Alkaline and acid hydrolytic processes in aerobic and anaerobic sludges: effect on total EPS and fractions

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Abstract Sludge samples from an upflow anaerobic sludge blanket (UASB) reactor and four submerged aerated biofilters (BFs) of a wastewater treatment plant (1,000 inhab.) were processed at bench scale by alkaline and acid hydrolysis with the objective to evaluate the organic matter solubilization, volatile solids (VS) destruction and the effect of hydrolytic processes on the extracellular polymeric substances (EPS) fraction of the sludge samples. The results showed that alkaline hydrolysis of sludge samples treatment with 1.0% total solids (TS) using NaOH 20 meq L^{-1} was more efficient on organic matter solubilization and VS destruction than acid hydrolysis. The EPS sludge content was also affected by the alkaline treatment of anaerobic sludge samples. The EPS concentrations (mg EPS/gVSS) on the anaerobic sludge after the alkaline treatment were significantly lowered according to sample height in the UASB reactor. Data indicated that the EPS sludge fraction is the main component affected by the alkaline hydrolytic process of anaerobic sludge samples.

Keywords EPS; hydrolysis; sludge; UASB; wastewater treatment

Introduction

The sludge management in wastewater treatment plants (WWTP) has been one of major environmental challenges in Brazil (von Sperling and Gonçalves, 2001; Gonçalves *et al.*, 2003). The first stage on the sludge management is the volume minimization at WWTP, that can be achieved by selecting wastewater treatment process with low sludge production (example: the association in series of anaerobic and aerobic processes).

The main benefits derived from combined aerobic and anaerobic reactors for wastewater treatment are well documented (Mergaert *et al.*, 1992; Van Haandel and Lettinga, 1994). Compactness, energy saving and low sludge production are some advantages of this association, when compared to conventional treatment facilities. The association of UASB reactors with fixed biofilm reactors is a subject of research, involving mainly percolating biologic filters in the aerobic treatment step (Kitao *et al.*, 1986; Collivignarelli *et al.*, 1990). Biofilters (BFs) constitute an excellent option for the post-treatment of UASB reactor effluents, due to their capacity to degrade the soluble compounds and filter the suspended particulates, in the same reactor. The UASB reactor promotes a primary treatment of domestic sewage and the aerobic biofilter promotes a secondary and tertiary treatment of UASB effluent. The sludge generated in the aerobic biofilter is backwashed to a UASB reactor where it is thickened and processed concomitantly with the raw sewage effluent to a UASB reactor.

The chemical hydrolysis under acid and alkaline conditions is one of management strategies for sludge minimization, characterized by the possibility to increase the soluble organic fraction of the sludge which can be converted into biogas by UASB reactor, i.e. converting the solid phase to gaseous phase in the WWTP. According to Muller (2001) the hydrolytic technologies can increase the soluble organic fraction of the sludge but can

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also be applied with the objective to improve the efficiency of biological processes such as phosphorus and nitrogen removal, improving the sludge dewatering process and scum inhibition. Some authors recommend for sludge minimization the combination of the hydrolytic technologies such as chemical and thermal or chemical and mechanical (sonication) among others (Chiu *et al.*, 1997; Weemaes and Verstraete, 1998).

The main question derived from the sludge hydrolysis treatment is the actual contribution of extracellular polymeric substances (EPS) fractions to soluble organic matter and available for methane conversion in the UASB reactor. EPS are metabolic polymeric products secreted by bacterial cells that tend to accumulate outside the cell, forming a gel-like matrix (Morgan *et al.*, 1990). They form a protective shield layer against environmental stresses, and also act as carbon and energy reserves. EPS were found crucial for sludge properties such as flocculation, dewatering and main constituents for both aerobic and anaerobic sludges (Nielsen *et al.*, 1996; Schmidt and Ahring, 1996). EPS are composed of a variety of organic substances. Carbohydrate was identified as the predominant constituent in the EPS fraction from sludge derived from domestic sewage of many pure cultures (Frølund *et al.*, 1996; Sutherland and Kennedy, 1996; Cescutti *et al.*, 1999; Shin *et al.*, 2001), whereas protein was found in substantial quantities in the sludges of many wastewater treatment reactors (Fang and Jia, 1996). Humic substance (Frølund *et al.*, 1995), uronic acid and deoxyribonucleic acids (DNA) (Zhang *et al*., 1999; Tsuneda *et al.*, 2001) were also detected in EPS; however, information about their concentration in EPS is scarce. These complex substances have great interference in sludge characteristics and basics for flocs, granule and biofilm generation. In domestic wastewater treatment plants, the EPS fraction are part of total volatile solids (VS), generally used to estimate the active biomass and important parameter for biological wastewater treatment projects.

