

Short communication

Anaerobic co-digestion of sewage sludge and grease trap: Assessment of enzyme addition

Andres Donoso-Bravo ^{a,b,*}, Maria Fdz-Polanco ^c^a Escuela de Ingeniería Bioquímica, Pontificia Universidad Católica de Valparaíso, General Cruz 34, Valparaíso, Chile^b INRIA-Chile, Chilean Informatics Research and Innovation Centre (CIRIC), Rosario Norte 555 of. 703, Las Condes, Santiago, Chile^c Department of Chemical Engineering and Environmental Technology, University of Valladolid, Prado de la Magdalena s/n, 47011 Valladolid, Spain

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ABSTRACT

Anaerobic co-digestion of grease trap and sewage sludge from a wastewater treatment plant is evaluated. Enzyme-lipase application, both addition and dosage, are evaluated by fitting the methane production of biochemical potential tests with the first order model. The enzyme addition effect, at 2, 5 and 10% of grease trap ($\%GT\text{VS}_{\text{FED}}^{-1}$) and the enzymes doses, between 0.25 and 1.67% (v/v), without and with grease trap presence were studied. Grease trap addition showed a negative effect on the waste biodegradability, which was completely overcome by the addition of lipase. Enzyme addition improved notably the methane production for all grease trap fractions studied. In regards to the dosage, the best result was achieved between 0.33 and 0.83% (v/v) of enzyme. The co-digestion of sewage sludge and grease trap may be a feasible process by using lipases due to the saving in operational costs and the increase in the biogas production

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1. Introduction

Anaerobic digestion is a consolidated technology in the treatment of organic solid waste. Among them, one of the most important applications has been the anaerobic treatment and stabilization of sewage sludge (SS) generated from the surplus of biomass produced in the aeration tank of activated sludge systems in wastewater treatment plants (WWTPs) [1]. From the anaerobic degradation of SS, biogas, a renewable energy source, is obtained which may be converted in thermal and/or electric energy.

Grease trap (GT) is a solid waste (scum layer) obtained from the flotation process in a WWTP, which is composed by several types of oil and fat. Normally, GT is collected and disposed in landfills; however, this waste must be stabilized prior to disposing due to strict regulations and the associated problems with this type of waste (such as foul odors). Anaerobic digestion might be used in order to degrade and stabilize GT along with SS. An important advantage of this is that the same existing facilities for sewage sludge degradation can be used, thereby not comprising further investment costs. Moreover, the GT, due to its high lipid-content, represents an attractive source for biomethanization, due

to the higher methane yield obtained when compared to proteins or carbohydrates. Anaerobic co-digestion is a process where several wastes are used as substrate, for instance, SS and lipid-rich waste [2]. Luostarinen et al. [3] found that the co-digestion of SS and GT (from meat industry) improved the biogas production and methane yield at low and high GT concentrations. Davidsson et al. [4] evaluated the anaerobic digestion of these wastes in batch and continuous pilot-scale digestion tests, obtaining that the addition of GT increased methane yield and methane potential in batch tests. In any case, the research in this field is preliminary and several operational conditions must be studied in order to improve the process.

The enzymes application can improve the anaerobic degradation of lipids, since catalyzes the hydrolysis of long chain fatty acids. Enzymes are biodegradable and harmless for the anaerobic treatment processes and aquatic ecosystems; in addition, their contribution to the BOD in the waste stream is negligible. Lipases have been used in anaerobic treatment of fat-wastewater [5–7]. However, there is a lack of literature regarding the enzyme lipase application in anaerobic co-degradation of solid-lipid waste such as GT and SS. The aim of this study is to assess the effect of lipase addition and its dosage (Biolipase L[®]) in the anaerobic co-digestion of GT and SS. A simplified mathematical model was used to estimate some kinetic parameters in order to compare the methane production profiles as well as to count with some criteria for a preliminary economical evaluation of the lipase application.

* Corresponding author at: Escuela de Ingeniería Bioquímica, Pontificia Universidad Católica de Valparaíso, General Cruz 34, Valparaíso, Chile. Tel.: +56 32 2274178.

