Specific Methanogenic Activity of anaerobic sludge from different wastewater treatment systems

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Anaerobic technology in wastewater treatment is widespread in Brazil, being benefited by climate conditions, besides presenting advantages such as low cost, low solids production, and tolerance to high organic loads. In anaerobic systems occurs the process known as anaerobic digestion, which is the degradation of complex organic matter by microorganisms in the absence of oxygen, producing the biofuel known as biogas, composed mainly of methane (CH_4), carbon dioxide (CO_2), water and hydrogen sulfide (H_2S). However, the monitoring of an adapted biomass with high microbial activity in anaerobic reactors is essential for ensuring the anaerobic process efficiency. Several methods have been proposed to evaluate the anaerobic microbial activity, based on the characterization of Specific Methanogenic Activity (SMA). SMA is defined as the maximum CH_4 production capacity by an anaerobic microbial community (or anaerobic sludge), where the conversion of organic substrates into CH_4 occurs in a controlled manner in laboratory, such as substrate addition and temperature control, in order to favor the metabolism of methanogenic archaea. We aimed to analyze SMA from different anaerobic samples to select the biomass with higher methanogenic capacity for subsequent inoculation in a pilot-scale UASB reactor, in addition to allowing the monitoring of the anaerobic treatment operational performance of different wastewater systems.

This study assessed the SMA of three biomasses from different anaerobic systems: i) a septic tank sludge from a kennel; ii) a prototype UASB reactor sludge for swine wastewater treatment; and iii) a real scale UASB reactor sludge from a wastewater treatment plant (WWTP) treating university sewage. The experiment was performed to evaluate the maximum capacity of the three sludges, under anaerobic conditions and in laboratory-scale bioreactors, in converting organic substrates to CH_4. After the sampling in each system, the concentration of total volatile solids (TVS) was measured according to APHA (2012), to obtain the volume of sludge to be added to 100 mL flasks, using a substrate/microorganism ratio of 0.5 gCOD/gTVS. The experiment was performed in triplicate. The flasks were sealed with rubber bung and aluminum sealing, maintaining anaerobic atmosphere after the expulsion of oxygen by N_2 and incubated at 30±2 ºC. Sodium acetate was used as a substrate in combination with macro and micronutrient solutions. The volume of CH_4 daily produced was directly measured through CH_4 volume measurement (Aquino *et al*, 2007) by washing biogas with NaOH 15%. After 30 consecutive days, accumulated CH_4 volume data was processed in programming language *R 3.6.3* for SMA determination. For each anaerobic sludge, a sigmoidal model was adjusted to the observed data (Figure 1). Logistic and Gompertz models were used for comparison and all model coefficients were significant at 0.001 probability. SMA values obtained in mL_{CH4} were converted to $gCOD_{CH4}$ using stoichiometric conversion factor (F = 394 $mL_{CH4}.(gCOD_{CH4})^{-1}$, considering temperature of 30 °C and pressure of 1 atm).

Table 1 presents SMA values obtained for each anaerobic sludge sample, in addition to the TVS values and TVS/TS ratio (Total Solids). The SMA of the prototype UASB sludge sample was the highest (0.045 $gCOD_{CH4}.(gTVS\ day)^{-1}$) among the analyzed sludge samples; however, the inflection point of the curve (Figure 1b) occurred previously (4th day) if compared with that of WWTP UASB sludge. This demonstrates that prototype UASB biomass had faster adaptation (or lag phase) to the environment. Likewise, it presented a shorter exponential phase (or log phase), occurring in approximately 6 days (from the 3rd to the 9th day). This is the phase that occurs the largest methanogenesis activity, allowing to obtain the maximum CH_4 production rate through the higher curve slope. On the other hand, the lag and log phases were longer for WWTP UASB biomass (Figure 1c), which were respectively up to the 8th and 25th days, with a SMA of 0.034 $gCOD_{CH4}.(gTVS\ day)^{-1}$ and an inflection point occurring on the 17th day. Thus, the phase of higher CH_4 yield occurred in a wider time interval for this sample. These facts explain the higher CH_4 accumulated volume during the test period by the WWTP UASB sludge (100.1 mL_{CH4}) compared to that of the prototype UASB sludge sample (57.7 mL_{CH4}). Furthermore, the curve was better adapted to the data of the WWTP UASB sludge (R^2 = 0.9989) in relation to those of the prototype UASB sludge (R^2 = 0.9777). SMA values obtained for these two sludge samples from different-scales UASB reactors were according to some results found in the literature. Alves *et al* (2005) recorded, for swine wastewater biodigester sludge, a SMA of 0.0331 $gCOD_{CH4}.(gTVS\ day)^{-1}$, using the same method used in this study, whereas Almeida *et al* (2011) reported, for UASB reactor sludge treating dairy wastewater, a SMA of 0.040 $gCOD_{CH4}.(gTVS\ day)^{-1}$. In a previous study carried out by Paez (2019), the value of 0.055 $gCOD_{CH4}.(gTVS\ day)^{-1}$ was obtained for the same WWTP UASB sludge used in this study. For domestic sewage, the usual values are in the range of 0.1 to 0.4

