

# A FULL-SCALE UASB REACTOR FOR TREATMENT OF PIG AND CATTLE SLAUGHTERHOUSE WASTEWATER WITH A HIGH OIL AND GREASE CONTENT

L. A. S. Miranda<sup>1</sup>, J. A. P. Henriques<sup>1,3\*</sup> and L. O. Monteggia<sup>2</sup>

<sup>1</sup>Biotechnology Center, Department of Biophysics, Federal University of Rio Grande do Sul, UFRGS, Fax +(55) (51) 3316-7309, Av. Bento Gonçalves 9500, Prédio 43421, Campus do Vale, Cx. P. 15005, CEP: 91501-970, Porto Alegre - RS, Brazil.

E-mail: pegas@cbiot.ufrgs.br, E-mail: luismiranda@pop.com.br

<sup>2</sup>Institute of Hydraulic Research, Federal University of Rio Grande do Sul, Bento Gonçalves 9500, CEP 91501-970 - Porto Alegre - RS, Brazil.

E-mail: monteggia@iph.ufrgs.br

<sup>3</sup>Biotechnology Institute, University of Caxias do Sul - UCS, Caxias do Sul - RS, Brazil.

(Received: July 31, 2003 ; Accepted: May 9, 2005)

**Abstract** - This paper discusses the performance of an 800m<sup>3</sup> full-scale UASB reactor in treating meat-packing plant and slaughterhouse effluents containing high concentrations of oil and grease (O&G) (413-645 mg/L), resulting in a COD/O&G ratio of 26-32%. Those macromolecules were considered responsible for the unbalance of the system resulting in a total washout of the biomass. The removal of O&G from the influent using a physicochemical system (coagulation-flocculation) improved the physical characteristics of the anaerobic sludge, controlling the biomass washout. Reactor performance was significantly improved when the COD/O&G ratio influent was maintained in the 10%. The COD and O&G removal rates obtained after implantation of the physicochemical system were 70-92% and 27-58%, respectively. The specific methanogenic activity (SMA) of the biomass shows towards a tendency stabilisation and adaptation to the substrate influent. Pretreatment of the influent allowed the maximum organic load to be increased (1.46 to 2.43 Kg COD/m<sup>3</sup>.d) and improved the quality of the effluent.

**Keywords:** Anaerobic digestion; Oil and grease; Specific methanogenic activity; UASB reactor.

## INTRODUCTION

The technology of anaerobic digestion has become well established in the treatment of industrial and urban effluents because of factors such as its low implementation and maintenance costs, its excellent organic matter removal rate and the production of methane. A better understanding of the microbiological and hydraulic mechanisms that regulate the system has contributed to the development of more compact and modern high-rate

reactors, such as the upflow anaerobic sludge blanket (UASB), which combines operational simplicity and efficiency. Effluents generated by agro-industry, especially by meat packing, have high levels of proteins, lignocellulose compounds and fats as soluble matter and suspended solids and require preliminary treatment before they can be biologically treated (Martinez et al., 1995; Oliva et al., 1995; Sayed et al., 1987, 1988a, b). It has been reported that anaerobic treatment of slaughterhouse wastewater at mesophilic temperatures is slowed

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\*To whom correspondence should be addressed

down or impaired because of high concentrations of suspended solids (SS), particularly fat particles (Masse et al., 2001). The application of a pretreatment to hydrolyse SS, especially fats, may improve the anaerobic digestion of unsettled slaughterhouse wastewater (Rajan et al., 1989).

Masse et al. (2001) observed that 35% of the neutral fats in slaughterhouse effluents hydrolysed to free LCFA when treated with pancreatic lipase 250 (PL-250) enzyme, increasing the rate of removal in an anaerobic reactor operating in batch regime. The pretreatment had a significant effect on reduction of the size of fat particles, but only a small effect on their digestibility, which was only 4% higher than that of the control substrate after a 69-hour digestion. The authors concluded that this type of pretreatment can be more efficient in reactors that operate at high temperatures, since the rate of LCFA removal can be increased (Masse et al., 2001a).

The stability of these systems depends on a series of biochemical reactions during the anaerobic digestion of organic matter, whose main characteristic is an important syntrophic interaction in the initial steps of the treatment – hydrolysis, acidification and biogas production (Monteggia, 1991; Silveira et al., 1999; Schink, 2001). However, fats, oil and grease, especially long-chain fatty acids (LCFAs), cause operational problems when continuously discharged at high concentrations, inhibiting several bacteria involved in the process (Koster & Cramer, 1987; Sayed et al., 1988; Tay & Yan, 1996; Thiele et al., 1988), thus requiring a pretreatment phase.