E-mail addresses: andres.donoso@ucv.cl, [\(A. Donoso-Bravo\).](mailto:adonosobravo@gmail.com)

2. Materials and methods

2.1. Anaerobic batch tests

Batch anaerobic digestion tests were carried out to assess the SS-GT biodegradability. All the experiments were done in duplicate. Anaerobic sludge (inoculum) from a pilot-scale anaerobic digester treating mixed waste-activated sludge was used as inoculum for the anaerobic test. Serum bottles, with a volume of 60 ml were used. Reactors were incubated at 35 °C and magnetically stirred. Anaerobic biodegradability was calculated following biogas production and composition. The results of biogas test were expressed in specific units (mlCH₄ g VS⁻¹).

2.2. Substrate and enzyme characteristics

For this experiment mixed SS (60% primary sludge and 40% secondary sludge) and GT from a conventional WWTP in Valladolid (Spain) were used. 1 ml⁻¹ of macro-micronutrients and 1 g L⁻¹ of sodium bicarbonate were added in all assays in order to supply the necessary elements and enough alkalinity to maintain the pH above 7. The characterization of the substrates (SS and GT) and the inoculum for experiment 1 and 2 are presented in Table 1.

Two series of anaerobic batch experiments were carried out in order to evaluate: (1) the effect of the enzyme addition and (2) the enzyme dosage. For experiment 1, three concentrations of GT were evaluated: 2, 5 and 10 (%w as %GTVS_{FED}⁻¹). In the assays with enzyme addition a constant dose of 0.25 (% v/v) was added. In experiment 2, four enzyme doses were tested 0.25, 0.33, 0.83 and 1.67 (% v/v) and two types of batch tests were carried out: only SS and SS+GT (5%w). For all the experiments, a certain volume of inoculum and substrate (SS and GT) was added in order to maintain the substrate-inoculum ratio of 0.5 g VS_S g VS_I⁻¹. A commercial lipase, BIOLIPASA L (Catalog no. 9001-62-1), obtained from Biocion S.A., Spain, was used. This formula comes in a liquid transparent solution and has an activity of 50,000 U g⁻¹. This lipase has an increase activity from pH 7 to 10 and an optimum temperature of 30 °C, although it remains very active between 15 and 40 °C.

2.3. Kinetic model

In the case that the hydrolysis of the particulate organic matter is the rate limiting step, a first order equation (Eq. (1)) may be used to estimate the hydrolysis rate (k_h , d⁻¹) and the biodegradability extent or anaerobic biodegradability (B_0 , mlCH₄ g VS⁻¹) from a batch test [8]. Afterwards, these parameters can be used, as a first approximation at least, of the parameters of more complex models of the co-digestion in a continuous system [9,10].

$$B = B_0 \cdot (1 - \exp(-k_h \cdot t)) \quad (1)$$

where B is the methane production (mlCH₄ g VS⁻¹) and t is the time of the assay (d). Nonlinear optimization by the least squares procedure is applied to calculate the unknown parameters (B_0 and k_h) and by minimizing a cost function (Eq. (2)), which measures the difference between the experimental measurements and the corresponding simulated value. Matlab® was used to solve the least squared procedure.

$$J(\theta) = \min \sum_{t=1}^N (y_{\text{exp}}(t) - y(t, \theta))^2 \quad (2)$$

where J is the objective function, y_{exp} is the experimental methane production (as mlCH₄ g VS⁻¹), y is the corresponding simulated value, θ is the vector of parameters and N is the number of measurements. The accuracy of the estimated parameters was obtained through the Fisher information matrix (FIM), which summarizes the quantity and quality of information obtained in the experiment and assuming proper model selection with no data autocorrelation and uncorrelated error, the inverse of the FIM (Eq. (3)) gives us an estimation of the parameter estimation covariance matrix (C_j)

$$C_j = (F(\theta))^{-1} \text{ where } F(\theta) = \sum_{i=1}^N \left[\frac{\partial y_i(t, \theta)}{\partial \theta} \right]^T Q_i^{-1} \left[\frac{\partial y_i(t, \theta)}{\partial \theta} \right] \quad (3)$$

Finally, once the covariance matrix is available, an approximation of the standard deviation (σ) of the parameters can be estimated through Eq. (4).

$$\sigma(\theta_i) = \sqrt{C_j} \quad (4)$$

Table 1
Substrate and inoculum characterization.