Material and methods

Wastewater treatment plant, Universidade Federal do Espirito Santo (WWTP-UFES)

The sludge samples in this work were obtained from WWTP processing domestic sewage of 1,000 inhabitants consisting of anaerobic (UASB) reactor followed by post-treatment with aerated submerged biofilters (BFs) with main operational parameters listed in Table 1. The discharged sludge from aerobic BF is sent to the UASB reactor for thickening and digestion.

Sludge sampling and conditioning

Anaerobic sludge derived from UASB reactor was collected at tap 1 and tap 3 (0.25 and 1.25 m respectively) and characterized for pH, VS, total solids (TS) and chemical oxygen demand (COD) according to APHA (1995). Aerobic sludge was collected during the backwashing of biofilters by six consecutive discharges intercalated with 3 min forced aeration. The samples were concentrated by gross filtration and diluted to the TS (%)

Table 1 Main operational characteristics of WWTP-UFES

Parameter	UASB + BF
Population (inh)	1,000
Q (average) (L/s)	1.0
Area (m^2)	5.3
Height (utile)(m)	5.3
Volume (m^3)	26.0
HRT (hour)	8.0

concentration of 1.0, 3.0 and 5.0%. After this physical conditioning process, the samples were kept at room temperature for 24 hours before the hydrolysis assays.

Sludge hydrolysis

Sludge samples were processed in a 2.0 L reactor by continuous rotation (150 ± 2 rpm) in a jar apparatus at room temperature (25 ± 3°C). Acid and alkaline hydrolysis were performed with concentrations of 1, 3, and 5.0% TS and increased concentrations of 0, 20, 40, 80 and 100 meq/L of acid (H_2SO_4) and sodium hydroxide (NaOH) respectively. The hydrolytic assays were performed with 8 hours with 1 hour interval for the following monitored parameters: pH, VS, TS, COD total, COD filtered, according to APHA (1995). The volatile solids destruction was also monitored using sludge samples of 1% TS and 20 meq/L acid or sodium hydroxide, centrifuged at 3,500 g for 20 min and TS and VS assessed in settled sediment and supernatant (not filtered) fractions after 6 hours processing. The reaction kinetics was also investigated by processing the sludge samples with 5% TS, 20 meq/L in acid (H_2SO_4) and alkaline (NaOH) conditions during 8 hours, with monitoring interval of 1 hour and following sludge parameters of pH, TS, VS, COD total and COD filtered.

EPS extraction and quantification

Anaerobic sludge samples collected at 0.25 and 1.25 m of UASB reactor at WWTP-UFES were submitted to alkaline and acid hydrolysis with 20 meq/L of NaOH and H_2SO_4 for acid and alkaline sludge hydrolysis respectively during 6.0 hours reaction. The EPS extraction of initial and final sludge processing was performed according to Judice (1991) by using NaOH as the extractor agent. The EPS fractions analyzed were: proteins by microbiuret method, carbohydrates by the phenol-sulphuric acid (Dubois *et al.*, 1956) and lipids by the sulfophospho-vanillin method.

Results and discussion

Sludge characterization and hydrolysis

The main sludge characteristics are listed in Table 2, showing that the anaerobic sludge from UASB reactor bed (0.25 cm) has a TS concentration of 4.0% (±0.68%) as compared with the aerobic biofilter sludge with approximately 1.0% ± 0.68% TS and moreover presenting a higher relationship VS/TS = 77.07 ± 1.19%. Also, it can be inferred that organic matter in both sludges is formed by insoluble material as seen by low COD filtered/COD total relationship.