Hanaki et al. (1981) conducted an extensive study of the inhibitory effect of LCFA on anaerobic digestion. They concluded that LCFAs affect obligate hydrogen-producing acetogenic bacteria, which are responsible for the  $\beta$ -oxidation of LCFAs, as well as hydrogenotrophic and acetotrophic methanogenic archaea, which convert the intermediates resulting from the  $\beta$ -oxidation process. They also found that LCFA disappear from the solution and accumulate in the solid phase within 24 hours. This is in accordance with theories that relate LCFA inhibition to physical interactions between the acids and the microbial membranes. The main result is a difficulty in anaerobic biomass granulation and granule flotation (Menju et al., 1997) and impaired syntrophic interaction between microbial groups (Tay & Yan, 1996).

Effluents generated by agro-industry, especially by meat packing, contain a significant amount of dissolved protein in the form of gelatin and natural carbohydrates, such as polysaccharides. Cellulose and starch are the most abundant polysaccharides in slaughterhouse and meat-packing wastewater and

most are insoluble in water, forming colloidal suspensions (Sanders, 2001). A considerable amount of the organic matter in slaughterhouse effluents is composed of lipids found alone or mixed with carbohydrates and proteins (Lalman & Bagley, 2002; Schink, 2002), and glycerol and myristic (C14:0), palmitic (C16:0), stearic (C18:0), oleic (C18:1) and linoleic (C18:2) acids constitute approximately 90% (Sanders, 2001). Removal of lignocellulose compounds and fat is necessary prior to application of high organic loading rates.

The specific methanogenic activity (SMA) test is a powerful tool for the study of anaerobic processes. It can be used to assess the behaviour of sludge under the influence of potentially inhibitory compounds, to establish the relative degradability of various substrates, to observe changes in sludge activities due to a possible build-up of inert materials (e.g., metals after a long period of reactor operation) and to evaluate batch kinetic parameters (Monteggia, 1991).

Optimum conditions for the anaerobic process can be established using this test. The use of acetate,  $\text{CO}_2 + \text{H}_2\text{O}$ , ethanol, formate, butyrate and propionate as substrates allows evaluation of the interactions between the microorganisms responsible for acetogenesis and methanogenesis (Monteggia, 1991).

This paper describes the adverse effects of O&G on the performance of a full-scale UASB reactor treating wastewater from a slaughterhouse and meat-packing plant. In addition, it describes the solution adopted to provide stable and efficient operation of the full-scale anaerobic reactor.