	Grease trap		Sewage sludge		Inoculum	
	Exp 1	Exp 2	Exp 1	Exp 2	Exp 1	Exp 2
TS (g L ⁻¹)	35.1	28.5	27.9	32.1	29.3	28.3
VS (g L ⁻¹)	26.1	26.8	15.5	23.3	16.7	15.7
Grease (g L ⁻¹)	22.1	22.7	–	–	–	–

2.4. Analytical methods

Total solids (TS) and volatile solids (VS) concentrations in all samples were determined by heating at 105 °C during 24 h for total solids and 550 °C during 2 h for volatile solids concentration and grease content by Soxhlet extraction according to the procedures described in Standard Methods for Examination of Water and Wastewater [11]. Biogas volume was measured manually by a pressure transmitter (Druck, PTX 1400, range 1 bar) in the head space of each reactor. After the daily pressure measurement, the biogas in the head space was released, what reduced the pressure in the head space to atmospheric pressure. This pressure difference was converted into biogas volume, using the ideal gas Law and standard conditions ($P=1$ bar and $T=0$ °C) as reference. The methane content was measured by gas chromatography through the injection of 1 ml of sample directly in the column.

3. Results and discussion

3.1. Enzyme addition influence

The kinetic parameters B_0 and k_h , obtained from fitting Eq. (1) with the cumulative methane production data for each experiment, are shown in Table 2.

Fig. 1 shows that the addition of the lipase increases the total methane production of the anaerobic co-digestion of SS and GT in batch conditions for all the GT concentrations. Nonetheless, the methane content of the biogas was not affected by the use of the lipase, keeping a value within 63–68%. In Table 2 (experiment 1) the estimated values of the kinetic parameters along with their respective standard deviation of the model are presented. The enzyme addition caused a notable enhancement of the waste biodegradability for all the studied conditions in regards to those assays without enzyme. For 2%, 5% and 10% of GT, the enzyme addition increased B_0 , in about 130%, 127% and 78%, respectively. This sharp increase may be explained by the fact that without the enzyme, the presence of GT may have impaired the substrate accessibility for the anaerobic biomass [12], which was not the case when enzyme was present. These mass transfer problems had to take place, as it is confirmed by the results discussed in the next sub-section, since the hydrolysis of GT as such does not justify the values of the increase in the biodegradability. In regards to the hydrolytic coefficients, the obtained values are in the range of the common reported values for anaerobic degradation of solid waste [13]. However, the lipase addition does not produce any significant change in the solid properties since the coefficient values do not vary substantially.

For all GT concentrations enzyme doses, the first order model adequately described methane production as it can be observed from the values of the determination coefficient (r^2) which indicates the goodness-of-fit of the model

3.2. Enzyme dosage effect

Once the effect of the lipase addition was evaluated, several enzyme doses (0.25, 0.33, 0.83 and 1.67%, v/v) were tested, with and without the presence of GT. Nevertheless, it is worth to point out that this experiment was carried out with substrate and inoculum from the same origin but taken at different time, which can certainly exert an effect on the results, as discussed below. The enzyme addition without GT (only SS as substrate) was tested in order to evaluate the potential effect of the enzyme on the SS degradability and its contribution to the total methane produced. Figs. 2 and 3

Table 2

Estimated parameter values of the anaerobic degradation of sewage sludge and grease trap of the two evaluated experiments.

GT content (% w)	B_0 (mlCH ₄ g VS ⁻¹)	k_H (d ⁻¹)	r^2	B_0 (mlCH ₄ g VS ⁻¹)	k_H (d ⁻¹)	r^2
	Without-enzyme			With-enzyme		
Experiment 1						
2	140.3 ± 3.6	0.091 ± 0.008	0.940	322.1 ± 4.4	0.091 ± 0.004	0.984
5	160.9 ± 3.6	0.080 ± 0.006	0.965	365.3 ± 5.3	0.115 ± 0.007	0.976
10	243.5 ± 4.4	0.126 ± 0.010	0.957	434.3 ± 5.0	0.112 ± 0.005	0.985
Enzyme dose (% v/v)	B_0 (mlCH ₄ g VS ⁻¹)	k_H (d ⁻¹)	r^2	B_0 (mlCH ₄ g VS ⁻¹)	k_H (d ⁻¹)	r^2
	SS			SS + 5% GT		
Experiment 2						
0.25	349.6 ± 2.8	0.288 ± 0.009	0.997	299.6 ± 2.6	0.270 ± 0.009	0.997
0.33	292.5 ± 3.3	0.293 ± 0.013	0.994	452.4 ± 3.7	0.220 ± 0.006	0.998
0.83	368.1 ± 3.3	0.239 ± 0.007	0.997	469.5 ± 4.4	0.209 ± 0.006	0.997
1.67	363.8 ± 9.4	0.209 ± 0.017	0.979	393.5 ± 6.3	0.260 ± 0.015	0.992

show the methane production and the model fit for the batch tests with SS as substrate and the co-digestion with GT, respectively. The parameters B_0 and k_H obtained from the fitting procedure are shown in Table 2 (experiment 2).

