ELSEVIER

Bioresource Technology 80 (2001) 37–43

BIORESOURCE TECHNOLOGY

Influence of different natural zeolite concentrations on the anaerobic digestion of piggery waste

Z. Milán [a], E. Sánchez [b], P. Weiland [c], R. Borja [d,*], A. Martín [e], K. Ilangovan [f]

[a] *División de Estudios sobre Contaminación Ambiental (DECA-CNIC), P.O. Box 6990, La Habana, Cuba*
[b] *División de Consultores Ambientales (CONAM), Calle 23, No. 802, Esq. B, Vedado, La Habana, Cuba*
[c] *Institut für Technologie und Biosystemtechnik (FAL), Bundesallee 50, D-38116 Braunschweig, Germany*
[d] *Instituto de la Grasa (C.S.I.C). Avda Padre García Tejero 4, E-41012 Sevilla, Spain*
[e] *Dpto. Ingeniería Química, Facultad de Ciencias, Edificio C-3, Campus Universitario de Rabanales s/n, Ctra. Madrid-Cadiz, Km 396, 14071 Córdoba, Spain*
[f] *Instituto de Ingeniería de la UNAM, P.O. Box 70-472, D.F., 04510, Mexico*

Received 28 December 2000; received in revised form 18 March 2001; accepted 1 April 2001

Abstract

The effect of different natural zeolite concentrations on the anaerobic digestion of piggery waste was studied. Natural zeolite doses in the range 0.2–10 g/l of wastewater were used in batch experiments, which were carried out at temperatures between 27°C and 30°C. Total chemical oxygen demand (COD), total and volatile solids, ammonia and organic nitrogen, pH, total volatile fatty acids (TVFA), alkalinity (Alk) and accumulative methane production were determined during 30 days of digestion. The anaerobic digestion process was favored by the addition of natural zeolite at doses between 2 and 4 g/l and increasingly inhibited at doses beyond 6 g/l. A first-order kinetic model of COD removal was used to determine the apparent kinetic constants of the process. The kinetic constant values increased with the zeolite amount up to a concentration of 4 g/l. The values of the maximum accumulative methane production (G_m) increased until zeolite concentrations of 2–4 g/l. The addition of zeolite reduced the values of the TVFA/Alk ratio while increasing the pH values, and these facts could contribute to the process failure at zeolite doses of 10 g/l.

Keywords: Anaerobic digestion; Piggery wastewater; Stimulation; Natural zeolite; Kinetics

1. Introduction

One of the most important problems of pig breeding is the waste management which requires an adequate system of treatment and disposal. The high strength and large volume of piggery wastes cause a nuisance effect to the environment. Among the different wastewater treatments employed to reduce the pollution of piggery wastes, anaerobic digestion is considered one of the most promising processes (Kimchie et al., 1988; Hobson and Shaw, 1973, 1974; Hobson, 1981, 1985, 1992; Sánchez et al., 1995).

Anaerobic processes are efficient in reducing the concentration of organic matter of piggery waste. The utilization of the methane gas obtained at the process could give a significant reduction of the cost of waste treatment. Anaerobic treatment processes have also been applied to very strong and soluble wastes. It has been pointed out that an alternate aerobic – anaerobic treatment is a very efficient system to reduce the organic and nutrient concentrations (Varstaen, 1975; Sánchez et al., 1995). Anaerobic treatment of piggery waste offers the advantage that the biogas produced contains 50–70% of methane. On the other hand, removal efficiencies between 75% and 85% in chemical oxygen demand (COD) have been obtained at digestion times of 20–30 days at 35°C, and 60–70% at 25°C, using batch processes (Sánchez et al., 1995).

Clay minerals and other surface – active materials have been reported to influence microbial and enzymatic transformation of a variety of substances, including ammonium, sulfur, carbohydrates, proteinaceous materials and phenolic compounds (Pérez et al., 1989; Borja et al., 1993a, 1996). In addition, according to previous results (Stotzky and Rem, 1966; Sánchez and Roque, 1987; Borja and Banks, 1993; Borja et al.,

---

* Corresponding author. Tel.: +34-95-4692516; Fax: +34-95-461-6790 or +34-95-4691262.

E-mail address: rborja@cica.es (R. Borja).

