

# Influence of operational conditions on biofilm specific activity of an anaerobic fluidized bed reactor

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**Abstract** A key parameter in water and wastewater treatment technology is the biomass activity in terms of substrate removal ability. The effects of organic load rate and percentage of bed expansion on biofilm specific methanogenic activity were determined in an anaerobic fluidized bed reactor treating wine-distillery wastes in the thermophilic range (55°C). The proposed activity tests are highly reproducible: an experiment with three identical tests has shown that the standard deviation with respect to the mean values is less than 3%. Specific tests are applied to measure the maximum methanogenic activities of the biomass carrier in lab-scale anaerobic biofilm reactors. These tests have been successfully applied for monitoring the support colonization process and the evolution of biofilm activity in reactors, anaerobic filter and fluidized bed, with different operating conditions. The results show a dependence between the percentage of bed expansion and the specific activity of methanogenic microbiote on biofilm. There is a relationship between the percentage of bed expansion, the shear stress on the biofilm and the hydrodynamic conditions in the system. Initial biofilm detachment can be compensated with the increase of biomass and of its activity due to the reduction of the substrate diffusional limitations to the microorganism growth inside the support pores.

**Keywords** Biofilm; methanogenic activity determination; thermophilic anaerobic bacteria

## Introduction

High-rate anaerobic digesters, which also retain biomass, have a high capacity to treat numerous organic industrial wastewater and hence low site-area requirements. The major biofilm processes developed are based on two general configurations, flocculation of anaerobic bacteria consortium (UASB reactors) or bacterial attachment to different media support (stationary packed beds, both upflow and downflow, and fluidized- or expanded bed reactors) (Pérez *et al.*, 2001).

A key parameter in water and wastewater treatment technology is the biomass activity in terms of substrate removal ability. However, this parameter is not always linearly correlated with the conventional biofilm descriptors as dry weight, COD or biofilm thickness (Lazarova and Manem, 1995). For a better prediction and control of the biofilm process efficiency, this activity descriptor should be used.

The aim of this paper is to quantify the effect of the organic load rate (OLR) and the percentage of bed expansion (E) on the methanogenic specific activity (AM) in an anaerobic fluidized bed reactor treating wine-distillery wastes in the thermophilic range (55°C).

## Materials and methods

### Anaerobic fluidized bed

A transparent Plexiglass column with a cross-section of 5.11 cm<sup>2</sup> and length of 170 cm was used. Its bottom was moulded into a conical shape to promote uniform fluidization of media and colonized particles (bioparticles). The reactor temperature was maintained at 55 ± 1°C by circulating water through the water jacket and the biogas generated was collected in a gas meter. The effluent from the fluidized bed reactor was recycled through a variable speed membrane pump in order to provide upflow velocities for media and to maintain the

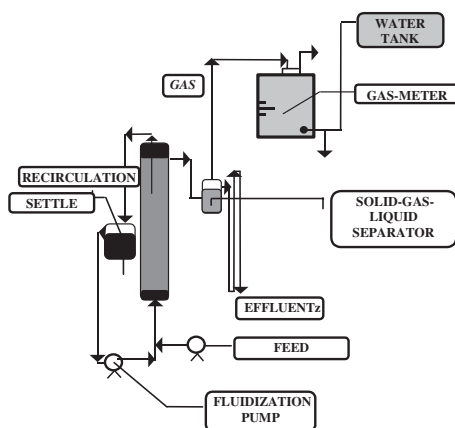
different expansions of the bioparticle bed (percentage of bed expansion, E), as shown in Figure 1. The feed utilised was wine-distillery wastewater (Perez *et al.*, 2001). The feed was supplied by a continuous feeding methodology with diluted and neutralised vinasses with 15 gCOD/L (Garcia-Morales *et al.*, 2000).

The experimental conditions, organic load rate (OLR), hydraulic retention time ( $\theta$ ), percentage of bed expansion (E) (relationship between the active reactor volume operated like a fixed bed (300 cm<sup>3</sup>, active volume) and the volume of the expanded bed) and the average of COD removal efficiency (COD<sub>r</sub>, %), are presented in Table 1.

The media support used consisted of open-pore sintered-glass beads (SIRAN™) developed and marketed by Schott Glaswerke. The SIRAN™ bead carrier is produced by sintering a mixture of glass and saltpowder following by washing process which elutes the non-sinterable salt. The resulting glass sponge has a well-defined pore size distribution. This material was chosen because of its characteristics (surface area: 87 m<sup>2</sup>/L, pore volume: 55–60%) and also because it could be calcined to measure attached biomass concentration like total volatile solids.

### Activity determination

The specific methanogenic activity determination was carried out by using an activity test. The activity test procedure involves incubating the colonized supports from the anaerobic fluidized bed reactor with a specific substrate (acetate) in sealed anaerobic vials. Vials of 125 cm<sup>3</sup> with rubber stoppers and crimp seals are used. The quantity of colonized biomass is measured by using a calibrated cylinder. The non-attached biomass present in the support was removed by washing the support with water tempered to 55°C. The biomass concentration (X<sub>0</sub>) was measured by analysing the volatile suspended solids concentration



**Figure 1** Scheme of the experimental fluidized bed reactor

**Table 1** Operational conditions of the anaerobic fluidized bed reactor

OLR gCOD/L/d	$\theta$ day	E %	COD <sub>r</sub> <sup>(*)</sup> %
13.3 ± 1.6	1.2	0	98
		25	
		100	
29.9 ± 2.5	0.5	25	93
		50	
		100	

(\*) average of COD removal efficiency (%)