In regards to the application of enzyme in the anaerobic degradation of sewage sludge, the B_0 remains mostly constant in all enzyme doses. The methane content of the biogas remains within the range observed for experiment 1. The cumulative profile of methane obtained at 0.33% (v/v) appears to be a result of some problems in the pressure measurement or methane leakage since there is not a consequent decrease for the next enzyme dose. On the other hand, a slight decrease of the k_H is observed as the enzyme doses increase, which must have been caused by some ammonia accumulation such that the change in the methane production profile lead to some parameters underestimation. This has been previously observed when lipases are used in anaerobic degradation of sewage sludge [14].

When GT is added (5%), there is a clear positive effect of the enzyme addition on the biodegradability when the dose is equal or higher than 0.33%. At 0.25%, there was not a clear effect of the enzyme addition on the degradation kinetic compared to the one performed at the same conditions in experiment 1, which has to be related with the GT characteristics used for this experiment. Lipases action is an interfacial phenomenon which depends on the concentration and the interface characteristics, thereby in this case, a 0.25% is a too low concentration in order to overcome the mass transfer limitation for the lipase can access to the lipids present in the GT. Furthermore, these interfacial problems have been seen increased when substrates with a high suspended solids content are used, such as SS [15]. The best results are achieved when (0.83%, v/v) of enzyme was used. However, the obtained biodegradability at 0.33% (v/v) is only a 3% less, thereby in cost-benefit terms; the lower dose may be the best option. At the highest dose, a reduction in the biodegradability is observed. The decrease in B_0 indicates that

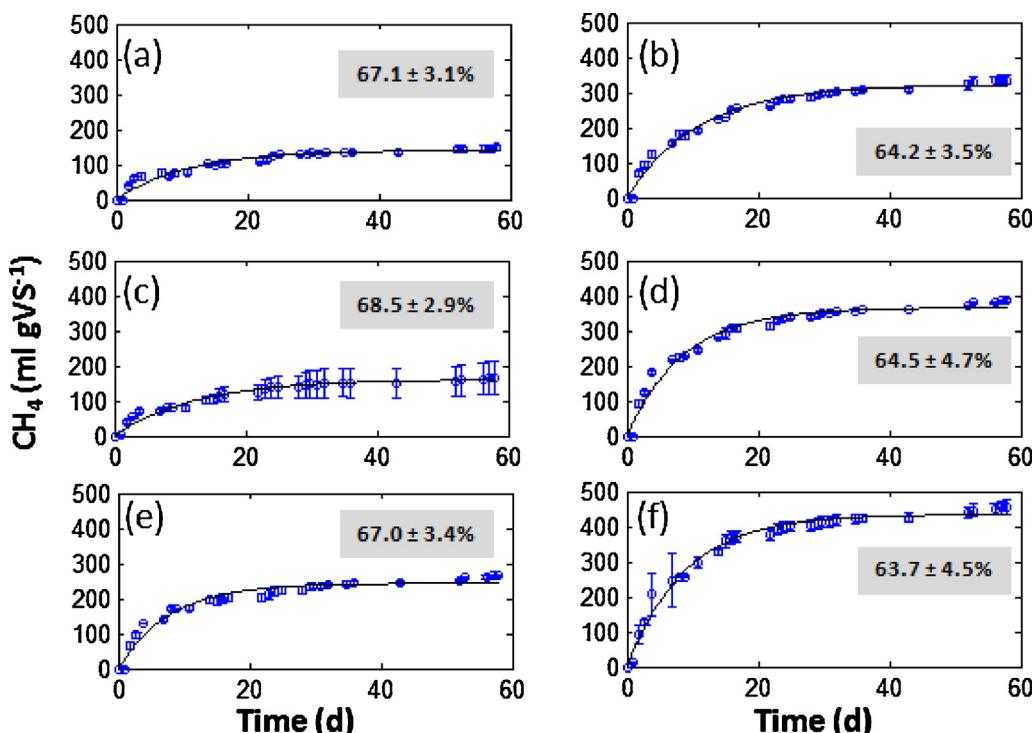


Fig. 1. Methane production from the anaerobic digestion of sewage sludge and grease trap (a) 2% GT (b) 2% GT + lipase (c) 5% GT (d) 5% GT + lipase (e) 10% GT (f) 10% GT + lipase. Solid line: model prediction, blue points: experimental information. The percentage of the methane in the biogas is shown in the grey textbox. (For interpretation of the references to color in figure legend, the reader is referred to the web version of the article.)