The hydrolytic assays were performed as a function of total solids (TS%) concentration of 1, 3 and 5% and acid or alkaline concentration (0 to 100 meq/L) during

Table 2 Main characteristics of aerobic and anaerobic sludge samples used in present work. SD = standard deviation; n = number of samplings

Parameter	Aerobic sludge (biofilter)			Anaerobic sludge bed (UASB)		
	Average	SD	n	Average	SD	n
pH	6.86	0.20	24	6.84	0.23	18
TS(mg/L)	986.24	197.79	23	40,813.3	6,750.5	18
TS(%)	0.99	0.02	23	4.08	0.68	18
VS (mg/L)	6,784.60	176.26	23	31,449.7	5,139.7	18
VS/TS(%)	68.14	7.30	23	77.07	1.19	18
COD total (mg/L)	1,100.20	335.80	24	41,620.8	11,871	18
COD filtered (mg/L)	76.92	18.45	24	260.01	80.52	18
CODfilt/DQOtotal (%)	7.54	2.53	24	0.65	0.17	18

the fixed time of 8 hours. The results were expressed as COD filtered/COD total, to emphasize the solubilization due to hydrolytic processes shown in Figure 1 for aerobic and anaerobic sludge. Comparing the hydrolytic processes, it can be seen that alkaline hydrolysis was more efficient to increase the soluble fraction for both aerobic and anaerobic sludge. No statistical differences were observed for acid treatment although a slight tendency to increase the relationship at higher TS% was observed.

After the definition of the optimal TS% values for hydrolytic processes, by using a referential and fixed value of 40 meq/L of hydrolytic agent, the efficiency of the hydrolytic process was evaluated as a function of the acid and soda concentration adopting a fixed concentration of 1% TS for both sludges. Comparing the hydrolytic treatment after 8 hours and expressed with the relationship COD filt/COD total, it was shown that higher optimal values of COD filt/COD total were achieved in the alkaline hydrolysis using 60 meq/L and 20 meq/L of NaOH for aerobic and anaerobic sludge respectively, meaning that the alkaline hydrolytic process was more efficient for anaerobic sludge as compared with results observed for aerobic sludge. The acid treatment did not show any statistical differences among doses and showed low hydrolytic efficiency for both sludges with maximum of organic matter solubilization (COD filter/COD total) of 11.0%.

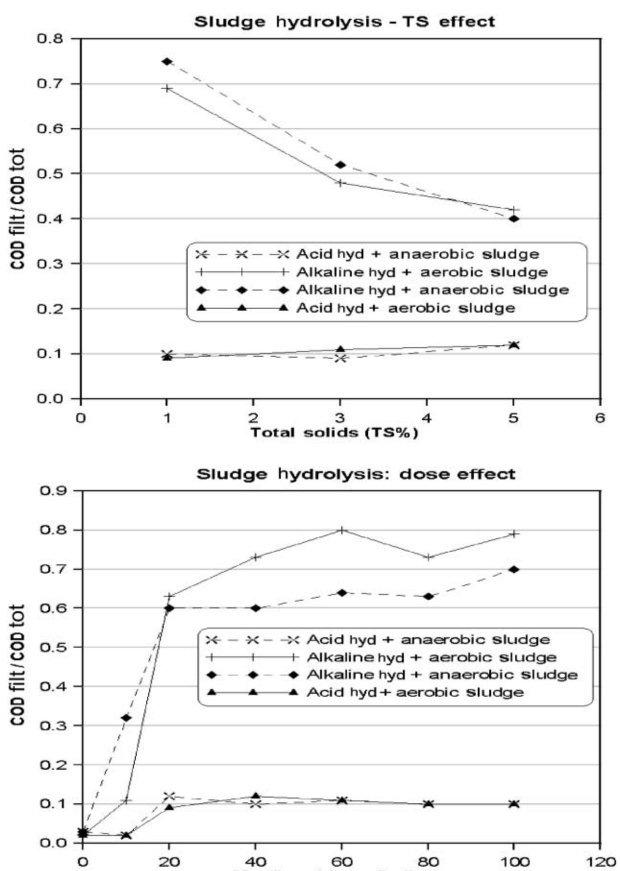

Figure 1 Effect of sludge solubilization expressed as COD filt/COD total as a function of TS% and fixed 40 meq/L of hydrolytic agent and effect of hydrolytic agent concentration of 0–100 meq/L with fixed 1.0% TS for aerobic and anaerobic sludge after 8 hours of hydrolytic processing

The time-course reaction hydrolysis of aerobic and anaerobic sludge is expressed in Figure 2. It was shown that alkaline hydrolysis fitted a "first hour saturated" model for both sludges and the acid hydrolysis fitted a "linear" model for both sludges. In fact, the alkaline hydrolysis can solubilize almost 60% of organic matter sludge in the first hour reaction. There is no apparent reasonable explanation for these trends which will have to be further investigated.

