The effect of operational conditions on the sludge specific methanogenic activity and sludge biodegradability


ABSTRACT

The effects of hydraulic retention time (HRT) and influent COD concentration (COD_{inf}) on Specific Methanogenic Activity (SMA) and the biodegradability of an anaerobic sludge need to be elucidated because of the discordant results available in literature. This information is important for the operation of anaerobic reactors and design of the sludge post-treatment unit. For this study, sludge samples obtained from eight pilot-scale Upflow Anaerobic Sludge Blanket (UASB) reactors were tested. The reactors were fed with municipal wastewater and operated with different sets of HRT and influent concentrations until the steady state was established. The results show that at a lower HRT, sludge with relatively higher SMA develops. A slight trend of declining SMA at increasing COD_{inf} was found for reactors operated at longer HRTs; however, further experiments are necessary for more definitive conclusions. The sludge from reactors operated at longer HRTs and with lower COD_{inf} resulted in lower biodegradability. Results also showed that it is ineffective to design a UASB reactor with a longer HRT to cope with organic shock loads.

Key words | biodegradability, HRT, influent concentration, SMA, UASB reactors

INTRODUCTION

The metabolic capacity or specific methanogenic activity (SMA) of anaerobic sludge depends on various operational parameters, including hydraulic retention time (HRT), upflow velocity ($V_{up}$), organic loading rate (OLR), influent chemical oxygen demand (COD_{inf}), sludge retention time (SRT), operational temperature, presence of inhibiting factors or xenobiotics compounds, and reactor configuration (Lettinga 1995). However, published results about the effect of HRT, $V_{up}$, and COD_{inf} on SMA are contradictory. Jawed & Tare (1996) investigated the performance of an Upflow Anaerobic Sludge Blanket (UASB) reactor fed with diluted molasses and concluded that increasing the OLR by reducing the HRT led to a decrease in SMA. The same observation was made by Kalyuzhnyi et al. (1996).

They operated a lab-scale UASB reactor fed with a mixture of glucose and acetate under different operational conditions and found that decreasing the HRT from 6.3 to 4 h caused a decrease in the SMA. On the other hand, O’Flaherty et al. (1997) used upflow hybrid reactors fed with a solution of volatile fatty acid (VFA) and alcohol to assess HRT and $V_{up}$ effects on the SMA. They concluded that by decreasing the HRT from 8 to 4 h and raising the $V_{up}$ from 0.01 to 0.5 m/h resulted in a 200% increase in the SMA. According to Kato et al. (1997), in reactors treating diluted wastewater containing ethanol, the low substrate concentration was the main reason for the relatively low SMA of the sludge. On the other hand, Jawed & Tare (1996) investigated the performance of a UASB reactor fed with diluted molasses...
and found that an increase in COD_{Inf} led to a decreasing SMA. Ghangrekar et al. (1996) operated lab-scale UASB reactors fed with sewage under different HRTs, V_{up}, and COD_{Inf}. They found that the sludge SMA did not follow any trend with respect to these parameters. With respect to the discordant information available in the literature, it is clear that the effects of operational conditions on the SMA still need to be clarified. The knowledge about the effect of these parameters on the SMA can provide insight about the capacity of the UASB reactors to withstand organic and hydraulic shock loads.

Sludge biodegradability is a parameter used to estimate the fraction of the organic material in the sludge that can be biologically converted into methane. The effect of the operational conditions imposed on UASB reactors regarding the biodegradability of the sludge has been scarcely reported in literature. As mentioned by Mgana (2003), it is very difficult to compare the data obtained by the few researchers who carried out studies on this area, as each of them followed different experimental procedures.

The objective of this study is to evaluate the effect of COD_{Inf}, HRT, SRT, and V_{up} on both the SMA and biodegradability of sludge samples withdrawn from different UASB reactors.