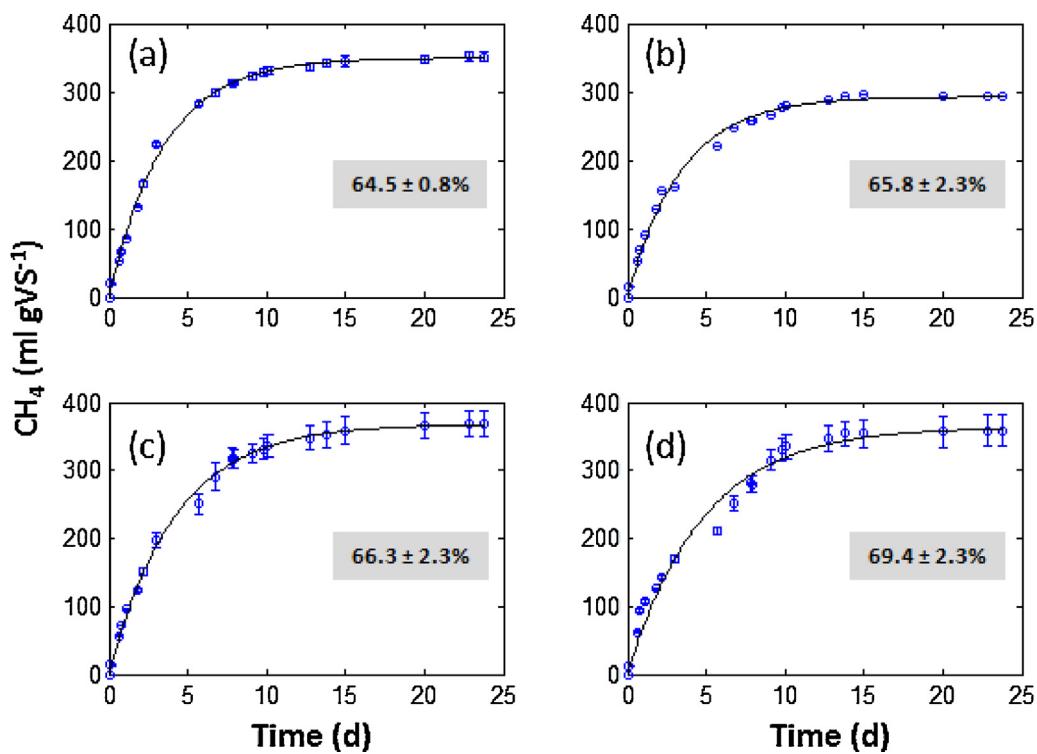


Fig. 2. Methane production from the anaerobic digestion of sewage sludge at several enzyme doses (a) 0.25 (b) 0.33 (c) 0.83 and (d) 1.67% (v/v). Solid line: model prediction, blue points: experimental information. The percentage of the methane in the biogas is shown in the grey textbox. (For interpretation of the references to color in figure legend, the reader is referred to the web version of the article.)