0960-8524/01/$ - see front matter
PII: S0960-8524(01)00064-5

1993a,b,c,d, 1996), zeolite has been found to be a successful microbial support in mesophilic anaerobic digestion of different wastewaters, due to the following characteristics:

1. The high capacity for immobilization of microorganisms.
2. The capacity for improving the ammonia/ammonium ion equilibrium.
3. The possibility of reducing the ammonia and ammonium ions in solution.

Natural zeolite has also been used in other steps of several wastewater treatment technologies. This material is an efficient ion exchanger for removing the ammonium ion from anaerobic effluents and municipal wastewater. Ion-exchange processes using homoionic natural zeolite have shown ammonium removal from waters with efficiencies close to 95% (Milán et al., 1997). Zeolite is also a good adsorbent of small molecules. On the other hand, natural zeolite is an attractive filtering media not only for effluents with low suspended-organic content but also for high suspended-matter effluents (Reyes et al., 1997; Milán et al., 1999). In the present work, the effect of different natural zeolite doses on anaerobic digestion of piggery waste was studied in laboratory-scale reactors operating in batch conditions.

2. Methods

2.1. Piggery waste

Piggery waste manure from a farm of 4500–5000 pigs located near to the laboratory (Havana city) was used as substrate. A manure–water dilution ratio 1:3 was prepared so as to obtain a waste with the typical composition of the greater part of cuban pig farms. The characteristics of the substrate are shown in Table 1.

Table 1
Characteristics of the piggery waste used[a]

Parameters	Average value[b]
Total COD	26 800
TS	22 100
VS	16 100
TSS	11 100
VSS	9100
Total phosphorous	230
Organic nitrogen	860
Ammonia nitrogen	410
TVFA	5270
Alkalinity	3380
pH	6.8

[a] TS, total solids; VS, volatile solids; TSS, total suspended solids; VSS, volatile suspended solids; TVFA, total volatile fatty acids (as acetic acid). All units, except pH, are expressed in mg/l.

[b] Values are averages of 15 determinations on different lots of waste; there was virtually no variation (less than 5%) between analyses.

2.2. Natural zeolite used

The zeolite used in the experiments was obtained from the Tasajera deposit located in Villa Clara Province, Cuba. The characteristics of the zeolite employed are given in Table 2. The natural zeolite had a particle size $\leqslant 1.0$ mm.

2.3. Equipment

The experiments were carried out at laboratory-scale using 11 batch reactors, each with a working volume of 5 l. The reactors were hermetically closed by rubber caps provided with two holes, one for sample extraction and raw waste addition to restore the original volume and the other for the biogas outlet. Each experiment was performed with the addition of a different dose of natural zeolite. The digesters were mixed by a magnetic stirrer at 350 rpm.

The methane produced in the process was measured by water displacement using 2 l Mariotte reservoirs fitted to the reactors. Tightly closed bubblers containing a NaOH solution (15%, w/v) to collect the CO_2 produced in the process were intercalated between the reactors and Mariotte reservoirs. The cumulative volume of methane produced from each digester was measured every day. The digestion time was 30 days. The experiments were carried out at room temperature, which varied in the range 27–30°C. Each experimental run was carried out in duplicate. As the deviations between replicate samples were always less than 3%, mean values are reported in the corresponding tables and figures.

2.4. Inoculum

The inoculum of each digester was 1 l of well-digested piggery sludge (180 days retention time) obtained from a full-scale anaerobic digester near the laboratory. The characteristics of the inoculum used are summarized in Table 3.

Table 2
Characteristics and phase composition of the natural zeolite used

Chemical composition (%)		Phase composition (%)	
SiO_2	58.05	Clinoptilolite	35
Al_2O_3	11.94	Mordenite	15
Fe_2O_3	4.36	Montmorillonite	30
MgO	0.77	Others[a]	20
CaO	5.94		
Na_2O	1.5		
K_2O	1.2		
IW[b]	12.09		
Total	95.85		

[a] Others – Calcite, Feldspar and Quartz.

[b] IW – Ignition wastes.

Table 3
Characteristics of the inoculum[a]

Parameter	Value	Units
COD	65 300	mg/l
TS	6.8	%
VS/TS	0.61	–
pH	7.4	–

[a] Values are averages of 5 determinations of a single lot of biomass used as inoculum. There was virtually no variation (less than 3%) between analyses.