## MATERIALS AND METHODS

### Wastewater Characteristics

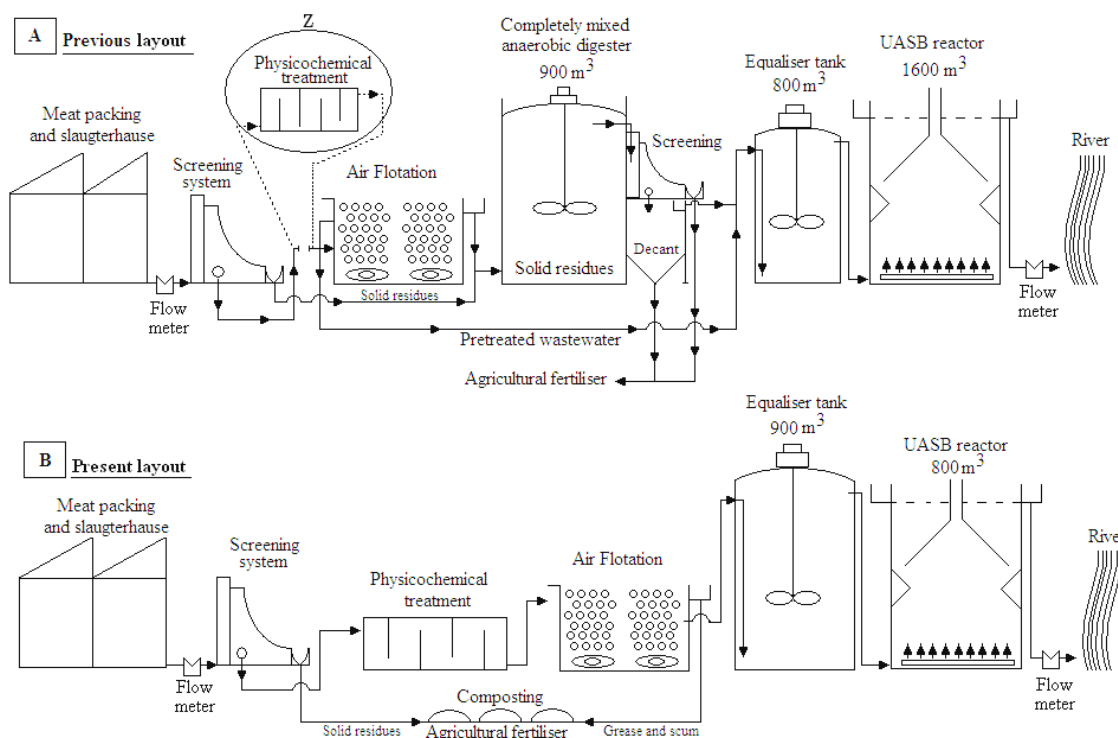
The volume of effluent from the UASB reactor, generated by the industrial process at full capacity and measured by a flow meter (Figure 1a), was 50  $\text{m}^3/\text{h}$  over a 14h operating period. Characterisation of the raw effluent was based on an intensive sampling procedure during normal operation of the plant. Samples were taken from Monday to Friday every hour during 4h in the morning and afternoon. The samples from each period were mixed to produce a one-liter sample. The average temperature was 28°C, the color was between 1800 and 3000 mg/L Pt-Co and turbidity was between 780 and 1200 NTU. Table 1 contains the average values of pH, O&G, COD,  $N_{\text{total}}$  and  $P_{\text{total}}$ , in accordance with established procedures (APHA, 1995) and  $\text{Protein}_{\text{total}}$  obtained during the intensive characterisation period.

## Effluent Treatment System (ETS)

### ▪ Previous Layout

The ETS was originally composed (Figure 1a) of a screening system for removal of solid residues generated during the slaughtering and meat-processing phases and sent for digestion in a 900 m<sup>3</sup> complete mixture anaerobic reactor. The retention time of the solids in the reactor was approximately 75 days. After stabilisation, the solids were used in agricultural fertilisation. The effluent generated

during this phase was mixed with the industrial effluent and sent to an equalising tank for temperature homogenisation and adjustment of pH. The pH was maintained between 6.8 and 7.2 by automatic addition of 40% NaOH. After equalisation (Figure 1a - Equaliser tank) of the fluxes, the effluent was sent to a 1600 m<sup>3</sup> UASB reactor. The hydraulic detention time in the UASB reactor was 54.8 h. After 150 days of operation, a physicochemical treatment was implemented with the objective of reducing the influent O&G as shown in detail "Z" (Figure 1a).



**Figure 1:** Layout of wastewater treatment system before (a) and after (b) modifications. Prenda S/A, Santa Rosa, RS, Brazil.

### ▪ The Present Layout

The present layout (Figure 1b) is comprised of a screening system for removal of solid residues and a physicochemical treatment for O&G removal. The cationic polymers tested were selected under laboratory conditions using a jar test apparatus. Nalco N9909 (quaternary ammonium-substituted hydroxy ethyl cellulose) and Adesol G9046 (linear cationic base on polyacrylamides with a high molecular weight), which have a long chain and high positive charge density. The polymer used in the full-scale system was G9046 and the chemical dosage was 20mg/L.

The pretreated wastewater was sent to an equaliser tank where temperature and pH were monitored on-line and maintained at 25 °C and between 6.8 and 7.2,

respectively. When necessary, a 40% NaOH solution was automatically added to the equalised flow to maintain the pH within the selected range.

The equalised wastewater was sent to a UASB reactor with a 800 m<sup>3</sup> capacity, an internal diameter of 9.4 m and a height of 11.6 m. The hydraulic retention time (HRT) was in the range of 18 to 27 h.

### Analytical Methods

Data were acquired from UASB reactor operation for 180 days before and after implementation of the physicochemical treatment (Figure 1a - Previous layout) and for 660 days (between March, 1999 and December, 2000) after the modifications (Figure 1b - Present layout).

The quality of the anaerobic biomass was studied during 660 days using the SMA with 2000 mg/L of acetate as substrate. Samples of anaerobic biomass were collected at 30-day intervals for SMA analyses and analysed in duplicate (Figure 6). Routine samples (COD, pH, O&G and TVS) of influent and effluent wastewater were collected twice a week and analysed in duplicate, according to established procedures (APHA, 1995).