Solids destruction

Results of VS in settled sediment of aerobic and anaerobic sludge after six hours of hydrolytic treatment are shown in Table 3. The alkaline hydrolysis processing was more efficient in VS removal fraction than acid treatment, as seen by the % removal of 12–19.8% for alkaline process of aerobic and anaerobic sludge and 6.0–10.4% removal derived from acid treatment of the same aerobic and anaerobic sludge. These values may represent the susceptibility of active biomass or EPS material to alkaline conditions as compared with the acid conditioning process. One possible explanation is based on the natural acidic buffering capacity of both sludges as they carry an excess negative charge of organic matter material from the matrix.

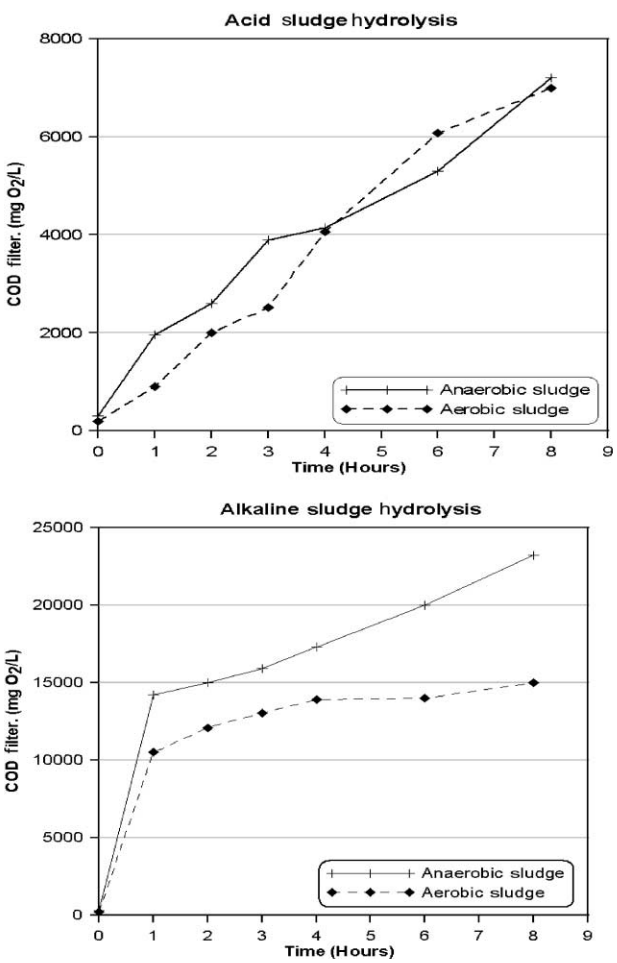

Figure 2 Hydrolysis data from aerobic and anaerobic sludge hydrolysis with 5% TS and 20 meq/L in acid (H_2SO_4) and alkaline (NaOH) conditions during 8 hours, expressed as COD filtered (mg O_2/L)

Table 3 Solid destruction expressed as % of volatile solids removal for aerobic and anaerobic sludge under alkaline and acid hydrolysis after 6 hours of processing using fixed TS of 1% and 20 meq/L of hydrolytic agent (H_2SO_4 or NaOH). SD = standard deviation

Sludge	Time (h)	Alkaline hydrolysis			Acid hydrolysis		
		VS (g/L)		% removal	VS (g/L)		% removal
		Average	SD		Average	SD	
Aerobic	0	9.10	0.537	19.8	9.52	0.275	10.4
	6	7.30	0.164		8.53	0.791	
Anaerobic	0	6.90	0.050	12.0	7.26	0.238	6.0
	6	6.07	0.141		6.85	0.310	

Effect of hydrolysis on EPS content

Results from anaerobic sludge samples submitted to alkaline and acid hydrolysis evaluated for total and EPS fractions at two UASB reactor heights are shown in Figure 3. The total EPS content expressed as mg EPS/g VS in the settled sediment of sludge samples was influenced by hydrolytic treatment and sample height at UASB reactor. The alkaline hydrolysis was more efficient to remove the EPS content of anaerobic sludge samples as compared with the acid treatment. Sampling height at UASB reactor (tap 1 and tap 3) also influenced the total EPS and fractions of sludge samples. Generally, samples from higher layer or blanket in the UASB reactor were more susceptible to EPS reduction by hydrolytic attack, probably due to lower TS% and increased active biomass products expressed as VS content. Comparing the EPS fractions, the same trend was observed, i.e. significant reduction due to alkaline treatment and more significant reduction as a function of reactor height.

