# *Microbial solvent regeneration in biotreatment of air contaminated by styrene*

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**ABSTRACT**

In this study, the biodegradation of a styrene-polluted waste gas in a reactor containing 5 L of a biphasic mixture (10:90% v/v) of organic solvent (silicone oil) – water – biomass was investigated to establish the ability of a microbial solvent regeneration. Reproducible microbial solvent regenerations have been observed. The regeneration time, which increases with the increase of the styrene load (varying from 543 to 1800 mg), leads to elimination capacity up to 48 $g_{styrene}.m^{-3}{}_{mixture}.h^{-1}$. The solvent regeneration requires roughly 1.2 molecule of oxygen per molecule of styrene and corresponds to the first steps of the biodegradation of the styrene.

## 1 INTRODUCTION

Styrene is of major importance in the petrochemical and polymer-processing industries, which can contribute to the pollution of natural resources via the release of styrene-contaminated effluents and off-gases. Generally the produced polluted air flows are high in volume with low styrene concentrations (around 200 $mg.m^{-3}$) corresponding to the application area of bioprocesses and some biofilter utilizations are described (Cox *et al.*, 1996; Arnold *et al.*, 1997; Jorio *et al.*, 2000; Zilli *et al.*, 2001; Dehghanzadeh *et al.*, 2005) but with the needing of high bed volume. Another possibility in biological air treatment is the use of a bioscrubber, combination of a column for the pollutant air to liquid transfer and a biological reactor for the solvent recycling. However, for waste gases containing hydrophobic compounds having low solubility in water (like styrene), it is necessary to use a mixture of a non-biodegradable organic solvent and water. The pollutant is preferentially transferred from the gas phase to the organic solvent, and

diffuses to the aqueous phase in order to be degraded by the microorganisms. Thus complete regeneration of the mixture of organic solvent – water is possible. There are only few studies devoted to the regeneration of the organic solvent for styrene biodegradation (see Dumont *et al.*, 2006a). Consequently, the aim of this study is to present preliminary results about a solvent regeneration due to the degradation of the styrene by a mixed culture able to use this molecule as the unique source of carbon. Silicone oil is specifically used as solvent and the regeneration is followed by the measure of the oxygen variation in the gas phase in a batch reactor.

## 2. MATERIALS AND METHOD

### 2.1 Microorganisms and chemical

The mixed culture was obtained from the Nantes (France) wastewater treatment plant. Styrene was used as the sole source of carbon and energy and a nutrient solution consisting in an aqueous solution of $H_8N_2O_4S$ and $H_2KO_4P$ was used according to the quantity of carbon to keep the C/N/P ratio around 100/5/1. The biomass was progressively acclimated to styrene.

Silicone oil (Rhodorsil fluid 47V5, dimethylpolysiloxane) was obtained from Rhodia Company. The physical properties at 25°C are: viscosity, 5 mPa.s; density, 910 $kg.m^{-3}$; styrene solubility, 38g/L (Dumont *et al.*, 2006b); oxygen solubility, 7 higher than in water (Dumont *et al.*, 2006a). The styrene solubility in water at 25°C is 320 mg/L (Kirk-Othmer, 1983).

### 2.2. Experimental setup

The description of the reactor used in thus study is shown in Figure 1. The reactor has an 11.5 L total volume (height 0.33 m, diameter 0.21 m). In the experiments, air was supplied from a compressor and sparged through an elliptical distributor (75x150mm) with 50 holes (1mm diameter). All experiments were carried out at a constant temperature of 25°C maintained by a thermostatic bath. The total volume of the mixture was 5 L (silicone oil volume fraction: 10%) allowing to absorb roughly 3g of styrene (Dumont *et al.*, 2006b). Each experiment was carried out according to the following sequential procedure:

1) A synthetic waste gas stream was prepared by passing compressed air through a styrene generator filled with liquid styrene (flowmeters 3 and 4 opened; valves 1 and 5 opened; valve 2 closed). The known styrene stream bubbled through the mixture silicone oil – water – biomass during 1 hour in order to be absorbed by the liquids. The styrene concentrations in the gas phase were measured simultaneously at the inlet and outlet of the reactor using a