### MATERIALS AND METHODS

#### Experimental set-up

The experimental investigation was carried out using eight pilot-scale UASB reactors which had a volume of 120 L each, a height of 4m, and were fed with domestic sewage at 27°C. The pilot-scale reactors were denominated by R^{HRT}_{COD}, where the superscript index stands for the hydraulic retention time, and the subscript index stands for the total influent COD. Both are the averages during steady state conditions. When the steady state had been established, the sludge samples were withdrawn from taps located at four different heights on the reactors to assemble composite samples, which were immediately used for the analysis of SMA, biodegradability and solids. The main operational parameters are presented in Table 1.

#### Specific methanogenic activity tests

The SMA was determined following the procedure described by Jawed & Tare (1999). The tests were carried out in 0.6 L serum bottles (with a working volume of 0.4 L), which were filled with sludge (5 g/L), sodium acetate (2.5 g/L), distilled water, and pH buffer (2.5 g/L of sodium bicarbonate). Both nutrients and trace elements were added

### Table 1 | Operational parameters

<table>
<thead>
<tr>
<th>Reactor</th>
<th>HRT (h)</th>
<th>V_{up} (m/h)</th>
<th>COD_{Inf} (mg/L)</th>
<th>COD_{Dis} (mg/L)</th>
<th>COD_{Inf} (mg/L)</th>
<th>COD_{Dis} (mg/L)</th>
<th>VFA_{Inf} (mgAc./L)</th>
<th>OLR (kgCOD/m^{3}.d)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Set 1–UASB reactors operated with similar COD_{Inf}, but different HRT and OLR</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>R3_{16}</td>
<td>6</td>
<td>0.64</td>
<td>816 ± 45</td>
<td>566</td>
<td>250</td>
<td>180</td>
<td>3.3</td>
<td></td>
</tr>
<tr>
<td>R4_{70}</td>
<td>4</td>
<td>0.95</td>
<td>770 ± 38</td>
<td>459</td>
<td>312</td>
<td>164</td>
<td>4.6</td>
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</tr>
<tr>
<td>R7_{87}</td>
<td>2</td>
<td>1.90</td>
<td>787 ± 31</td>
<td>513</td>
<td>275</td>
<td>164</td>
<td>9.4</td>
<td></td>
</tr>
<tr>
<td>Set 2–UASB reactors operated with the same HRT, but different COD_{Inf} and OLR</td>
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<td></td>
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<tr>
<td>R3_{55}</td>
<td>6</td>
<td>0.64</td>
<td>816 ± 45</td>
<td>566</td>
<td>250</td>
<td>180</td>
<td>3.3</td>
<td></td>
</tr>
<tr>
<td>R3_{355}</td>
<td>6</td>
<td>0.64</td>
<td>555 ± 36</td>
<td>421</td>
<td>135</td>
<td>88</td>
<td>2.2</td>
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<tr>
<td>R3_{58}</td>
<td>6</td>
<td>0.64</td>
<td>298 ± 19</td>
<td>216</td>
<td>82</td>
<td>42</td>
<td>1.2</td>
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<tr>
<td>R1_{95}</td>
<td>6</td>
<td>0.64</td>
<td>195 ± 15</td>
<td>75</td>
<td>12</td>
<td>28</td>
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</tr>
<tr>
<td>Set 3–UASB reactors operated with similar OLR, but different HRT and COD_{Inf}</td>
<td></td>
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</tr>
<tr>
<td>R3_{16}</td>
<td>6</td>
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<td>816 ± 45</td>
<td>566</td>
<td>250</td>
<td>180</td>
<td>3.3</td>
<td></td>
</tr>
<tr>
<td>R3_{58}</td>
<td>4</td>
<td>0.95</td>
<td>558 ± 31</td>
<td>383</td>
<td>175</td>
<td>103</td>
<td>4.2</td>
<td></td>
</tr>
<tr>
<td>R3_{532}</td>
<td>2</td>
<td>1.90</td>
<td>352 ± 18</td>
<td>235</td>
<td>117</td>
<td>62</td>
<td>3.4</td>
<td></td>
</tr>
</tbody>
</table>