2.5. Experimental procedure

The digesters were inoculated as mentioned above. Four liters of piggery waste were added to each digester. Eleven experimental runs were performed by adding 0.2, 0.4, 0.6, 0.8, 1, 2, 4, 6, 8 and 10 g of zeolite per liter of waste contained in the reactor and one reactor without zeolite was used as control. After zeolite and wastewater addition the cumulative methane production was measured. The duration of each experiment was 30 days, the time interval for maximum gas production and COD removal in each case.

2.6. Analyses

Basic process parameters were determined twice a week taking 100 ml of sample. The following parameters were determined: COD, total and suspended solids, nitrogen (organic and ammonia), pH, total volatile fatty acids (TVFA) and alkalinity (Alk). These analyses were carried out according to Standard Methods for the Examination of Water and Wastewater (APHA, 1989). Ammoniacal N determination was carried out by distillation of the samples previously buffered at pH 9.5 with a borate buffer solution and titration with NaOH of the distillates collected in excess sulfuric acid.

2.7. Kinetic model

In order to evaluate kinetically the effect of zeolite addition on the anaerobic digestion of piggery wastewater the first-order kinetic model developed by Grau et al. (1974) for COD degradation was used

$$S/S_0 = \exp -[k_1\ t(X_0/S_0)], \tag{1}$$

where S is the concentration of organic matter (g COD/l) at any digestion time, t (days), S_0 is the initial substrate concentration (g COD/l), k_1 is the first-order kinetic constant (days^{-1}) and X_0 is the initial concentration of microorganisms expressed as grams of volatile suspended solids (VSS) per liter. Taking into account that the values of X_0 and S_0 are assumed constant during the experiment and the product $k_1(X_0/S_0)$ is an apparent constant (k'_1), Eq. (1) is simplified to

$$S/S_0 = \exp -(k'_1 t). \tag{2}$$

To calculate the value of the apparent kinetic constant, k'_1, Eq. (2) is transformed as follows

$$-\ln(S/S_0) = k'_1\ t \tag{3}$$

indicating that a plot of $-\ln(S/S_0)$ versus t should give a straight line with a slope equal to k'_1 with intercept zero if the model is applied to the experimental data.

3. Results and discussion

Figs. 1 and 2 show the values of accumulative methane production (G) and COD, respectively, during the digestion time. The final COD decreased as the zeolite dose increased and the minimum COD value was obtained at doses in the range of 2–4 g/l. A further increase of the zeolite doses showed a significant decrease of the organic matter removal. Fig. 1 shows the accumulative methane production (G) at different doses of zeolite. It can be observed that the value of G during the first week of experiment increased at zeolite doses in the range 0.2–6 g/l indicating that zeolite was a stimulant for the process. When the zeolite doses increased beyond 6 g/l (digester numbers 10 and 11) the methane production dropped significantly and the form of the curves

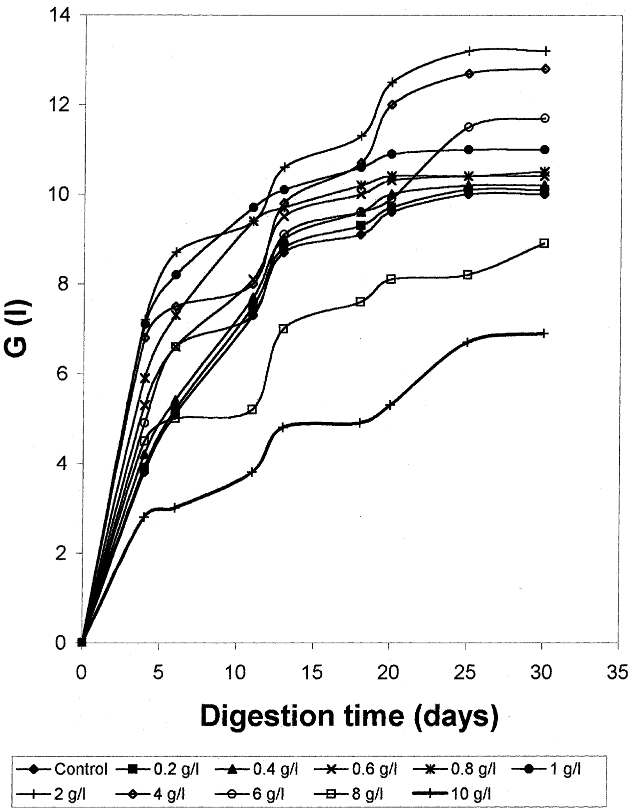

Fig. 1. Variation of the volume of methane accumulated, G (l), with time (days) for the different zeolite doses used.