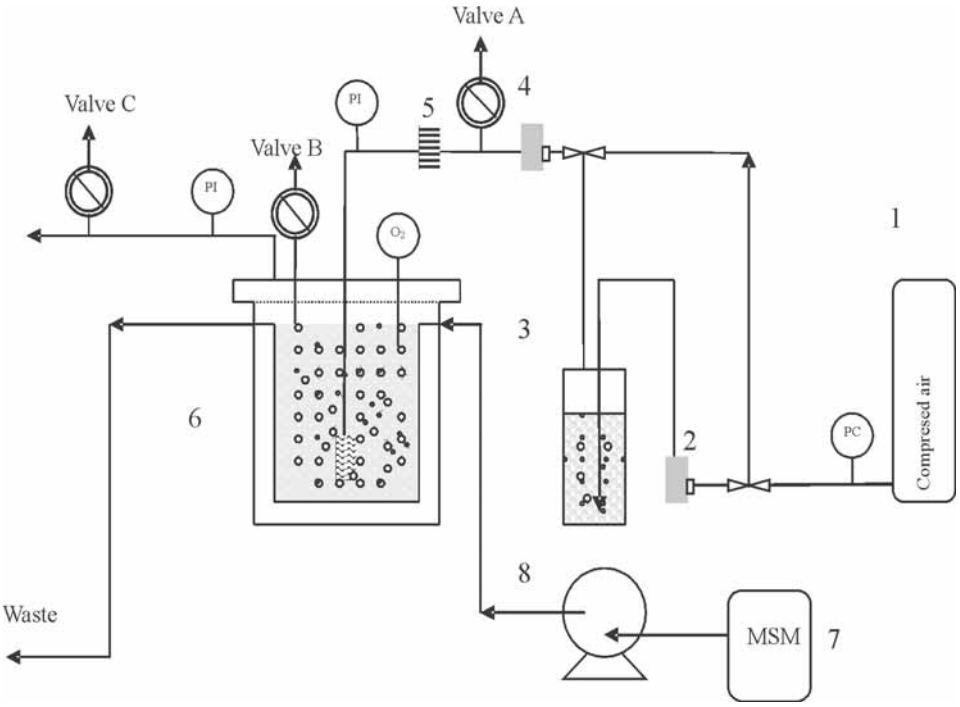


Figure 1. Scheme of the laboratory scale multiphase reactor.

Flame Ionisation Detector (Combustion HFR 400 FFID) calibrated before each experiments with standards. From these measures the amount of styrene absorbed by liquids is known.

2) The system was then completely isolated from the outside air by closing the valves 1 and 5. By closing the flowmeter 4 and opening the valve 2, a finite volume of air (6.7 L) was continuously flowed through the mixture in order to supply the biomass in oxygen (gas flow rate 0.9 $10^{-4}$ $m^3/s$). The decrease in oxygen concentration in air due to the biodegradation of the styrene by microorganisms was monitored and recorded as a function of time during 23h for further analysis. The oxygen fraction in the gas phase was determined using a paramagnetic oxygen analyser (Cosma Cristal 300). It was assumed that: (i) in the reactor the ideal gas law is applicable to calculate the number of moles of oxygen absorbed by the liquids (temperature and pressure of the gas phase was low), (ii) the presence of silicone oil in the emulsion does not change Henry constant for oxygen in water, (iii) the response time for the paramagnetic oxygen analyser is <5 s which is smaller than the mass transfer response time of system.

