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Temperature Conversion (Mesophilic to Thermophilic) of Municipal Sludge Digestion

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A protocol proposed is described here to characterize for the conversion from mesophilic to thermophilic conditions in a pilot-scale anaerobic digester that operates with municipal mixed sludge. Furthermore, the performance of thermophilic operation relative to the previous mesophilic operating status was evaluated. The performance was evaluated in terms of a number of parameters that included organic removal rate (ORR) (kgVS/m^3·d and kgCOD/m^3·d), biogas and volumetric methane production rate (m^3/m^3·d), pH, total acidity (mg acetic acid/L) and acidity/alkalinity relationship. The digester was initially operated with an organic loading rate (OLR) of 1.26 kgVS/m^3·d and a solids retention time (SRT) of 27 days under mesophilic conditions (35°C). The solids destruction efficiency was found to be 54.3%, while the volumetric biogas production in the digester reached 0.36 m^3/m^3·d. The strategy selected for the conversion from mesophilic to thermophilic digestion involved slowly increasing the temperature of the digester (0.38° C/d) until it reached 43°C. In this way, the temperature of the digester was raised from 43 to 45°C and then operated at a constant 45°C. The performance parameters at this temperature indicated that the digester was unstable. For this reason the OLR was decreased until feeding was suppressed. The reactor operated at 45°C for 32 days, and the temperature of the digester was then raised from 45 to 50°C (without feeding). The temperature was subsequently raised to 50 – 52°C with the system operating at variable SRT (65-52 days), and finally, the temperature was increased at a rate of 0.13°C/d until it reached 55°C. At thermophilic conditions (55°C), the OLR studied was 1.48 kgVS/m^3·d (SRT: 27 days), and under these conditions the solids destruction efficiency was 53.3% VS, and the biogas produced in the digester reached 0.32 m^3/m^3·d. *AIChE J,* 51: 2581–2586, 2005
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Introduction

Anaerobic digestion has been, and continues to be, one of the most widely used processes for the stabilization of wastewater treatment plant sludge. The widespread use of this technique over other stabilization processes stems from the potential advantages, which include higher methane production (in excess of that required for operation of the process), a reduction of 30 to 50% of volatile solids requiring ultimate disposal, and a high rate of pathogen destruction. However, conventional anaerobic sludge digestion is associated with several problems and these have limited the application of this technique. A number of processes have been reported for the upgrading of sludge digestion, with thermophilic anaerobic digestion (55°C) being an alternative to mesophilic anaerobic digestion (35°C).[1, 2]

In general, thermophilic anaerobic plants offer attractive kinetic advantages when they are compared with mesophilic and cryophilic systems. The thermophilic anaerobic process

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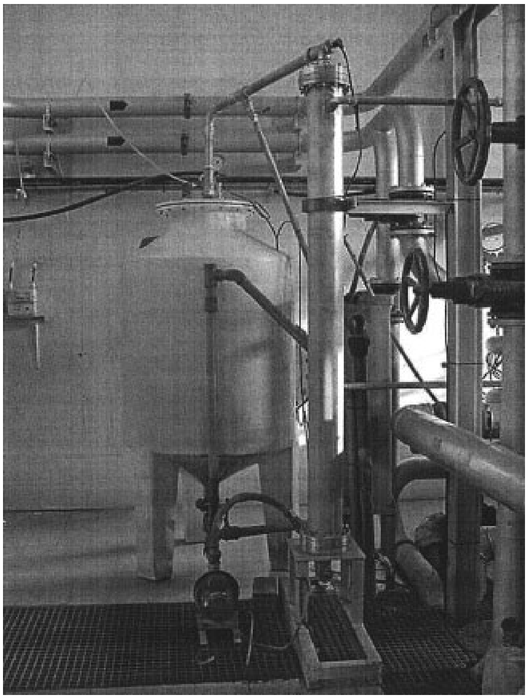

Figure 1. Pilot-plant digester used in this research.

leads to an acceleration of biochemical reactions, and a higher efficiency in terms of the degradation of organic matter in comparison with the mesophilic process.[1,3,4] The efficiency in the destruction of pathogens is also considerably higher.[5,6,7,8] These features are of great importance, because they enable the use of a digester with a smaller volume and/or operation with a higher loading rate.[9] The higher degradation efficiency is associated with a higher level of biogas production and a lower content of volatile solids in the digested sludge, which represents a smaller output of stabilized sludge with better dewatering properties.