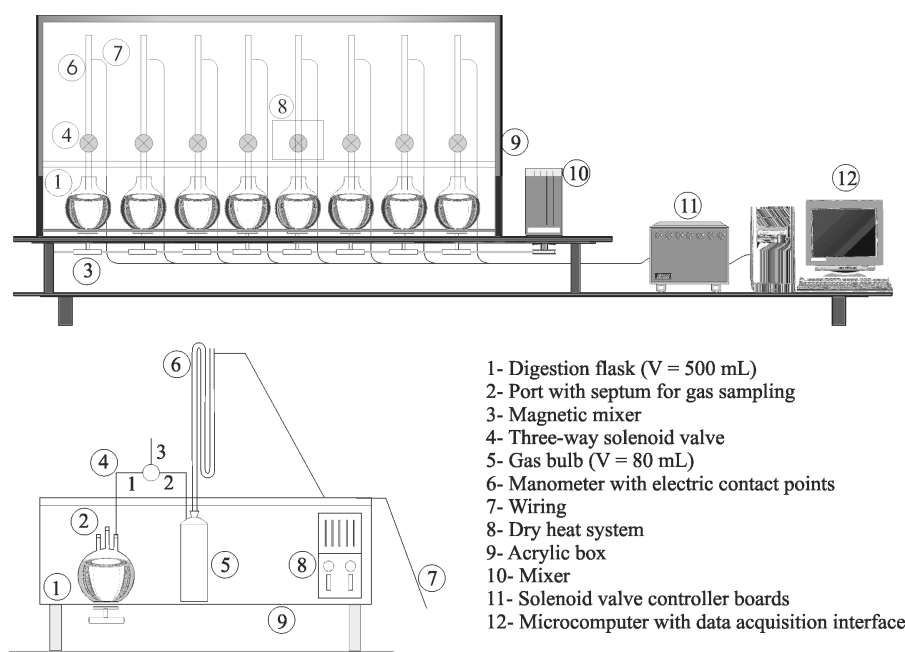
#### ▪ Determination of Protein Content

The protein content of the wastewater was determined by the bicinchonic acid (BCA) method using a BCA protein assay kit (Pierce, Rockford, IL, U.S.A.) and bovine serum albumin as the standard solution. The samples were collected following the procedures described in Wastewater Characteristics and analysed in triplicate.

#### ▪ Specific Methanogenic Activity

Determination of SMA involved samples collected 1.0 m from the base of the reactor at 30-day intervals between March 1999 and December 2000. Within that period, 22 samples were obtained and analysed in duplicate, resulting in 44 tests. The method and equipment used to determine SMA were those used by Monteggia (1991) (Figure 2). Five hundred ml glass reactors were connected to a mixing system powered by a magnetic field, thus reducing the possibility that the gas produced would leak out. Whenever desired, the mixing rate could

be changed using a velocity controller. A dry heating system with one resistance and two microventilators was used to circulate the air. A thermostat controlled the resistance. Data were acquired using semi-continuous biogas flow gauges, pressure sensors (previously calibrated using manometers), a control board that stored the records and solenoid valves. Pressure was controlled as follows. The water column in the manometer was displaced by the accumulation of gas produced. When the water column closed the circuit between the two electrodes, the storage board recorded the pulse and the solenoid valve opened, releasing a volume of gas. A sample of sludge with a TVS concentration of approximately 4000 mg/L was used. Enough of a mineral solution containing essential substances for the metabolic activity of microorganisms was added to the sludge in each reactor to make a total volume of 450 mL. The mineral solution contained  $\text{KH}_2\text{PO}_4$  ( $1.5 \text{ g.L}^{-1}$ ),  $\text{K}_2\text{HPO}_4$  ( $1.5 \text{ g.L}^{-1}$ ),  $\text{NH}_4\text{Cl}$  ( $0.5 \text{ g.L}^{-1}$ ),  $\text{Na}_2\text{S.7H}_2\text{O}$  ( $0.05 \text{ g.L}^{-1}$ ) and yeast extract ( $0.2 \text{ g.L}^{-1}$ ). After the sludge and mineral solution (dilution water) were added, the pH was adjusted with sodium bicarbonate to within the neutral range. Gaseous nitrogen was used to purge the occasional traces of oxygen and replace atmospheric air contained in the upper portion of the digestion vials (reactors). The period of sludge acclimatisation to the experimental conditions was approximately two hours, after which the substrate was added. Test was considered to have started at the moment of substrate injection (2000 mg/L of sodium acetate).