## 3 RESULTS AND DISCUSSION

Tests of sequential styrene degradation were conducted over a continuous period of 40 days. During these operations input styrene concentrations in the gas phase were adjusted in order to change the styrene load in the mixture which varied from 543 mg to 1800 mg. Several tests were carried out for the same styrene load. Typical examples of oxygen decrease in gas phase versus time are presented in Figure 2 (absorbed styrene around 1150 mg). According to the data shown in Figure 2, it appears that results can be reproduced. Firstly, the oxygen fraction dramatically decreases due to styrene degradation and secondly increases very slowly to reach an equilibrium plateau. This slight increase in probably due to the oxygen balance between the three phases air – silicone oil – water. At the end of each experiment, the oxygen concentration in the water phase was measured and compared with the theoretical value which should verify the oxygen balance between air and water. Experimental and theoretical results are in good agreement. For a styrene load of 1150 mg, the reproducible results presented in Figure 2 indicate that the whole mass of pollutant is degraded during the first 5 hours of the experiments. It can be assumed that this period represents the regeneration time of the solvent by the microorganisms. Figure 3 presents experimental data obtained for different styrene loads. As it can be observed, the time of degradation logically increases with the styrene load and the oxygen fraction decreases by about the same proportion as the styrene load increases. From these data, it can be possible to estimate the elimination capacity of the mixture and the mole number of atomic oxygen used to degrade one mole of styrene (table 1). The obtained elimination capacities are in the same order of magnitude of elimination capacities measured in biofilters: Cox *et al.* (1996), using perlite-packed filters to enrich styrene-degrading fungi, reported styrene elimination capacities of 70 $g.m^{-3}_{\text{filter bed}}.h^{-1}$. Arnold *et al.* (1997), using peat as filter material, reported 30 $g.m^{-3}_{\text{filter bed}}.h^{-1}$. Zilli *et al.* (2001) using a filter packed with a mixture of peat and glass beads (4:1) inoculated with the styrene degrader *Rhocococcus rhodochrous* AL NCIMB 13259, recorded an elimination capacity of 63 $g.m^{-3}_{\text{filter bed}}.h^{-1}$ and Dehghanzadeh *et al.* (2005), using yard waste compost mixed with shredded hard plastics in a 75:25 v/v ratio of plastics inoculated with thickened municipal activated sludge, obtained 45 $g.m^{-3}_{\text{filter bed}}.h^{-1}$. In contrast, Jorio *et al.* (2000) recorded maximum elimination capacities up to 141 $g.m^{-3}.h^{-1}$. In the specific case of biodegradation due to microorganisms in a biphasic aqueous-organic mixture, Osswald *et al.* (1996) measured in a stirred reactor similar elimination capacities (ranged from 23 to 63 $g.m^{-3}.h^{-1}$).

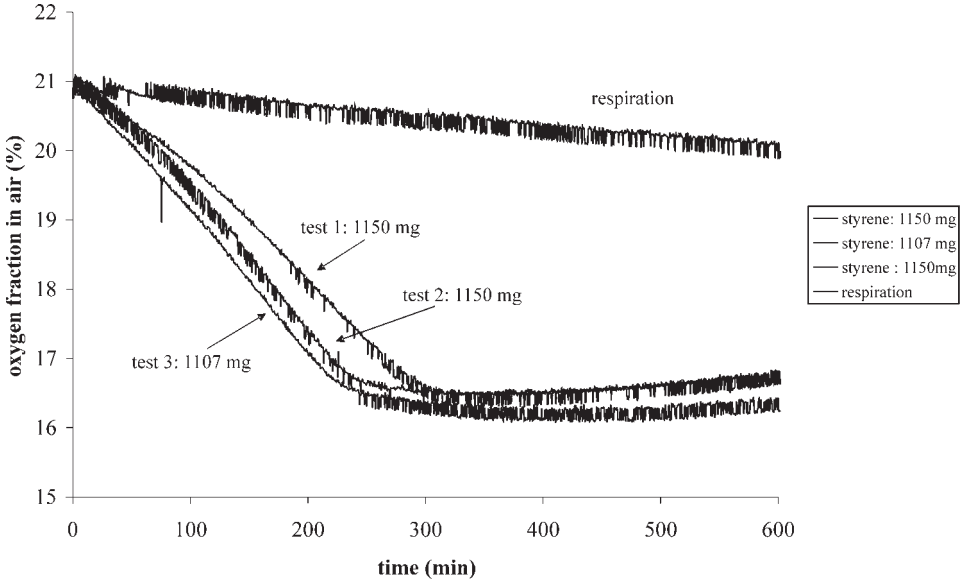


Figure 2. Typical examples of oxygen decrease in the gas phase versus time due to styrene elimination (styrene load: around 1150 mg in 5 L mixture).

