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# A new method to determine the microbial kinetic parameters in biological air filters

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#### ABSTRACT

This paper presents a new method to determine kinetic parameters of the biodegradation of various pollutants in a biofilter. Toluene, a readily biodegradable volatile organic compound, and methane, a hydrocarbon and a greenhouse gas, have been chosen as the target pollutants. The new protocol utilized biomass immobilized on bed pellets; these directly sampled from a continuous steady-state biofilter. The comparison of this method with the conventional experimental protocol utilizing micro-organisms suspended in a liquid medium was made using the pollutant toluene. Indeed, with both methods, the kinetic parameters have been evaluated by following the microbial growth in batch, thermostated reactors, using determined amounts of pollutant substrate. This experiment has confirmed the pertinence of the new procedure. The interesting features of the new method are that: (1) it is easy to operate (no preliminary treatment of the bed samples) and (2) it provides reproducible parameters that represent the real biofilter case more adequately than liquid cultures. In addition, modeling of the experimental specific growth rates in the case of toluene has shown that the values obtained with the use of solid extracts can be correlated by a Haldane's formulation, where  $\mu^* = 17.6 \, \text{day}^{-1}$ ,  $K_h = 13.2 \, \text{g m}^{-3}$ , and  $K_i = 2.5 \, \text{g m}^{-3}$ . The maximum specific growth rate  $(3.1 \,\mathrm{day}^{-1})$  was reached for an initial concentration of toluene near  $5.0 \,\mathrm{g\,m}^{-3}$ . The determination of the experimental specific growth rates of micro-organisms in the methane biofilter has also been performed. This study allowed highlighting two methane concentrations' ranges: from 1000 to 14500 ppmv and from 14500 to 27000 ppmv. For the first range, the Monod model proves to be suitable with the kinetic parameters:  $\mu_{\text{max}} = 0.43 \,\text{day}^{-1}$  and  $K_m = 5.37 \,\text{g m}^{-3}$ . For the second range, neither the Monod nor the Haldane's formulation could directly be used. However, a mathematical adjustment of the Monod model allows to find kinetic parameters  $\mu'_{\text{max}} = 1.09 \, \text{day}^{-1}$  and  $K'_m = 7.59 \, \text{g m}^{-3}$ . The biomass yields for the tested methane concentrations have also been determined and showed two different tendencies, depending on the same two ranges. For the first range of methane concentrations, the biomass yield was quite constant with an average value around 0.36 g biomass (g methane)<sup>-1</sup> while for the second range, it could be approached by a polynomial second-order regression. The maximum value of the biomass yield obtained on the second range was 0.8 g biomass (g methane)<sup>-1</sup> at a methane initial concentration of 20000 ppmv.

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#### 1. Introduction

Biofiltration of contaminated gaseous effluents is a pollution control technology that became popular in Europe in the 1960s, and that has experienced a significant expansion in North America since 1980 (Kim and Sorial, 2007; Civilini, 2006; van Groenestijn and Kraakman, 2005; Janni et al., 2001). The interesting properties of biological filters are: they usually operate at room temperature and pressure; and the microbial activity (metabolic energy) converts pollutants like toluene (a volatile organic compound) or methane (a greenhouse gas) into mineral products (water, carbon dioxide, etc.) and biomass, which

makes them quite inexpensive processing methods, well adapted for the treatment of low to moderate inlet loads (Leson, 1998). The applications for biofiltration on the industrial scale are now wide (e.g. wastewater treatment plants, food, paint and resins industries, soils decontamination, etc.) (Rappert and Müller, 2005; Kennes and Veiga, 2004). To support further applications, preliminary laboratory and pilot-scale studies are required to optimize the operation parameters (filtering media, flow rates, concentrations, irrigation rate, nutrients, etc.), but also to develop models that describe and predict biofilters' behavior, either at steady-state or under transient conditions. Most of the existing models differ in their assumptions concerning the operating regime (steady-state or transient conditions), or concerning the transfer/equilibriums mechanisms (diffusion, dispersion, adsorption, etc.) between the three phases coexisting in a biofilter. Nevertheless, all of them have to consider the main cause of

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pollutants removal: the microbial activity. Thus, biodegradation kinetic expressions constitute the heart of modeling. Depending on the operating regime and on the model assumptions, either enzymatic or microbial kinetics can be considered. Enzymatic biodegradation, described by Michaelis-Menten's macrokinetics, has been considered by several authors (Gebert et al., 2003; Lim, 2001; Dammann et al., 1999; Wani et al., 1999; Ottengraf and van den Oever, 1983) for the modeling of continuous steady-state biofilters, without biomass evolution in the case of methane or toluene biodegradation. However, most authors associate the rates of pollutants biodegradation with microbial growth rates by considering Monod or Haldane microkinetic approaches for the modeling of biofilters under steady-state or transient conditions (Schwartz et al., 2001; Jorio et al., 2005; Arcangeli and Arvin, 1999; Tang and Hwang, 1997; Baltzis et al., 1997; Shareefdeen and Baltzis, 1994).

The kinetic parameters involved in the model equations can be experimentally determined, and several methods, operating on batch cultures with varying substrate concentrations, have been proposed in the literature. Most of them utilize bacterial strains, either pure or that are isolated from the filtering media, suspended in liquid growth media (Lin and Cheng, 2007; Jorio et al., 2005; Arcangeli and Arvin, 1999; Acuna et al., 1999; Schirmer et al., 1999; Juteau, 1997; Smith and McCarty, 1997; Tang et al., 1997; Oh et al., 1994; Ottengraf and van den Oever, 1983). The drawbacks of these methods are that (1) they necessitate prior operations for the conditioning of the biomass; (2) they do not necessarily represent the real growth media (the solid bed pellets), which more likely contain consortia of interacting micro-organisms, among them the degrading species; and (3) they do not reflect the mass transfer constraints that exist in a biofilter. To date, a few works only have focused on the experimental protocols for application to solid growth media (Govind and Wang, 1997).