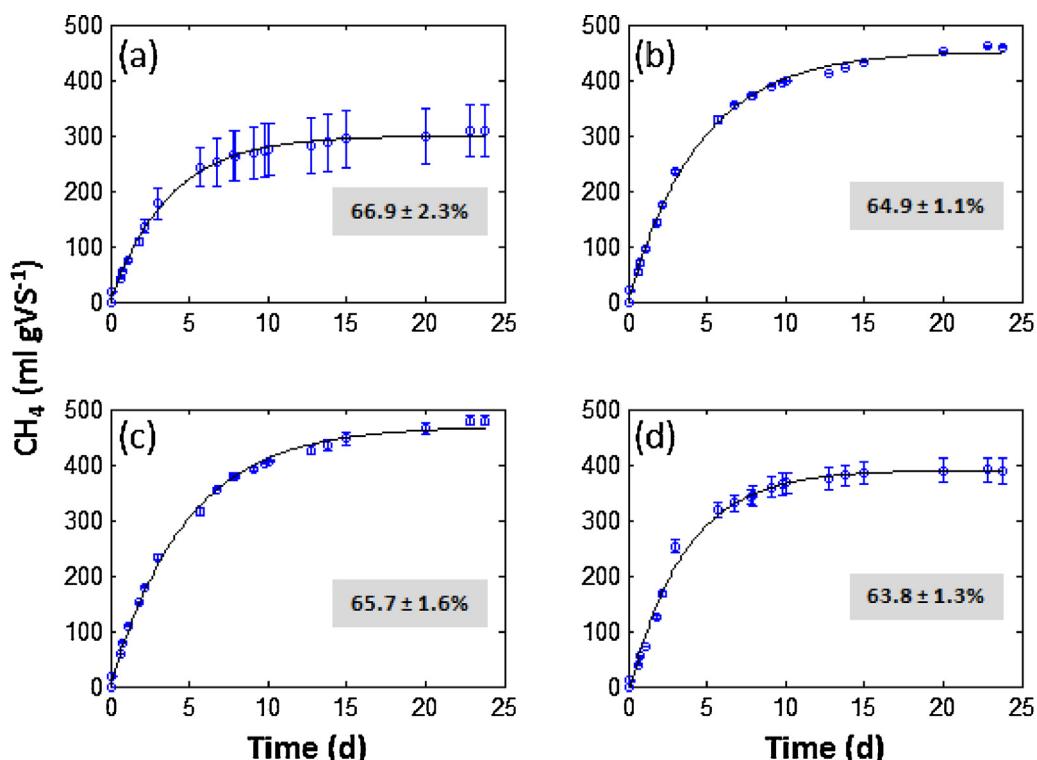


Fig. 3. Methane production from the anaerobic co-digestion of sewage sludge and grease trap at several enzyme doses (a) 0.25 (b) 0.33 (c) 0.83 and (d) 1.67% (v/v). Solid line: model prediction, blue points: experimental information. The percentage of the methane in the biogas is shown in the grey textbox. (For interpretation of the references to color in figure legend, the reader is referred to the web version of the article.)

some of the products from the lipids hydrolysis were not degraded during the assay time. Long chain fatty acid (LCFA), known for being slowly anaerobically degraded, have been observed to be produced when high concentration lipases are used [16].

As the previous case, the reaction curve-type model agrees with the experimental data for all enzyme doses, showing determination coefficients (r^2) above 0.99.

4. Economical evaluation

The economical assessment of the benefit of applying lipase for the anaerobic co-digestion of SS and GT cannot be done directly since batch tests were used in this case, and continuous or semi-continuous systems are usually used in full scale plants. Nevertheless an approximation can be made through the use of the kinetic parameters estimated during the fitting process. Taking as example, for instance, the results from experiment 1 (with 5% of GT and an enzyme dose of 0.25%, v/v) and a given waste flow of $100\text{ m}^3\text{ d}^{-1}$, the daily cost of enzyme addition would be roughly 4225 € d^{-1} . This cost should be offset by the reduction in investment and operational expenses of the digester. Taking the inverse of the hydrolytic coefficient, for the case when no enzyme was added a minimum of 12.5 d as HRT is obtained whereas for the case of enzyme usage 8.7 d, hence a reduction of around 30% in the HRT, consequently in the size (volume) of the digester. Less digester volume entails less heating, mixing, investment, among other cost. On the other hand, and from the anaerobic biodegradability parameters more than double of methane may be generated. This increase in the methane production may represent a significant benefit, which, nonetheless, it depends on the local conditions in regards to the energy sector. Other important aspects to take carefully into account are the impact of the enzyme application on the dewaterability properties and the disposal requirements as well as the final dealt price of the enzyme which can be negotiated as a wholesale.

5. Conclusion

Grease trap from wastewater treatment plant is an increasing concern in the plant solid waste management. This study demonstrates the feasibility of the anaerobic co-digestion of sewage sludge and grease trap by using lipase. The enzyme addition, lipase-type, can benefit the biodegradation process, increasing significantly the production rate and the total production of methane. Lipase doses between 0.2 and 0.5 ml (0.33–0.83%, v/v) showed the best results in terms of biodegradability enhancement. First order model may be used to obtain some kinetic parameters from the methane production, which can be used to get a preliminary basic design of a continuous process.

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