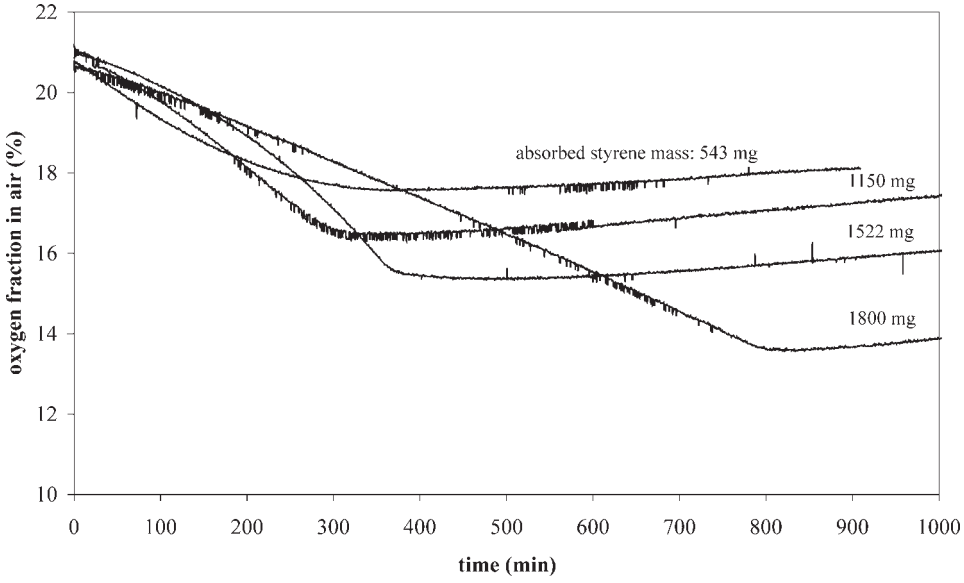


Figure 3. Oxygen fraction decrease in the gas phase versus time for different styrene loads (total liquid mixture: 5 L; silicone oil volume fraction 10%).

On average the biodegradation of one molecule of styrene implies 1.2 moles of atomic oxygen. According to O'Leary *et al.* (2002), two main pathways for the aerobic degradation of styrene exist: an initial oxidation of the vinyl side-chain and a direct attack on the aromatic nucleus (Figure 4). For theses two pathways, the conversion of styrene in either 3-vinylcatechol or phenylacetic acid uses one mole of atomic oxygen. Then it can be assumed that the first steps of the styrene conversion to soluble intermediates in water are sufficient to regenerate the organic solvent for further applications without needing the complete biodegradation in $CO_2$, water and biomass. Obviously, it should be necessary to carry out consecutively a large number of sequential procedures in order to verify that the mixture is not polluted by degradation products, i.e. the solvent regeneration time remains constant for a given styrene load.

Table 1.
Elimination capacity of the biphasic mixture and ratio $O_2$/styrene according to the styrene load.

| **Styrene load in the mixture (g)** | **mole $O_2$ mole styrene removed** | **Elimination capacity ($g_{styrene}.m^{-3}_{mixture}.h^{-1}$)** |
|---|---|---|
| 0.543 | 1.7 | 20 |
| 1.150 | 1.1 | 42 |
| 1.522 | 1.0 | 48 |
| 1.800 | 1.2 | 27 |

## 4 CONCLUSIONS

The following conclusions can be drawn from the preliminary experimental results presented in this study:

(i) reproducible microbial solvent regeneration has been observed. The regeneration time, which increases with the increase of the styrene load, leads to elimination capacity ranged from 20 to 48 $g_{styrene}.m^{-3}_{mixture}.h^{-1}$.
(ii) the solvent regeneration requires roughly 1.2 molecule of oxygen per molecule of styrene which corresponds to the first steps of the biodegradation of the styrene.

Obviously, further experiments will be necessary to complete this preliminary results including the continuous measurement of the pH of the emulsion, the dissolved oxygen in the aqueous phase, the styrene concentration in the gas phase, the identification of the microorganisms present in the mixture and if possible the identification of the intermediary products of the styrene conversion by the microorganisms.