The overall data derived from the main effects of hydrolytic processes and anaerobic sludge sample height in UASB reactor on total and EPS fractions are consolidated in Table 4 as a tentative scheme to visualize the main trends correlated with the discussed processes.

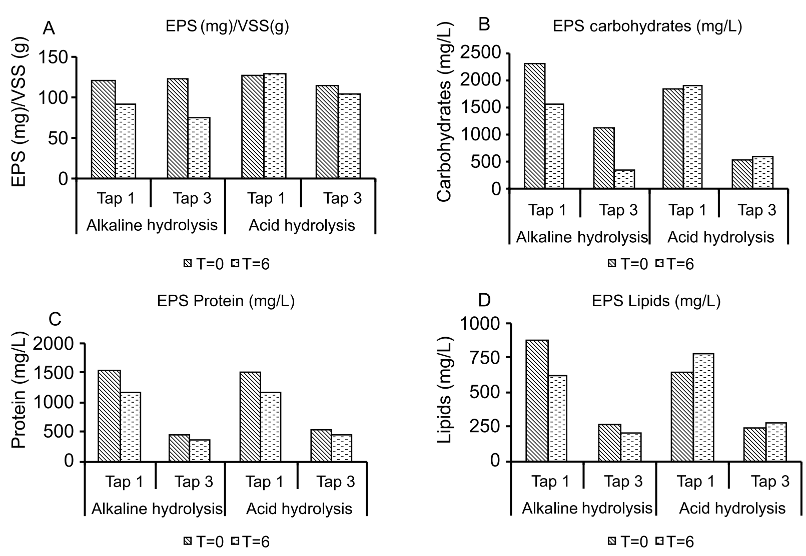

Figure 3 Total EPS (A) and main EPS fractions identified as carbohydrates (B), proteins (C) and lipids (D) from anaerobic sludge samples at tap 1 (0.25 m) and tap 3 (1.25 m) from UASB reactor of WWTP-UFES submitted to alkaline and acid hydrolysis. n = 6; T = hours

Table 4 Consolidated data from total and EPS fractions from this study, considering the sludge samples from different UASB layers and alkaline and acid hydrolysis processes after 6 hours treatment. Increasing (↑), decreasing (↓) and no effect (↔)

Parameter/fraction	Tap 1 (0.25 m)		Tap 3 (1.25 m)	
	Acid	Alkaline	Acid	Alkaline
Total EPS	↔	↓	↓	↓
EPS carbohydrates	↔	↓	↔	↓
EPS protein	↓	↓	↓	↓
EPS lipids	↔	↓	↔	↓

Cost analysis

The anaerobic sludge hydrolysis process using 20 meq/L of NaOH in a 4% TS anaerobic sludge with a VS/TS value of 77% represents an alkali consumption of 0.8 kg/ton. Considering the soda price of US$110.00 per ton we can estimate the cost of sludge alkaline processing of US$ 0.08 per ton. Combining the discharging raw sludge managing costs of US$ 29.00 per ton and the VS destruction value of 12% (Table 3) for anaerobic sludge under alkaline processing, we can infer that sludge managing costs can be reduced to around US$ 25–26 which means a US$ 3.00–4.00 reduction cost per ton, assuming that all hydrolyzed VS can be converted to gas phase (biogas) by the UASB reactor receiving that hydrolyzed sludge.

Conclusions

- The hydrolytic processes can be applied to sludge derived from aerobic and anaerobic wastewater treatment. The alkaline hydrolysis using NaOH at 20–40 meq/L, at room temperature, is a more efficient process as compared with acid hydrolysis processed with the same sludge samples.
- The main effect of alkaline hydrolysis is to solubilize the organic matter content of aerobic and anaerobic sludge as expressed by the relationship of COD filtered/COD total. The effect can be seen on the solids destruction expressed as VS representing the active biomass and biomass products in sludge matrix.
- The alkaline hydrolysis can solubilize almost 60% of organic matter from anaerobic and aerobic sludges in the first hour reaction hydrolysis, fitting a saturated model as opposed to acid hydrolysis which tends to fit a “linear model” for sludge hydrolysis.
- The main target of hydrolytic attack is the total extra polymeric substances EPS and its fractions represented by carbohydrates, proteins and lipids.
- The solubilized fractions of VS derived from biomass of EPS fractions can be assimilated and metabolized inside UASB reactors, minimizing the sludge generation and increasing methanization in wastewater treatment plants.

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