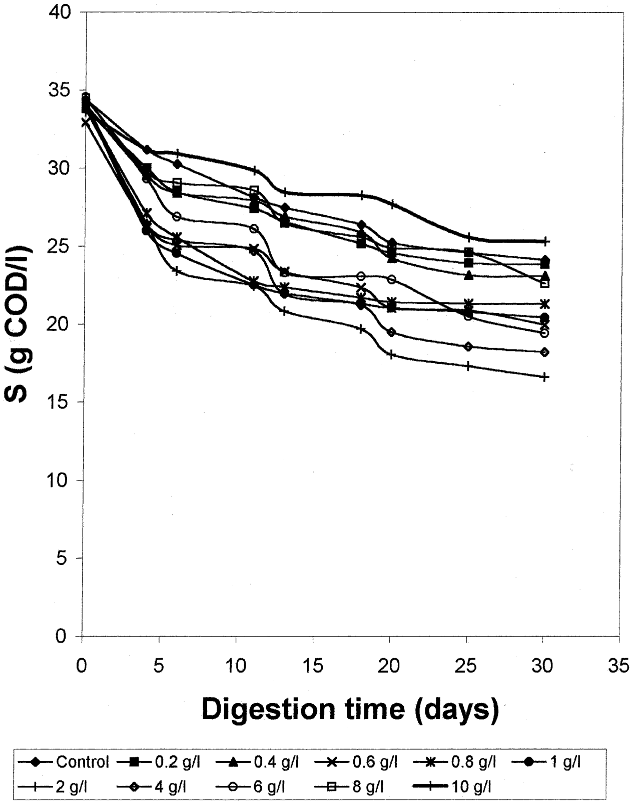

Fig. 2. Variation of the COD values (g/l) with time (days) for the different zeolite doses used.

changed drastically during the first seven days, showing small regions of low or no production of methane. The maximum methane production (G_m) was also influenced by the zeolite doses. Fig. 3 shows the values of G_m obtained at different doses of zeolite. The maximum value of G_m was in the range 2–4 g/l, and the minimum at 10 g/l. The effect of natural zeolite on G_m could be explained by the increase of the organic matter degraded during the digestion and, therefore, of the methane volume produced for the above-mentioned doses, considering that G_m is the product of the initial substrate concentration (S_0, g COD/l) and the methane yield coefficient (Y_p, l methane/g COD removed) (Borja et al., 1993c,d).

Fig. 4 shows a plot of $-\ln(S/S_0)$ values as a function of digestion time (days) for some zeolite doses, as an example. Representation of the experimental data as indicated, Eq. (3), gives straight lines with the intercept virtually at zero. Table 4 shows the values of the first-order kinetic constant for COD removal determined according to Eq. (3). The values of the constants were very similar in the range 0–1 g/l, under such doses the influence of zeolite on the process was not significant. At doses in the range 2–4 g/l, the kinetic constant increased between 80% and 100 % compared to the control. At higher doses the kinetic constant decreased, achieving a minimum value of 0.01 d^{-1}, for a zeolite dose of 10 g/l, 17% lower than that observed for the control. The significant reduction on the methane gas production, the maximum methane production and the apparent first-order kinetic constant at doses higher than 8 g/l could be explained by different factors. The increase of total solids concentration due to the zeolite addition provokes at the same time a reduction of the free available water affecting the transport of nutrients and metabolites in

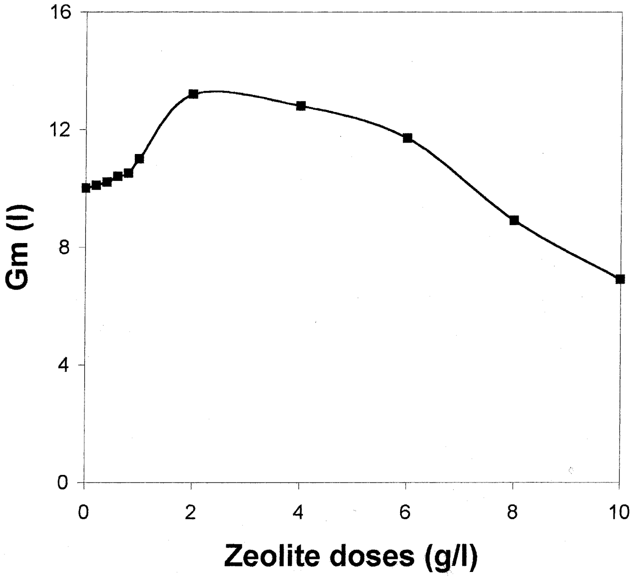

Fig. 3. Effect of the zeolite doses on the maximum volume of methane accumulated (G_m).