$gCOD_{CH4}.(gTVS\ day)^{-1}$ (Chernicharo, 2007). Regarding the sludge sample from the kennel septic tank, despite the similarity of its curve shape (Figure 1a) with that of the WWTP UASB sludge, it presented a SMA of 0.007 $gCOD_{CH4}.(gTVS\ day)^{-1}$ and a CH_4 accumulated yield of 9.1 mL_{CH4}, low values comparing with those achieved by the other two sludge samples tested and those found in the literature.

WWTP UASB sludge presented higher CH_4 accumulated yield in a longer log phase period, despite not presenting the highest SMA among the samples. Therefore, we concluded that it is the most suitable sludge to be used as inoculum for the pilot-scale UASB reactor, which will possibly allow higher substrate (wastewater) degradation efficiency and biogas yield.

Table 1. SMA of the anaerobic sludge samples and their respective values of TVS concentration, TVS/TS ratio, and CH_4 accumulated yield.

Sludge sample	TVS ($g\ L^{-1}$)	TVS/TS ratio	CH_4 accumulated yield (mL)	SMA ($gCOD_{CH4}.(gTVS\ day)^{-1}$)
Kennel tank septic	47.3	0.45	9.1	0.007
Prototype UASB	30.8	0.42	57.7	0.045
WWTP UASB	49.7	0.36	100.1	0.034

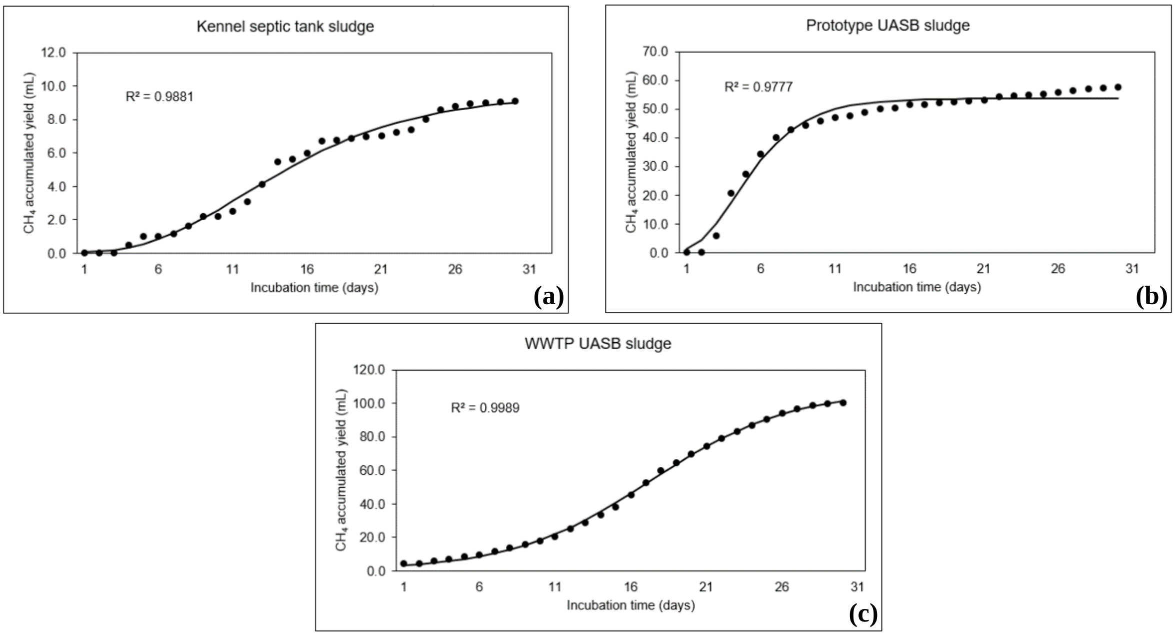

Figure 1. Sigmoidal curves adjusted to CH_4 accumulated daily yield data over 30 days of SMA testing, for the following sludge samples and respective adjustment models: (a) kennel tank septic and (b) prototype UASB, both adjusted by the Gompertz model; and (c) WWTP UASB, by the Logistic model.

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