**Figure 2:** Diagram of the anaerobic respirometer used for specific methanogenic activity tests (Monteggia, 1991).

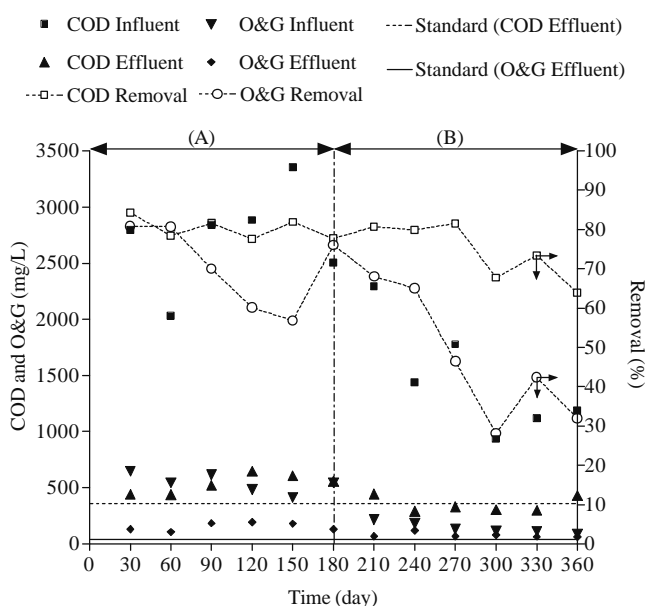
## RESULTS AND DISCUSSION

The reactor influent showed a high level of O&G in the first 180 days of operation resulting in a ratio O&G/COD of 20% (Figure 3 (A)). However weekly measurements based on composed samples (Table 1), reached values of 26-32 % in the same period.

The influent contained had high levels of O&G and total proteins (Table 1), which are the main components of this type of wastewater (Alves et al., 2001; Sanders, 2001). These components are rapidly hydrolysed to LCFA, glycerol and amino acids, respectively, in anaerobic systems (Novak & Carlson, 1970; Rinzema et al., 1994; Hwu et al.,

1998; Sanders, 2001).

The anaerobic reactor effluent COD values were high (438–646 mg/L) before implementation of the physicochemical pretreatment system (Figure 3, days 0 to 180). According to Hanaki et al. (1981), Rinzema et al. (1994), Hwu et al., (1998) and Lalman and Bagley (2002), the increase in effluent COD and O&G can be associated with the combined effects of two factors: (i) bacterial inhibition due to the acute toxicity of LCFA to both methanogens and acetogens, the two main trophic groups involved in LCFA degradation, and (ii) excessive adsorption of these substances to the sludge granules, causing the formation of a lipid film around the biomass particles.



**Figure 3:** Average monthly values for COD and O&G in the influent and effluent of the 800 m<sup>3</sup> reactor in 1997, before (A) and after (B) implementation of the physicochemical pretreatment system with polyelectrolyte. Each point represents the average of eight monthly samples.

**Table 1:** Average values of pH, O&G, COD, N<sub>total</sub>, P<sub>total</sub> and Protein<sub>total</sub> in the anaerobic reactor influent (with samples taken according to item 2.1) before implantation of the physicochemical system.

pH	COD (mg/L)	O&G(mg/L)	O&G/COD (%)	N <sub>total</sub> (mg/L)	P <sub>total</sub> (mg/L)	Protein <sub>total</sub> (mg/L)
6.8-7.4	1400-3600	452-957	26-32	133-179	25-42	51-93

Under these conditions, continuous operation of the reactor allowed an increase in of the size granules, which seemed to contain many cellulose fibres and fat particles, as the sludge was compacted in large blocks (Figure 4) due to the hydrophobicity caused by the lipid layer adsorbed to the granules. This process was probably responsible for the loss of sludge density, also resulting in the reduction in substrate diffusion into the granules and in the accumulation of biogas. In this situation, both granules and biomass blocks tended to emerge and

developed a superficial layer of scum and sludge with the consequent biomass washout. These effects may cause hydraulic problems and incomplete mixing of substrate and biomass, thus reducing the COD conversion rate.

