed, when the C_L concentration increased, the potential of transfer ($C^0{}_L$–C_L) decreased.

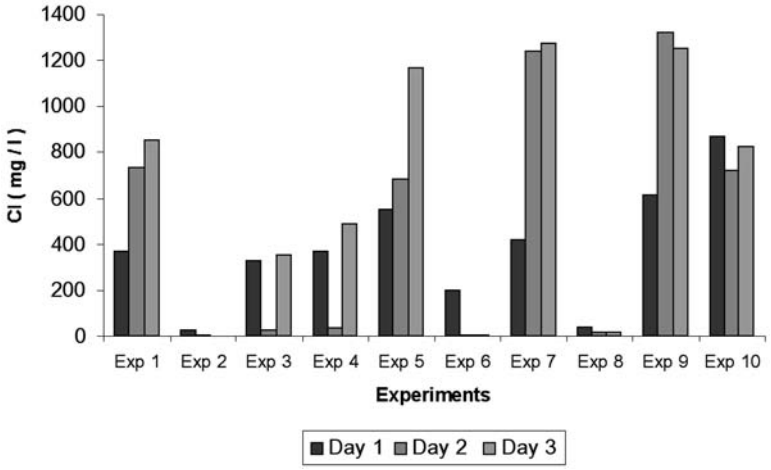

Fig. 7. Equilibrium concentration into biphasic biological reactor.

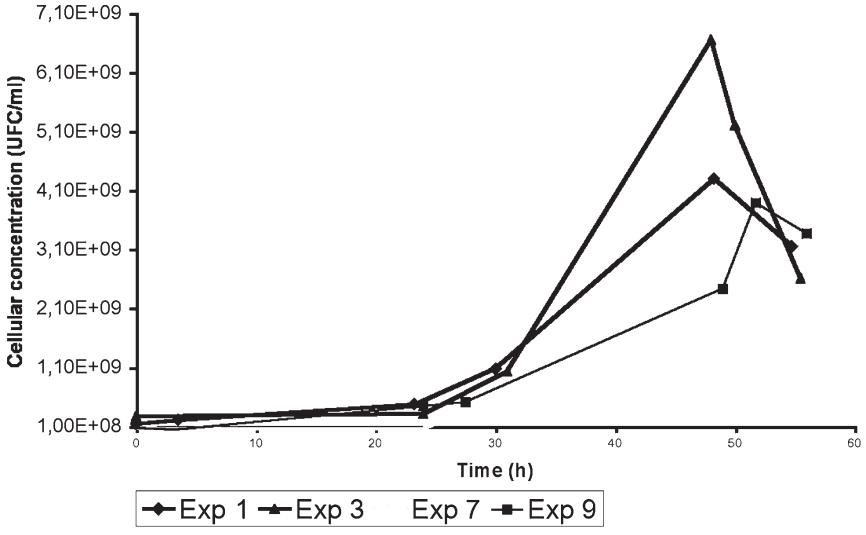

Fig. 8. Evolution of cellular concentration.

Evolution of Cellular Concentration in Bioreactor

The initial cellular concentration within the bioreactor is standardized. However, the evolution of the biomass is variable from one experiment to another. It is thus important to discuss the influence of cell multiplication on the effectiveness of the biodegradation. The evolution of cellular concentrations is presented in Figs. 8 (four repetitions of the central experiment) and 9 (other individual experiments).

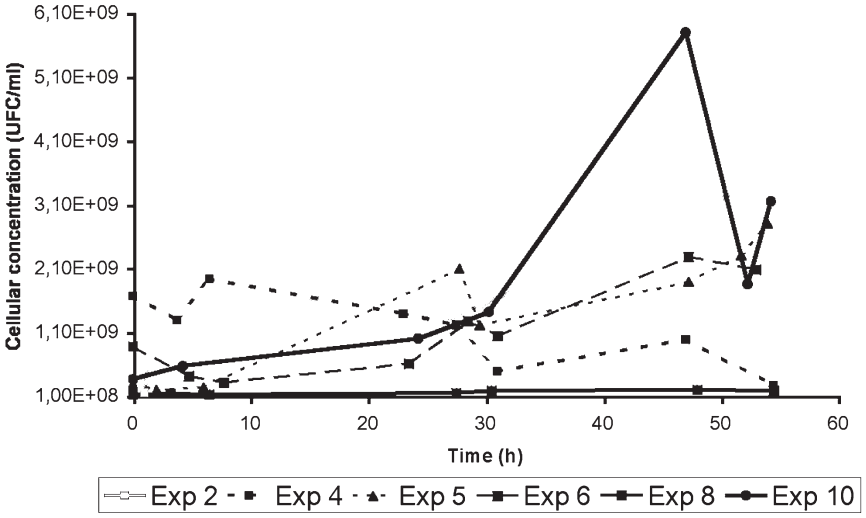

Fig. 9. Evolution of cellular concentration.

The variability of observed growth for a repetition of the central experiment (Fig. 8) indicates that there was not a relationship between the quantity of pollutant per unit of time and cellular multiplication. However, and generally, an adaptation phase of microorganisms was observed at the beginning of the experiment (24 h). The cellular concentration increased then slightly decreased during experiments. This observation must be owing to the increase in the degradation rate functions of time. It should be noted that cell multiplication was minimal for experiments corresponding to the lowest flows of IPB (0.57 and 0.11 mg/[min · L] for experiments 2 and 8, respectively). For small quantities of added carbon substrates, cell multiplication was then very low. On the other hand, growth was observed during the experiments with higher potential carbon sources.

In addition, the quadruple repetition of the central experiment (Fig. 8) allows comparision of four identical experiments and shows that the most significant biodegradation rate (82%; experiment 3) was obtained when the cell multiplication was the highest. Cellular concentration thus plays a role in the effectiveness of the bioreactor.

Conclusion

The results show that the use of a two-phase bioreactor with silicon oil as the second phase allows absorption of large quantities of IPB and good biologic degradation with the appropriate microorganism. Analysis of the results also shows that the biodegradation rate seems to be related mainly to the gas flow (L/min) compared to the IPB concentration (mg/Nm^3) in the gas effluent.

A limit of concentration in the liquid medium was also found, which led to a transfer limitation of the IPB from polluted gas toward the liquid medium by a reduction in the potential of transfer ($C^0{}_L$–C_L). In addition, it was shown that an increase in the cellular concentration during the experiment improved the effectiveness of the bioreactor. Thus, in the tested configuration, the two-phase bioreactor was able to degrade 7.5 mg/(min · L) of reactional medium (450g/[m^3 · h]).

It was also shown that an adaptation step of the biomass was necessary to reach substantial rates of abatement. However, the biomass was maintained and even increased during the experimentation.

The bioscrubbers usually used in the off-gas treatment were able to treat a polluted effluent with a concentration of 1000 mg/Nm^3 at a flow of 1.5 L/min (90 m^3/[m^3 · h]). This corresponds to a flow of 1.5 mg/(min · L) of bioscrubber *(14).*

The proposed reactor presents interesting opportunities in the biologic treatment of gas effluents polluted by aromatic compounds in high concentration. The suggested process might be applied in the range of concentration and flow for which thermal oxidation is too expensive (between 1 and 7 g/Nm^3).

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