COD_{Inf} and COD_{Dis} refer respectively to the suspended solids (SS) and dissolved fraction of the total influent COD; VFA_{Inf} refers to the influent volatile fatty acids; values are Confidence Intervals (α = 0.05); Data of reactor R3_{16} is repeated to create the three sets above.
to the bottles to prevent deficiency during the test. The following nutrients (g/L) were added: NH₄Cl (0.28), K₂HPO₄ (0.25), MgSO₄·7H₂O (0.10), CaCl₂·2H₂O (0.01), and CaCO₃ (0.60). For trace elements (1 mL/L) the following substances (mg/L) were added: FeCl₂·4H₂O (2000), H₃BO₃ (50), ZnCl₂ (50), CuCl₂·2H₂O (58), MnCl₂·4H₂O (500), (NH₄)₆Mo₇O₂₄·4H₂O (50), AlCl₃·6H₂O (90), and CoCl₂·6H₂O (2000). The activity tests were performed at 30°C and in shaking conditions of approximately 100 rpm. The total volatile solids (VS) content of the sludge was determined prior to the SMA test to calculate the specific activity. Methane production was monitored daily during the test by displacement using a Mariotti bottle filled with NaOH solution (5%w/w). All experiments were performed in triplicate.

**Sludge biodegradability tests**

The sludge biodegradability tests were conducted using serum bottles similar to those used for the SMA tests. Additionally, the same temperature and shaking conditions were applied. 400 mL of sludge was added to the bottle, but no nutrients, substrate, or buffers were added. The VS content of the sludge was determined prior to the biodegradability test to calculate the fraction converted to methane. All experiments lasted 30 days. The biodegradability was calculated with Equation 1 (Leitão 2004).

\[
\text{Bio} (\%) = \left( \frac{\text{COD}_{\text{CH}_4}^{30}}{\text{COD}_{X}^{X}} \right) \times 100
\]

Where: “Bio” is the Biodegradability of the sludge; COD₃₀CH₄ is the total amount of methane produced at the end of the test in terms of COD and calculated based on the Henry’s law; CODₓₓ is the initial mass of sludge added to the serum bottles and calculated based on the VS content of the sludge when assuming that 1 gVS is equivalent to 1.5 gCOD (van Haandel & Lettinga 1994).

All physical-chemical analyses were performed as recommended by the Standard Methods (1995).

**RESULTS AND DISCUSSION**

**Specific methanogenic activity**

SMA was higher for sludge collected from reactors which were operated at shorter HRTs and higher V_up (Set 1 in Figure 1). This phenomenon can be likely attributed to selective retention of sludge with higher SMA in reactors that were operated at higher upflow velocities (O’Flaherty et al. 1997). This mechanism appears to be confirmed by the results obtained with reactor R₂ (V_up of 1.90 m/h). At the end of the experiment, this reactor contained a certain amount of granular sludge with the highest SMA (0.59 gCOD/gVS.d). Higher SMA values that are found when reactors are operated at shorter HRTs can also be attributed to a high concentration of biomass which is grown under high volatile fatty acids (VFA) loading rates (VLR). The substrate for methanogenic microorganisms was partially independent of the anaerobic food chain, i.e., VFA is generally present in raw sewage due to some hydrolysis and acidification that occurs in the sewer system (approximately 180 mg/L as acetic acid). Moreover, VFA is also produced in the reactor during the acidogenic step. This introduced and/or produced VFA

![Figure 1](image_url)
may have improved the growth of this specific trophic group. Thus, an increased SMA at decreased HRT can be interpreted as a larger relative mass of methanogenic archaea (Guiot et al. 1992).

According to Zeeman & Lettinga (1999), the longer the SRT, the better the sludge quality in terms of methanogenic activity. However, this only seems to be true if the sludge age is increased by a decreased influent SS concentration (Set 2 in Figure 1). Even so, this rise does not seem considerable compared to Set 1. During the treatment of domestic sewage, a short hydraulic retention time leads to short SRT and a high SMA (Set 1 in Figure 1).