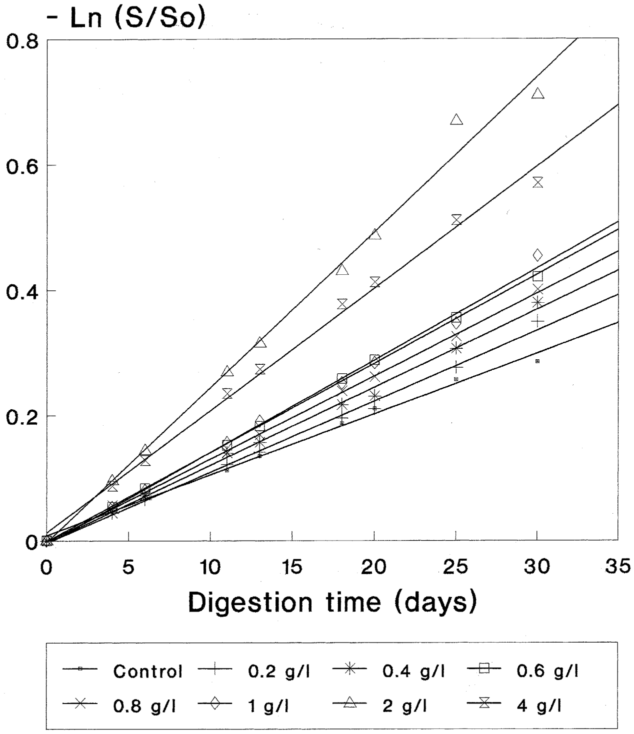

Fig. 4. Variation of the $-\ln(S/S_0)$ values with time for some zeolite doses.

Table 4
Apparent kinetic constant values for each dose of zeolite used[a]

Zeolite doses (g/l)	k_1' (d^{-1})
Control	0.012
0.2	0.011
0.4	0.012
0.6	0.014
0.8	0.013
1	0.014
2	0.024
4	0.021
6	0.019
8	0.013
10	0.010

[a] The variance coefficient (VC) was lower than 5% in all cases.

the vicinity of the zeolite particles and the microorganisms associated. Therefore, large amounts of zeolite could increase the apparent viscosity of the medium, thereby hindering mass transfer between the substrate and microorganisms responsible for the process and decelerating the process. In addition, a change of the NH_4^+–NH_3 equilibrium could also affect the performance of the process. The organic and ammonia nitrogen concentrations at different zeolite doses are summarized in Table 5. As can be seen, the concentration of ammonia exchanged and adsorbed increased with the zeolite doses, but the organic nitrogen concentration at the end of the experiment was not affected considerably by zeolite doses in the range 0.2–8 g/l. However, the ammonia concentrations in the digester liquors were higher as the zeolite doses were augmented. The presence of zeolite contributed to reduce the concentration of NH_3 and NH_4^+ in the solution, so reducing the inhibitory effect of these compounds. The addition of zeolite could reduce both NH_4^+, by ion exchange delivering Mg^{+2}, Ca^{+2} and Na^+ to the digester liquor, and NH_3, by adsorption of this species on the active areas of the material; the two mechanisms were favorable for the anaerobic digestion and a greater amount of ammonia could appear in the solution without causing process failure because the relative concentration of free ammonium ion decreased. A slight increase of the ammonia nitrogen concentration was observed at zeolite doses from 0.2 to 0.6 g/l. But when the doses increased a clear tendency to increase the ammonia nitrogen concentration was found with a maximum value observed at 10 g/l. The ammonia nitrogen concentration determined by the analysis was a partial measurement of the total ammonia concentration because it included ionized ammonia (NH_4^+) and a fraction of the non-ionized species (NH_3, NH_2O), and most of the latter could not be determined in the analysis because a part of these compounds escaped from the solution. Taking into account that the organic nitrogen compounds (protein, aminoacids, amines and urea) are transformed to ammonia as final product of the anaerobic metabolism, an equivalent amount of ammonia nitrogen (that includes NH_4^+, NH_3 and NH_2O) must be increased over its initial value. The theoretical concentration of ammonia specie in the liquid fraction is the result of a material balance and it is the difference between the theoretical concentration of ammonia nitrogen and the fraction removed by the zeolite. The toxic effect of ammonia nitrogen increased with the concentration and the pH. In the case of zeolite doses in the range 0.2–4 g/l the pH value varied from 6.8 to 7.3, while for doses between 6 and 8 g/l the pH was around 7.8 and, finally, at a dose of 10 g/l the value of pH was 8.1. The toxicity of ammonia is strongly influenced by the pH, which determines the equilibrium concentration of free ammonia to the ammonium ion in solution. Values as low as 150 mg/l of NH_3–N have been reported as being toxic to anaerobic digestion at pH 8 (Kugelman and McCarty, 1965). The increase of the concentration of ammonia in the supernatant of the digesters caused the reduction of the TVFA/Alk ratio, due to an overall increase of the Alk