Studies on the internal and external impact of mass transport in bacterial aggregates have produced contradictory results. While some studies report that internal and external diffusion limitations affect the rate of substrate utilisation (Dolfing, 1985; Hwu et al., 1998), thus directly affecting reactor efficiency, other

studies indicate that limitations in mass transport are not observed in biofilms as thick as 2.6 mm (Beer de et al., 1992; Schmidt & Ahring, 1991). In a study of anaerobic digestion of long-chain fatty acids in UASB reactors, Rinzema and Lettinga (1993), observed that concentrations of lipid in the influent equal to or higher than 50% COD caused a decrease in biogas production, reducing the effects of mixing and the contact between biomass and substrate.

These data and observations clearly suggest the need for a phase pretreatment of the biological reactor influent, aiming to reduce the level of O&G and SS continuously discharged into the system. The physicochemical pretreatment system had three steps: (i) chemical prehydrolysis with NaOH, (ii) enzymatic prehydrolysis using lipases and (iii) coagulation-flocculation with flocculent polymer (polyelectrolyte). In the first case, according to Masse et al. (2001), it is necessary to use from 150 to 300 meq of NaOH per effluent liter to obtain a 73% reduction in the size of fat particles generated in the process of meat slaughtering and industrialisation, in addition to significant increase in influent alkalinity

and pH. This alternative would require an operation for removing saponified fats and a high consumption of acids for the final adjustment of pH.

In the second case, the same author describes the need for high pancreatic lipase (PL-250) concentrations and over 1000 mg/L of bacterial lipase (LG-1000) to obtain a 60% reduction in fat particles, besides a four hour pretreatment time for PL-250 and a 24-hour pretreatment time for LG-1000 to obtain a reasonable results. Considering these limitations, pretreatment with polyelectrolyte was the most efficient option and was also easily carried out in the effluent treatment systems.

Similar results were obtained for the cationic polyelectrolytes Nalco N9909 and Adesol G9046. Both produced extremely well-defined flocs with a high resistance level. This effect occurred by formation of bridges, mainly between fat particles, cellulose fibres and colloidal organic matter in the influent. Adesol G9046 was selected due to the lower cost and coagulation capacity. Table 2 contains the values of O&G/COD ratios before and after the addition of polymer.



**Figure 4:** Appearance of the anaerobic sludge from the reactor before physicochemical pretreatment.

**Table 2: Range values for influent COD, O&G, O&G/COD ratio and volumetric organic load rate (OLR) before (days 0-180 - Figure 3) and after (days 180-360 - Figure 3) implantation of the physicochemical treatment.**

	COD (mg/L)	O&G (mg/L)	O&G/COD (%)	OLR (Kg COD/m <sup>3</sup> d)
Influent (days 0-180)	2030-3350	413-645	20	0.88 -1.46
Influent (days 180-360)	930-2300	87-215	9.3	0.41-1.0

After operating the physicochemical system for six months (Figure 3 – day 330 on) a severe biomass washout of the 1600 m<sup>3</sup> UASB reactor (Figure 1a - Previous layout) occurred, resulting in total failure of the system. This process probably occurred due to accumulation of O&G in the anaerobic biomass during the period that preceded implementation of the physicochemical system (Figure 3 - days 0 to180). This

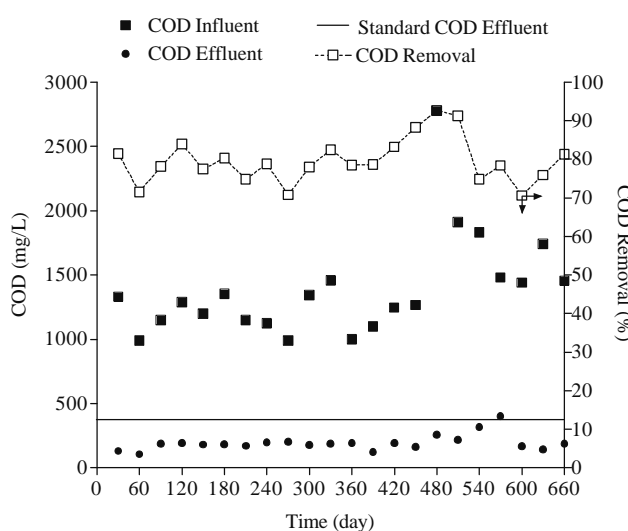
effect was also observed by Hwu et al. (1998), who studied the adsorption of oleic acid with granular sludge flotation in a UASB reactor and concluded that granular sludge flotation occurred at concentrations far below the toxicity limit, suggesting that complete washout of granular sludge would occur prior to inhibition.