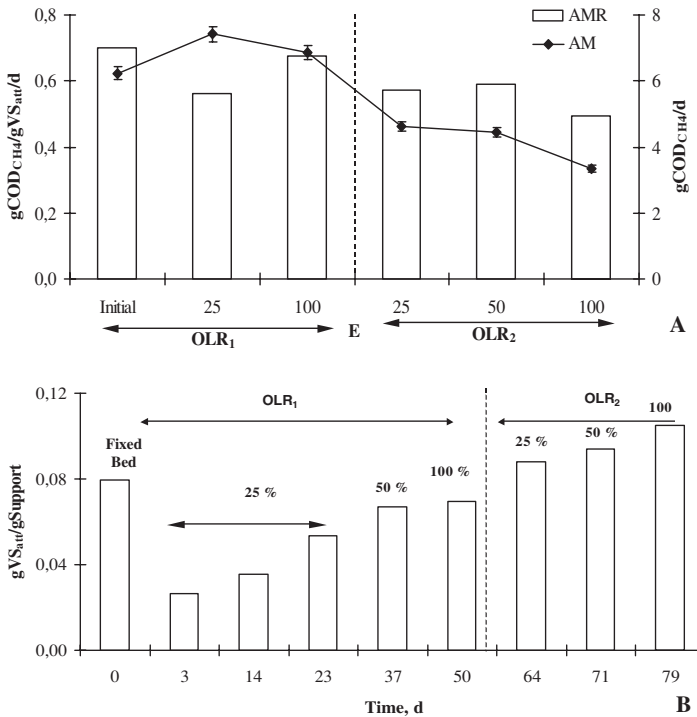
per gram of support (gVSS/g support). A selected medium containing the principal macro- and micro-nutrients (García-Morales *et al.*, 1996) was used. The colonized support (in a range between 0.5–1 for AM, expressed in gVSS/L) was added to this solution and then the vial was sealed and flushed for 5 min. with N<sub>2</sub>. In the methanogenic test, the gas-meter was connected at this moment. The reactor was immersed in a thermostatic shaking bath (Heto lab equipment, SB-22-A, 51 rpm (30% of maximum value); adjustable stroke length: 40 mm) for 1 hour to 55°C. After this time, the specific substrate (acetate with a concentration between 2–3 g/L) was added to each assay. Measurements began at this moment.

The determination of methanogenic specific activity (AM) was carried out by measuring the methane produced from acetate as the methanogenic substrate (Soto *et al.*, 1993). This parameter is the relationship between the maximum methane generation rate and the concentration of attached biomass in gVSS/L. Tests carried out under experimental conditions are highly reproducible. An experiment with three identical tests has shown that the standard deviation with respect to the mean values is less than 3%. The described test has been successfully applied for monitoring the support colonization process and the evolution of biofilm activity on reactors, anaerobic filter and fluidized bed, with different operational conditions (García-Morales, 1997).

**Results and discussion**

The evolution of methanogenic specific activity (AM, gCOD<sub>CH4</sub>/gVS<sub>att</sub>/d), specific activity extended to the whole active reactor, 300 cm<sup>3</sup>, (AMR, gCOD<sub>CH4</sub>/d) and attached biomass with the percentage of bed expansion (E) and the OLR are shown in Figure 2.

The relationship between the percentage of bed expansion, the increase in upflow velocities, the shear stress and the decrease in the diffusional limitations on the biofilm is well



**Figure 2** Evolution of several variables with the OLR and the percentage of bed expansion (E): A. methanogenic specific activity (AM ± 3%) and extended to AMR, and B. attached biomass (gVS<sub>att</sub>/g support)

known. The massive initial biofilm detachment when the operation of the fluidized bed starts (Figure 2B) can be compensated with the increase of biomass growth inside the support pores due to the reduction of the substrate diffusional limitations.

Figure 2A shows a different behaviour for each of the methanogenic activities, AM and AMR. On the first OLR (13.3 gCOD/L/d), methanogenic activity (AM) was observed to decrease due to a growth of biofilm in the internal zones of the particle that reduces the specific activity determined by activity test. For this OLR, due to this net biomass growth, there is an increase on the methanogenic activity extended to the whole active reactor (AMR). The growth of attached biomass can be observed on Figure 2B. Furthermore, there is not much substrate availability for a greater growth of the biomass due to the fact that the removal efficiency in this period is superior to 98%. On the other hand, on the second OLR (29.9 gCOD/L/d), there is a great increase of biomass on the support as a consequence of the greater substrate availability (Table 1). However, the specific AM determined by the test shows a decrease, from 25 to 100% of bed expansion, as in the first OLR.

The results of Figure 2B show a continuous increase of attached biomass in contrast with the decrease of the AM observed by using the activity test. This fact can be due to a difference between the hydrodynamic conditions in the fluidized bed reactor and in the activity test procedure, that could imply an underestimation of the real biomass activity inside the reactor in relation to that registered on the fixed bed operation.

## Conclusions

There is a high influence of upflow velocities, percentage of bed expansion (E), in the decrease of the diffusional limitations in those anaerobe strict microbiotes, like the methanogenic population. This phenomenon implies a growth of the biomass and an increase of the specific activity extended to the whole active reactor (AMR).

In the second OLR there is a substantial increase of biomass in the support due to the greater substrate availability. Also, the percentage of bed expansion permits an increase of biomass in the support due to the decrease of diffusional limitations. The difference between the hydrodynamic conditions in the reactor and in the activity test could imply an underestimation of the real biomass activity inside the fluidized bed reactor.

The support, under anaerobic thermophilic conditions, due to its properties of low density and specific area, is suitable for the immobilisation of slow-growing microorganisms (e.g. anaerobic thermophilic organisms) and for use as carrier on fluidized bed reactors.

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