Table 5
Influence of zeolite doses on the N concentration (organic and ammonium) after 30 days of digestion[a]

Zeolite doses (g/l)	Organic N (mg/l)	NH_4^+ (mg/l)	Concentration of NH_4^+ in the zeolite[b] (mg/g)
0	862	414	–
0.2	393	470	5.31
0.4	371	459	6.64
0.6	371	492	6.70
0.8	371	537	7.50
1	360	560	7.96
2	387	580	9.00
4	371	546	9.82
6	368	595	11.06
8	364	683	11.43
10	249	722	13.48

[a] Values are averages of 3 determinations on samples taken after 30 days of digestion. The differences between the observed values were less than 3% in all cases.

[b] Initial concentration of ammonia in natural zeolite: 2.59 mg NH_4^+/g.

Table 6
Influence of zeolite doses on the theoretical concentration of total ammonia in solution and TVFA/Alk ratio at the end of the digestion process

Zeolite doses (g/l)	Theoretical concentration of total ammonia in the solution (mg/l)	TVFA/Alk ratio at 30 days of digestion[a]
0	872	0.33
0.2	882	0.32
0.4	903	0.33
0.6	902	0.29
0.8	901	0.28
1	910	0.26
2	892	0.27
4	876	0.26
6	862	0.27
8	841	0.24
10	920	0.20

[a] Values of TVFA/Alk ratio are averages of 3 determinations on samples taken after 30 days of digestion. The differences between the observed values were less than 3% in all cases.

values, as could be observed in Table 6, in which are given the calculated values of total ammonia in the solution and the ratio of TVFA/Alk at different zeolite doses. From Table 6 it could be appreciated that the TVFA/Alk ratio had a tendency to decrease when the zeolite doses increased. The maximum concentration of total ammonia and the minimum TVFA/Alk ratio was found for a zeolite dose of 10 g/l, for which, the apparent kinetic constant achieved its lowest value and process inhibition appeared.

4. Conclusions

The addition of natural zeolite, at doses between 2 and 4 g/l, contributed to enhance the anaerobic digestion process of piggery waste by the increase of the kinetics of COD removal, the rate of methane production and the methane yield. This fact was determined by a reduction of the TVFA/Alk ratio to values favorable for the methanization of the organic matter. The values of the apparent kinetic constant of the process were calculated using a first-order model. The model appears to fit adequately the experimental data. The accumulated methane gas production was affected at zeolite doses of 8 and 10 g/l. At a zeolite concentration of 2 g/l the maximum methane yield was obtained. At concentrations higher than 6 g zeolite/l waste the process became affected and at a dose of 10 g/l a process inhibition appeared. Zeolite contributed to reduce the inhibitory effect of NH_3 and NH_2O by adsorption and of NH_4^+ by ion exchange, but inhibition could not be overcome at doses higher than 6 g zeolite/l waste.

Acknowledgements

The authors wish to express their gratitude to the "Alexander Von Humboldt" Foundation (Germany) and "Consejeria de Educación y Ciencia" of "Junta de Andalucia" (Spain) for financial support.

References

APHA (American Public Health Association), 1989. Standard Methods for the Examination of Water and Wastewater, 17th edn. APHA, Washington, DC, USA.

Borja, R., Banks, C.J., 1993. Semicontinuous anaerobic digestion of soft drink wastewater in immobilized cell bioreactors. Biotechnology Letters 15, 767–772.