The effect of the COD_{Inf} concentration on the SMA is not clear. It would appear that the influent concentration, ranging from 200–800 mgCOD/L, did not affect the SMA to any measurable extent and that the differences in sludge SMA found for those reactors which were operated with the same HRT are in the error range for such tests.

**Sludge biodegradability**

The biodegradability of sludge has an inverse relationship with the HRT, i.e., the shorter the HRT, the higher the biodegradability of the sludge (Figure 2). This is most likely because reactors operated at short HRTs are also inherently submitted to high OLR, and in the case of sewage, also generally submitted to a high SS loading rate. This high amount of entrapped SS reduces the SRT and therefore increases the biodegradability of the sludge. Another possible reason for higher sludge biodegradability when reactors are operated at shorter HRT is that the reactors are also exposed to a high VLR. The highly biodegradable material is then attributed to the high concentration of methanogenic biomass which was grown under the high VLR, as previously explained.

The results in Figure 3 show that the first hypothesis may be coherent with regards to the effect of SRT on the sludge biodegradability. Although the SS removal efficiency decreased for reactors operated at shorter HRTs, the SS loading rate (based on the removed SS) was higher. The second hypothesis is also consistent. Assuming that the SMA is proportional to the methanogenic bacteria content of the sludge, then for the same sludge biodegradability the differences in SMA can only be attributed to differences in the sludge’s contents of biodegradable SS.

This can be confirmed by applying the method described by Mgana (2003) for evaluating the degradable SS fraction in the sludge (X_{deg}). This method requires a linearisation of the cumulative methane curve produced during the biodegradability test (Equation 2). Only the linear part of the curve is taken into account for the linearisation, and the choice of the time when the straight line starts is based on the best fit for a linear regression, with Coefficient of Determination (R^2) closer to 1.

\[
\ln \left( \frac{X_0}{X_t} \right) = K_d \times t + a
\]

Where: X_0 is the total degraded SS (X_{deg} + X_{bm}) in the biodegradability test; X_t is the remaining degradable SS at any time during the experiment; “t” is the duration of the biodegradability test, and “a” is an empirical constant (0.087 and 0.415 for reactors R_{816} and R_{787} respectively).

The result of the method is presented in Figure 4 using reactors R_{816} and R_{787} as examples. If the stabilisation of the sludge had occurred according to single first order kinetics, then the biodegradability would probably be due to the decay of biomass (X_{bm}). This is because methane production, after a prolonged test period, originates from bacterial death and subsequent decay of dead cells (Seghezzo et al. 2002), as well as from the decay of poorly biodegradable matter. The results revealed that the initial parts of the curves in Figure 4 do not fit into the linearised methane production. This discrepancy can be interpreted as the contribution of
hydrolysed $X_{deg}$ and the decay of the biomass. Clearly, the discrepancy in Figure 4B exceeds that found in Figure 4A, indicating that the degradable SS concentration increased as the HTR decreased. In fact, during the first 10 days, the sludge of reactor R6 produced only 27% of the total recovered methane, whereas sludge of reactor R2 released an amount of 57% of methane. After this initial period, the gas production rate was very low, almost constant, and continued along the whole test time frame.

The values for the decay constant ($K_d$) used in Equation 2 (see straight lines in the plots) are within the range (0.004 to 0.050 $d^{-1}$) provided by the literature review of Batstone et al. (2002). The $K_d$ of sludge produced in reactors R6 and R2 were 0.030 and 0.043 $d^{-1}$ respectively. The latter indicates that a significant part of the sludge’s biodegradability for reactor R2 is due to degradable SS and that most of the sludge biodegradability for reactor R6 is due to biomass decay.

The results presented in Figures 2 and 3 show that the reactors operated with low COD$_{Inf}$ produced sludge with a lower biodegradability than reactors operated with high COD$_{Inf}$. This is because, according to Cavalcanti (2003), for a given HRT, the low total influent COD, and therewith low SS concentration, leads to a long SRT (Figure 3) and therefore the low sludge biodegradability.