Anaerobic digestion of waste requires balanced growth of various populations of microorganisms. The problem of a low thermophilic microbial population can be solved when a thermophilic inoculum is available, for instance from another existing thermophilic plant. However, the startup strategy for growing this inoculum is of extreme importance for success.[10]

Thermophilic digestion is usually attained by adapting a mesophilic system to higher temperatures. This dramatic temperature change probably selects those subpopulations of bacteria that are at low concentrations in the mesophilic system.[10] Thus, the transition from mesophilic to thermophilic temperatures may require a long acclimation period (the startup period for a thermophilic plant is one year or more — until gas production reaches an acceptable level), and may even fail due to the acidic pH and small amount of methane produced. A high sensitivity to temperature changes was observed by van Lier et al.[3] when sludge was adapted for a relatively short period to high temperatures. This phenomenon also occurs during the startup of thermophilic bioreactors if the methanogenic sludge is shifted from mesophilic to thermophilic conditions.[3] However, if the increase in the process temperature is gradual, the stability of the methanogenesis stage and the improvement in the thermophilic bacteria are much higher.[7,11]

This work described here concerns the protocol selected for conversion from mesophilic to thermophilic operation in a municipal sludge digester. The operating characteristics of the digester and its performance under thermophilic conditions relative to its previous mesophilic operating mode are also described.

Materials and Methods

Description of pilot digester.

The continuously stirred-tank reactor (CSTR) employed in this study had an operational volume of 0.15 m^3. A picture of the pilot-scale digester is shown in Figure 1. The desired temperature was maintained by recirculating temperature-controlled water through an internal coil. The reactor was inoculated with mesophilic sludge obtained from a full-scale 35°C continuously stirred-tank reactor (Guadalete WWTP, Jerez de la Frontera-Cadiz, Spain).

The characteristics of the prethickened combined primary and secondary waste sludge (feed) used during this study are presented in Table I (maximum, minimum and mean value of each parameter), and these values are typical values for raw sludge.

A certain volume (depending on the solid retention time imposed) of digested sludge was withdrawn from the reactor three times per day, and an equal volume of raw sludge was pumped into the recycle line of the digester.

The effluent from the digester was recycled through a variable-speed centrifugal pump in order to provide pseudo-completely mixed conditions in the liquid phase. Recycle flow was drawn from the bottom of the reactor and pumped to the top of the reactor in order to maintain mixed conditions in the digester.

Sampling and analysis.

The parameters measured were as follows: chemical oxygen demand (COD), total solids (TS_0, TS_e) and volatile solids (VS_0, VS_e) of influent and effluent, pH, individual volatile fatty acids levels (VFAs) and bicarbonate alkalinity of effluent, and gas production and composition of gas (methane and carbon dioxide percentages). All analytical determinations were performed according to "standard methods".[12]

The volume of gas produced in the reactor was directly measured by a mass-flow sensor and gas composition (methane and carbon dioxide) was determined by gas chromatography (SHIMADZU GC-14 B) using a stainless steel column packed with Carbosieve SII (1/8 in. dia. and 2 m length), and a thermal conductivity detector. The injected sample volume was 1 mL and operational conditions, were as follows: 7 min at 55°C; ramped at 27°C/min to 150°C; detector temperature: 255°C; injector temperature: 100°C. The carrier gas was helium and

Table 1. Main Characteristics of Raw Sludge

Parameter	Maximum Value	Minimum Value	Mean Value
COD (g/L)	102.2	36.0	71.7
pH	6.4	5.8	6.1
Total Solids (g/L)	78.6	30.2	55.0
% Volatile Solids	73.5	51.9	67.8
Volatile Solids (g/L)	46.5	22.2	39.0
Alkalinity (mg/L)	6000	4047	4870
Volatile acids (mg/L)	2112	607	1354