Borja, R., Sánchez, E., Weiland, P., Travieso, L., 1993a. Effect of ionic exchanger addition on the anaerobic digestion of cow manure. Environmental Technology 14, 891–896.

Borja, R., Durán, M.M., Martín, A., 1993b. Influence of the support on the kinetics of anaerobic purification of slaughterhouse wastewater. Bioresource Technology 44, 57–60.

Borja, R., Martín, A., Durán, M.M., Barrios, J., 1993c. Influence of clay immobilization supports on the kinetic constants of anaerobic digestion of dairy industry wastewater. Applied Clay Science 7, 367–381.

Borja, R., Martín, A., Luque, M., Durán, M.M., 1993d. Kinetic study of anaerobic digestion of wine distillery wastewater. Process Biochemistry 28, 83–90.

Borja, R., Sánchez, E., Durán, M.M., 1996. Effect of the clay mineral zeolite on ammonia inhibition of anaerobic thermophilic reactors treating cattle manure. Journal of Environmental Science and Health A 31 (2), 479–500.

Grau, P., Dohanyos, M., Chudoba, J., 1974. Kinetics of multicomponent substrate removal by activated sludge. Water Research 9, 637–642.

Hobson, P.N., Shaw, B.G., 1973. The anaerobic digestion of wastes from an intensive pig unit. Water Research 7, 437–449.

Hobson, P.N., Shaw, B.G., 1974. The bacterial population of piggery-waste anaerobic digesters. Water Research 8, 507–516.

Hobson, P.N., 1981. Anaerobic digestion of animal excreta and other agricultural wastes. Tribology Cebedeau 34 (455), 437–441.

Hobson, P.N., 1985. A model of anaerobic bacterial degradation of solid substrates in a batch digester. Agricultural Wastes 14, 255–274.

Hobson, P., 1992. Treatment of animal wastes and uses of treated residues. Biotechnologies for pollution control and energy. In: Proceedings of the Third Workshop of the Working Group on Biogas Production Technologies. CNREE Network on Biomass Production and Conversion for Energy, Braunschweig, Germany, pp. 48–70.

Kimchie, S., Lumbroso, R.E., Geller, Z., Abramovitch, D., Shelef, G., 1988. An integrative treatment process for piggery wastes based on anaerobic digestion. In: Tilche, A., Rozzi, A. (Eds.), Proceeding of the Fifth International Symposium on Anaerobic Digestion. Monduzzi Editore S.p.A., Bologna, Italy, pp. 635–639.

Kugelman, I.J., McCarty, P.L., 1965. Cation toxicity and stimulation in anaerobic digestion. Journal of Water Pollution Control Federation 37, 97–116.

Milán, Z., Sánchez, E., Weiland, P., De las Pozas, C., Borja, R., Mayari, R., Rovirosa, N., 1997. Ammonia removal from anaerobically treated piggery manure by ion exchange in columns packed with homoionic zeolite. The Chemical Engineering Journal 66, 65–71.

Milán, Z., Sánchez, E., Borja, R., Ilangovan, K., Pellón, A., Rovirosa, N., Weiland, P., Escobedo, R., 1999. Deep bed filtration of anaerobic cattle manure effluents with natural zeolite. Journal of Environmental Science and Health B 34 (2), 305–332.

Pérez, J.L., Carretero, M.I., Maqueda, C., 1989. Behavior of sepiolite, vermiculite and montmorillonite as supports in anaerobic digesters. Applied Clay Science 4, 69–82.

Reyes, O., Sánchez, E., Pellón, A., Borja, R., Colmenarejo, M.F., Milán, Z., Cruz, M., 1997. A comparative study of sand and natural zeolite as filtering media in tertiary treatment of wastewaters from tourist areas. Journal of Environmental Science and Health A 32 (6), 1695–1709.

Sánchez, E., Roque, R., 1987. Zeolite as support material in anaerobic wastewater treatment. Biotechnology Letters 9, 671–672.

Sánchez, E., Milán, Z., Borja, R., Weiland, P., Rodriguez, X., 1995. Piggery waste treatment by anaerobic digestion and nutrient removal by ionic exchange. Resources Conservation and Recycling 15, 235–244.

Stotzky, G., Rem, L.T., 1966. Influence of clay minerals on microorganisms I. Montmorillonite and kaolinite on bacteria. Canadian Journal of Microbiology 12, 547–562.

Varstaen, H., 1975. Three step treatment of piggery waste. Water Research 110, 927–931.