After restoration of the system, a new start-up was done employing a new UASB reactor (800 m<sup>3</sup>)

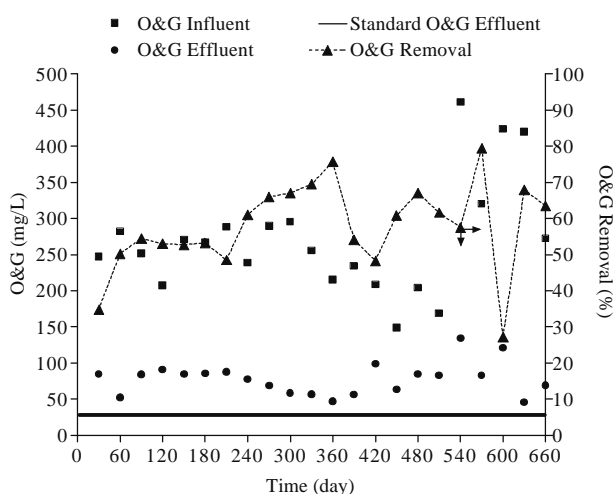
with the configuration shown in Figure 1b (Present layout). The new configuration provided stable and efficient performance in terms of COD effluent concentration ( $\leq 360$  mg/L), as demanded by environmental legislation in the state of Rio Grande do Sul (Brazil) (Figure 5). During the first year of operation of the new reactor, the monthly average of COD effluent varied from 104.7 to 201.5 (Figure 5) and in the following period (days 360 to 660 – Figure 5) it varied from 122.6 to 400.3. Only the average in the 19<sup>th</sup> month (day 570) was above (400.3 mg/L) emission standards. These results demonstrate the gradual adaptation of the biomass to the substrate as well as the beneficial effect of the physicochemical treatment, which can be verified by the decrease in the O&G/COD (influent) ratio.

However, the O&G concentration in the effluent (Figure 6) remained above the legal standards ( $\leq 30$  mg/L), showing the need for posttreatment.

Operational problems such as foaming, scum formation and washout of viable sludge are frequently reported to occur in anaerobic reactors due to the presence of lipids (Petruy, 1999; Öztürk et al., 1993; Lettinga and Hulshoff Pol, 1991, 1986). During the second period of monitoring (Figure 5-6 and Figure 7, days 0 to 660) COD removal was in the range of 70.6 to 92.6 %, showing the beneficial effect of the physicochemical treatment. During this period O&G removal varied between 27.2 and 57.9 % (Table 3). In addition it was possible to increase the OLR applied to the anaerobic reactor and also remove more COD (Table 3).



**Figure 5:** Measured values of COD in the influent and effluent of the 800 m<sup>3</sup> UASB reactor after the modifications (Figure 1b - Present layout). Each point represents an average of eight samples analysed in duplicate.



**Figure 6:** Measured values of O&G in the influent and effluent of the 800 m<sup>3</sup> UASB reactor after the modifications (Figure 1b - Present layout). Each point represents an average of eight samples analysed in duplicate.

**Table 3: Performance of the UASB reactors before and after modification.**

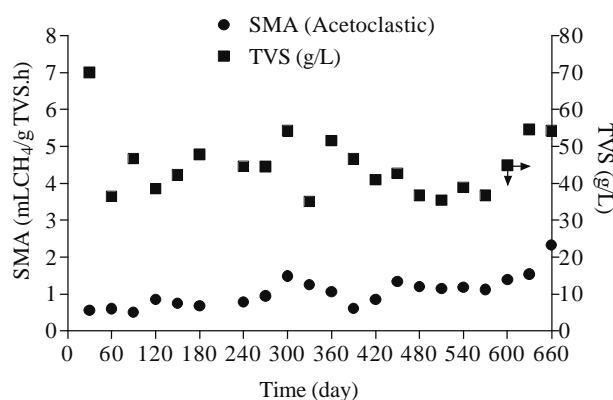
OLR (Kg COD/m <sup>3</sup> .d)	Previous Layout (Figure 1a) <sup>a</sup>			Present Layout (Figure 1b) <sup>b</sup>		
	0.88-1.46			0.86-2.43		
	Influent	Effluent	Removal	Influent	Effluent	Removal
COD (mg/L)	2030-3350	440-646	78-80 %	988-2774	104-400	70-92 %
O&G (mg/L)	413-645	130-193	68-70 %	140-460	27-134	27-58 %
pH	6.8-7.3	6.9-7.6		6.7-7.4	7.0-7.5	
Temperature (°C)	25 ± 1			25 ± 1		
O&G/COD (%)	20			14-17		

<sup>a</sup>1600 m<sup>3</sup> UASB reactor (Figure 1a – Previous layout) during days 0 to 180.

