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Development of particle-based biofilms for degradation of xenobiotic organic compounds

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Abstract

The aim of the experiments performed in this work was to develop a biofilm airlift suspension (BAS) process for the degradation of a mixture of organic sulfonates contained in the infiltration water from a contaminated site. To achieve this goal, active biomass growing on the contaminating xenobiotic organics as the sole source of carbon was obtained by enriching a mixed microbial culture sampled from an activated sludge treatment plant. After kinetic characterisation, the enriched culture was inoculated in the BAS reactor, where it colonised carrier particles and formed stable and uniform biofilms. In spite of the slow growth and degradation kinetics ($\mu_{max} = 0.014 \, h^{-1}$), due to high biomass concentration (up to $12 \, g_{VS} \, L^{-1}$) a high rate process was performed in the BAS reactor, achieving a degradation capacity of $8.7 \, kg_{COD} \, m^{-3} \, d^{-1}$, with an overall degradation efficiency of 70% based on COD measurements.

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1. Introduction

The biological oxidation of scarcely biodegradable compounds in wastewaters is accomplished by microorganisms characterised by slow growth rate, requiring long retention time to be kept in a reactor. To obtain long biomass retention time two conditions must be realised: (i) low rate of biomass wash-out and (ii) high reactor biomass concentration. In activated sludge processes, the biomass retention and, as a consequence,

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the treatment capacity are limited by the biomass concentration achievable in the oxidation tank. This limitation is particularly severe in the case of microorganisms with slow growth rates, resulting in large volumes to accomplish the degradation of target substrates and large areas to be occupied by the treatment works.

To overcome these limitations different types of particle-based biofilm reactors have been developed in the past two decades. In these reactors biomass grows in the form of nearly spherical, compact biofilms, which enable the attainment of large biofilm surface area and high reactor biomass concentration (Nicolella et al., 2000). As a consequence, high volumetric conversion

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rates can be obtained even with slow growing microorganisms, such as nitrifiers in the biofilm airlift suspension reactor (Frijters et al., 2000; Garrido et al., 1997) and methanogens in upflow sludge blanket (Lettinga et al., 1980), expanded bed sludge blanket (Zoutberg and Frankin, 1996) and internal circulation (Habets et al., 1997) reactors.

Aromatic sulfonates are a class of xenobiotic organic compounds which, depending on their molecular structure, may be recalcitrant to biodegradation or present slow biodegradation rates (Cook et al., 1999). These compounds are used as intermediates or obtained as byproducts in chemical and process industries for the production of dispersants, detergents, azo-dyes, concrete plasticizers and wetting agents. Whilst benzene- and mono-substituted naphthalene sulfonates are readily biodegradable (Nörtermann et al., 1986), more complex molecules, in particular those substituted with additional sulfonate, nitro, hydroxy and amino groups, are difficult to degrade (Cook et al., 1999; Nörtermann et al., 1994) and they resist biodegradation in activated sludge processes (Altenbach, 1996; Paxeus, 1996). Recalcitrant aromatic sulfonates are degraded by advanced treatments, such as chemical oxidation by Fenton's reagents (MacKay and Pignatello, 2001) or ozone (Shiyun et al., 2002), and electrochemical oxidation (Panizza et al., 2000).

This work illustrates the experimental procedure required to develop a biofilm airlift suspension (BAS) process for the degradation of a mixture of aromatic sulfonates contaminating an infiltration water, including fundamental investigations on microbial culture enrichment, growth and degradation kinetics and biofilm development. BAS reactors have already been successfully applied for the degradation of single aromatic sulfonates in synthetic wastewater (Wagner and Hempel, 1988), but there are no reports in the open literature on the application of particle-based biofilm reactors for the treatment of real wastewaters containing mixtures of these compounds.

2. Materials and methods

2.1. Leachate characterisation

The experiments were performed on the infiltration water collected at an industrial site in the north-west of Italy. The infiltration water (called 'leachate' in the following) is contaminated by the wastes of a chemical factory. Leachate samples pumped from the drainage wells at the factory site were collected once a week, transported to the laboratory and cooled at 4 °C. The composition of the leachate varied considerably during the period of experiments. In particular, the organic content ranges from 300 to $1200 \,\mathrm{mg_{COD} L^{-1}}$, with

nitrogen and phosphorous concentrations lower than $10 \text{ mg}_{N-NH_4} L^{-1}$ and $0.4 \text{ mg}_{P-PO_4} L^{-1}$, respectively. HPLC assays performed by the analytical laboratory at the factory site show that the organic content of the leachate is mainly due to aromatic sulfonates, including benzene sulfonates (1-amino benzene-4-sulfonic acid: $7-10 \text{ mg L}^{-1}$; 1-amino benzene-3-sulfonic acid: $15-22 \text{ mg L}^{-1}$), naphthalene sulfonates (naphthalene-1sulfonic sodium salt: $32-47 \text{ mg L}^{-1}$; naphthalene-2sulfonic sodium salt: $0-3 \text{ mg L}^{-1}$), disulfonates (1,6-NDS: $63-86 \text{ mg L}^{-1}$; 1,5-NDS: $3-9 \text{ mg L}^{-1}$; 2,6-NDS: $3-10 \text{ mg L}^{-1}$; 2,7-NDS: 19-31 mg L⁻¹; 2-OH-1,6-NDS: $0-1 \text{ mg L}^{-1}$; 2-OH-6,8-NDS: 13-29 mg L⁻¹; 2-OH-3,6-NDS: $3-8 \text{ mg L}^{-1}$; 2-amino naphthalene-5,7-disulfonic acid: $1-3 \text{ mg L}^{-1}$) and trisulfonates (2-OH-3,6,8-NTS: $2-10 \,\mathrm{mg}\,\mathrm{L}^{-1}$).

2.2. Inoculum and nutrient medium

The inoculum for the experiments performed in this work was a mixed culture of biomass obtained from the activated sludge process operating at the factory where the leachate originates. Biomass and activated carbon (added to the mixed liquor in the treatment plant at the factory site to reduce the toxicity of the leachate) from the aeration tank were separated by mechanical stirring and subsequent classification by sedimentation in Imhoff cones.

Since the leachate does not contain enough macronutrients, ammonium (NH₄Cl) and phosphate (KH₂PO₄) salts were added in order to reach a C:N:P ratio of 100:5:1. Variable amounts of salts were dosed depending on the measured COD concentration in the leachate and assuming an average COD/TOC ratio of 3.3 as determined by the analytical laboratory at the factory site. Micro-nutrients (e.g. 0.1 mg L^{-1} of MgSO₄ · 7H₂O and 0.5 ml L^{-1} of trace element solution formulated according to Vishniac and Santer, 1957) were also added to the leachate.

2.3. Biomass enrichment

Before inoculation in the BAS reactor, a period of enrichment was adopted to increase biomass activity and degradation efficiency. Biomass enrichment was performed in 2 L flask experiments. The leachate was initially diluted with nutrient medium to obtain a COD of around 200 mg L⁻¹. The dilution was then progressively reduced until the leachate was directly used as the growth medium. The volume of nutrient medium required to obtain a desired initial COD concentration was determined on the basis of the measured COD of the leachate (varying in the range 720–1100 mg L⁻¹ in the period of enrichment).

After inoculation with around 200 mg L^{-1} of biomass, the flasks were stirred, continuously sparged with air

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