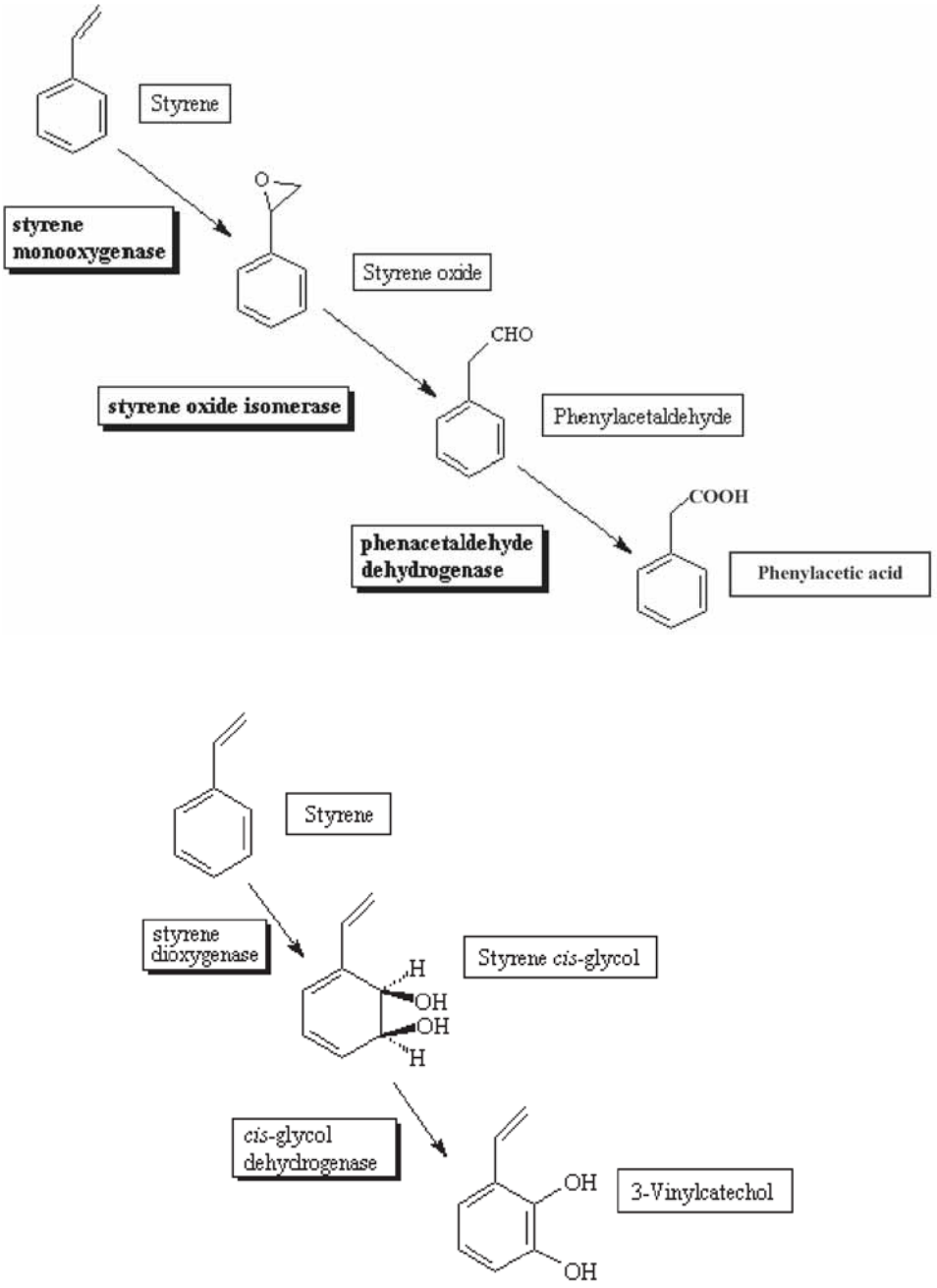


Figure 4. Two major pathways of bacterial styrene degradation: oxidation of the vinyl side-chain and a direct attack on the aromatic nucleus.

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