In this paper, a new method for the determination of the kinetic parameters is proposed. This protocol utilizes biomass samples extracted from the filter bed and directly inserted into the thermostated batch reactors. Two pollutants are chosen for this study: toluene (C<sub>7</sub>H<sub>8</sub>), as a representative of VOCs and methane (CH<sub>4</sub>) for greenhouse gases. Both the Monod and Haldane kinetic models are considered in respect to describing the experimental data. The values of the specific growth rates so obtained are compared when possible to values obtained by other authors reported in the literature. In addition, the kinetic parameters obtained with toluene after batch experiments with either liquid cultures or solid particles directly sampled are compared.

#### 2. Equipment and procedures

Two kinetic experimental methods have been tested. Both of them are operated in the batch mode, but they require the parallel operation of a continuous laboratory-scale biofilter from which bed samples are extracted and are either directly utilized for the kinetics test, or are pre-treated for the extraction of the micro-organisms.

#### 2.1. Continuous biofiltration setup

The continuous upflow biofiltration system is presented elsewhere for toluene (Delhoménie, 2002) and methane (Nikiema et al., 2005). In the case of toluene, the pellets of the filtering medium were composed of a mixture of compost (90% v/v) and a binder (10% v/v), and had an average particle diameter of around 10 mm, and a density of  $610\,\mathrm{kg}\,\mathrm{m}^{-3}$ . In the methane biofilter, the filter bed was an inorganic material with an average particle size of 5 mm and a density near  $1500\,\mathrm{kg}\,\mathrm{m}^{-3}$  (additional details on the inorganic material are not available to date due to a confidential agreement with the company providing the filter material). During the operation, the two packing beds (one for toluene and the other for methane)

were individually irrigated with 1L of nutrient solution per day. The compositions of the nutrient solutions for the two biofilters are presented in Delhoménie (2002) for toluene and Nikiema et al. (2005) for methane. The total gas flow rate was maintained at 24 and  $6\,\mathrm{m}^3\,\mathrm{day}^{-1}$  and the initial pollutant concentration fixed at 1.9 and  $4.8\,\mathrm{g\,m}^{-3}$ , respectively, for toluene and methane. The kinetic tests were undertaken once the steady-state condition was reached in each biofilter.

### 2.2. Kinetic protocol

The operation of a kinetic test consists in following the evolution of the substrates (C<sub>7</sub>H<sub>8</sub> or CH<sub>4</sub>) and carbon dioxide (CO<sub>2</sub>) mass changes versus time. Kinetic experiments are conducted in batch mode, in thermostated (25 °C) reactors: duplicate reactors (for the reproducibility) and a blank reactor (no substrate addition). The batch reactor is made of pyrex and has a volume of 0.5 L. Biomass extracts from the biofilter bed are poured into the reactor and all tests start as soon as the substrate is introduced into the reactor. Headspace gases are regularly sampled by means of a gastight syringe (100 µL, Hamilton), through a septum, so that the reactors remain leak-proof during the tests. Measurements of the C7H8, CH4 and CO2 concentrations were effected with a gas chromatograph coupled to a mass spectrometer (GC/MS, Hewlett-Packard, 5890 Series II, Column HP5, Supelco, Ontario, and CP-PoraBOND, Varian, Quebec, respectively, for toluene and methane analysis), calibrated with appropriate standards. The toluene, methane and carbon dioxide data collected can be related to the biomass concentrations' change involved in the microbial kinetics. The relationships between the biomass, pollutants and CO<sub>2</sub> depend on the considered method (liquid or solid extracts) and they will be exposed further in this paper. For each pollutant concentration, experiments were conducted at least two times and differences between the specific growth rates values obtained were less than 5% on average for all experiments.

#### 2.3. Preparation of liquid extracts

Before each test, 5 g of bed pellets were sampled from the various sections of the continuous biofilter and were mixed together. The pellets were agitated over 30 min in 50 mL of a solution made from a dilute commercial saline solution, Büshnell-Haas (3.27 g L $^{-1}$ , BH, Difco). The agitation was then followed by low-speed centrifugation (500 rpm, 3 min). The supernatant liquid from the centrifuged preparation provided the solution A. Then, a proteic dosage, providing the biomass concentration of solution A, was made according to the Bradford method, as described by Jorio et al. (2005) and utilized a UV–Visible spectrophotometer. The kinetic experiments were performed with the same initial mass of biomass:  $5\times 10^{-3}\,\rm g$ , so that a determined volume of solution A was poured into each of three batch reactors (duplicates and a blank), and the required 50 mL volume was completed with the BH solution.

#### 2.4. Preparation of solid extracts

The preparation of the solid extracts consisted of sampling a known amount: 5 g of biofilm in the case of toluene and 20 g of bed pellets for methane, extracted from the continuous steady-state biofilter (these amounts were previously optimized, to reduce the time required for the completion of each test). After the sampling, the pellets were left exposed for several minutes in the laboratory atmosphere, so that the substrate contained in the sample was volatilized, and were subsequently introduced into the three reactors (duplicates and blank). The blank reactor test verified that the sampled pellets did not initially contain residual substrate.

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