### Maximum methanogenic potential

Maximum Methanogenic Potential (MMP) is the maximum capacity of a reactor to convert an imposed VFA Loading Rate (VLR) into methane under optimal conditions (Equation 3), as stated by Leitão (2004). The VLR was calculated based on the influent VFA concentration and the VFA produced by acidogenesis in the reactor (Equation 4). VFA produced during the acidogenic step was calculated using Equations 5 and 6, according to Mahmoud et al. (2004). The difference between the MMP and VLR gives the “extra” VFA loading capacity. This “extra” capacity is considered to be a “reserve” capacity for the reactor to convert the VFA (introduced and/or produced) during a possible VFA overload. The results presented in Table 2 show that the “extra” VFA loading capacity significantly increases in systems operated at lower HRTs. This indicates that it is futile to design a...
UASB reactor with a long HRT to increase its capacity to cope with shock loads.

\[ \text{MMP} = \frac{(\text{SMA} \times M_X)}{24} \]  

\[ \text{VLR} = \left( \frac{\text{COD}^{\text{Inf}}_{\text{VFA}} + \text{COD}^{\text{Acid}}_{\text{VFA}}}{\text{COD}^{\text{Inf}}_{\text{VFA}}} \right) \times Q \]  

\[ \text{COD}^{\text{Acid}}_{\text{VFA}} = A_{\text{Tot}} \times \text{COD}^{\text{Inf}}_{\text{VFA}} \]  

\[ A_{\text{Tot}} = \frac{\left( \text{COD}^{\text{CH4}}_{\text{VFA}} + \text{COD}^{\text{Effl}}_{\text{VFA}} - \text{COD}^{\text{Inf}}_{\text{VFA}} \right)}{\text{COD}^{\text{Inf}}_{\text{Tot}}} \]  

Where: MMP (gCOD/h), SMA (gCOD/gVS.d), VLR (gCOD/h); \( M_X \) = Mass of VS in the reactors (gVS); \( \text{COD}^{\text{Inf}}_{\text{VFA}} \) = Influent VFA as COD (gCOD/L); \( \text{COD}^{\text{Acid}}_{\text{VFA}} \) = VFA produced during the acidogenic step (gCOD/L); \( Q \) = Flow rate (L/h); \( A_{\text{Tot}} \) = Acidogenesis based on total influent COD (gCOD/L); \( \text{COD}^{\text{CH4}}_{\text{VFA}} \) = Methane gas production in terms of COD (gCOD/L); \( \text{COD}^{\text{Effl}}_{\text{VFA}} \) and \( \text{COD}^{\text{Inf}}_{\text{VFA}} \) = Total VFA in the influent and effluent, respectively, as COD (gCOD/L).

### CONCLUSIONS

- Lower HRT leads to a higher sludge SMA. The influent concentration ranging from 200 to 800 mgCOD/L (fraction of SS in the range of 65–75%) has little effect on the SMA.
- Reactors operated with a long HRT and with a low \( \text{COD}^{\text{Inf}}_{\text{VFA}} \) produce sludge with low biodegradability.
- The high biodegradability of sludge produced in reactors operated at a low HRT is due to the high amount of entrapped degradable SS and the high concentration of biomass.
- There is no merit in designing a UASB reactor with long HRT to increase its capacity to cope with shock loads.

### ACKNOWLEDGEMENTS

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### REFERENCES


### Table 2

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Reactors</th>
</tr>
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<tbody>
<tr>
<td>Sludge mass—( M_X ) (gVS)</td>
<td>R1, R6</td>
</tr>
<tr>
<td>SMA (gCOD/gVS.d)</td>
<td>R1, R6</td>
</tr>
<tr>
<td>Max Meth. Potent.—MMP (gCOD/h)</td>
<td>R1, R6</td>
</tr>
<tr>
<td>VLR during steady state (gCOD/h)</td>
<td>R1, R6</td>
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<tr>
<td>Extra VFA loading capacity (gCOD/h)</td>
<td>R1, R6</td>
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With data from Table 2.