<sup>b</sup>800 m<sup>3</sup> UASB reactor (Figure 1b – Present layout) during days 0 to 660.

The SMA varied between 0.55 and 1.48 mL CH<sub>4</sub>/g TVS.h during the first year and 0.61 to 1.53 between days 360 and 630 (Figure 7). The variation in the SMA values can be attributed to the imprecisions in TVS determinations, which are a factor limiting the accuracy of the results (James et al, 1990), since the provision of an anaerobic environment which contained the necessary conditions (temperature, pH and redox potencial) and nutrients (nitrogen, phosphorus and trace elements) to obtain the maximum biological activity when utilising a test substrate were observed. The SMA measured from day 180 on showed continuous improvement of acetoclastic biomass activity. On the

other hand, in the same period there was a decrease in the TVS concentration of the inoculum used in each test. This reduction can be a reflex of the continuous loss of volatile solids with poor settling characteristics or of the expansion of the sludge blanket during sample collection. These results suggest a tendency for the biomass to stabilise, a behaviour also observed in the effluent COD values (Figure 5), which remained below the environmental standard for emissions, indicating the adaptation of the sludge to the substrate influent. Within 390 and 660 days of operation, an increase in acetoclastic SMA was observed, resulting in a better overall performance.



**Figure 7:** Average monthly SMA and TVS values for the new 800m<sup>3</sup> reactor during 660 days of operation after implementation of the physicochemical treatment system.

The capacity for methane production is a useful tool for estimating organic load in the reactor. An increase in the methanogenic capacity of the biomass allows treatment of a larger volume of wastewater in a reactor of the same volume, thereby allowing an increase in industrial production without physical alteration of the effluent treatment system, as can be seen by the results obtained after modifications of the layout.

The study suggests that the effects observed on an experimental scale and widely reported in the literature, may be case-specific and may not be

identically observable on an industrial scale, for which factors such as mixing effects, substrate-biomass interaction, adaptation of biomass to substrate, efficient removal of O&G from the influent, analysis of the volume and characteristics of the biogas produced are relevant and help in anticipating the detection of critical problems. In addition, analysis of the percentage of COD removed from the reactor alone is not sufficient to show that the process is devoid of problems. Actions to avoid biomass washout are a rigorously controlled of under the overall conditions of the operating system.



## CONCLUSIONS

Almost three years of monitoring a full-scale UASB reactor treating the effluent of a slaughterhouse and meat-packing plant with high O&G concentrations allowed the following conclusions to be drawn:

1. The O&G/COD ratio above 20% in the influent produced a gradual reduction in system efficiency, resulting in biomass washout and a general failure of the system.
2. The observed effects may be even more severe in meat-packing plant and slaughterhouse effluents due to synergism between and effects of accumulation of the various long-chain fat acids (LCFA) in the wastewater and sludge from this type of industry.
3. Our observations suggest that the inhibiting effect of O&G is related to its concentration in the influent and not to the O&G/biomass concentration ratio inside the reactor, since they have low biodegradability and tend to accumulate on the surface of sludge granules. During operation at high O&G concentrations (413-645 mg/L), the formation of biomass blocks was observed, probably as a result of hydrophobic interaction between the granules produced by the adsorbed O&G.
4. The implementation of a pretreatment using a flotation system with polyelectrolytes to remove excess O&G reduced the operational problems of active biomass washout.
5. Analysis of the rate of COD and O&G removal alone does not allow a reliable assessment of the anaerobic process in UASB reactors that treat complex effluents, particularly when the O&G/COD ratio is above 20% of the influent. In this situation, the biomass SMA and the hydraulic conditions must also be taken into consideration. The results also indicate the importance of more detailed studies on the adverse effects of O&G (mostly LCFA) on anaerobic biomass activity in UASB reactors.

## ACKNOWLEDGEMENTS

We would like to thank the meat-packing plant and slaughterhouse PRENDA S/A; GENOTOX – Genotoxicity Laboratory, Biotechnology Center – UFRGS; Institute of Hydraulic Research - UFRGS and FAPERGS for their financial support.

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