Table 2. Summary of Conditions During the Temperature Conversion

Stage	Stage Duration, Days	Feed	Temp. Increase Rate °C/d
Mesophilic, 35°C	90	Yes	—
Increasing 35 to 43°C	21	Yes	0.38
Maintenance at 45°C	32	Yes/no	—
Increasing 45 to 50°C	11	No	0.45
Maintenance at 50–52°C	42	Yes	—
Maintenance at 55°C	23	Yes	0.13
Thermophilic, 55°C	90	Yes	—

the flow rate used was 30 mL/min. A standard gas (from Carburos Metálicos, S.A; composition: 4.65% H_2; 5.33% N_2; 69.92% CH_4 and 20.10% CO_2) was used for calibration of the system.

Concentrations of individual VFA levels in the effluent were determined using a gas chromatograph (SHIMADZU GC-17 A) equipped with a flame-ionisation detector, and a glass column filled with Nukol (poly(ethylene glycol) modified by nitroterephthalic acid). Standard calibration solutions for each VFA were used. The temperatures of the injection port and detector were 200 and 250°C, respectively. Helium was the carrier gas at 50 mL/min. In addition, nitrogen gas was used at 30 mL/min flow rate.

Results and Discussion

Reactor operation.

Initially the digester was fed with raw sludge, as described in the Materials and Methods section and Table 1, at strength of 34.0 kg/m^3 to give an organic loading rate (OLR) of 1.26 kgVS/m^3·d or 2.47 kgCOD/m^3·d, with an SRT of 27 days.

The experimental study required 44 weeks of experimentation, data collection and analysis of the input and output sludge.

Steady-state conditions were reached for 27 days SRT at both mesophilic and thermophilic conditions in order to analyze and compare the results obtained. The attainment of the steady state was verified after an initial period (3 times the HRT) by checking the performance parameter values.

A summary of the conditions during the temperature conversion of municipal sludge digestion is given in Table 2.

Performance and operation parameters for the evolution of the process during the stable mesophilic and thermophilic operation, as well as during the transition from mesophilic to thermophilic temperature, are shown in Table 3. This table shows, for each parameter, the mean value and final value as the average of the last three data points in each stage.

The individual VFA levels (acetic, propionic, n-butyric acids and total VFA, as mg/L) in the effluent of the reactor from mesophilic and thermophilic conditions are shown in Table 4.

Mesophilic process startup.

The startup and development of the mesophilic digester began on April 4, 2000. For the first 13 weeks, the digester was maintained at mesophilic temperature and 27 days SRT until reaching stable operation.

Under these conditions (organic loading rate 1.26 kgVS/m^3·d or 2.47 kgCOD/m^3·d), the volatile solids destruction efficiency and organic removal efficiency were 54 and 53%, respectively, and the volumetric methane production rate was 0.36 – 0.38 m^3 CH_4/m^3·d. The operational pH was maintained at 7.5, and the acidity/alkalinity ratio was 0.09.

The volatile acetic acid concentrations were always in the range 200 – 600 mg acetic acid/L, with a mean value of 515 mg acetic acid/L and a total VFA concentration of approximately 876 mg acetic acid/L at steady state conditions, as can be observed from Table 4.

Mesophilic-thermophilic conversion process.

The strategy initially selected to change from mesophilic to thermophilic digestion involved a slow, gradual temperature increase designed after a method employed by Rimkus et al. in Chicago,[7] and Peddie et al. in Vancouver.[8] An alternative approach would have been to rapidly heat the digester to thermophilic temperatures and hold the temperature until a stable thermophilic regime established itself. However, this strategy was attempted by Peddie et al.[8] in Vancouver, and the digestion stopped as the mesophilic microorganism became deactivated and the thermophilic microorganism were building up.

The procedure attempted by Garber et al.[5] to obtain a thermophilic culture was to raise the temperature of the digester at a rate of 0.6°C/d until the desired temperature was reached. This method, however, "soured" the digester. In an effort to overcome this difficulty, the temperature of the digester in this

Table 3. Summary of Performance Parameter Data for Digester during the Conversion at Each Temperature

Stage	Value	T, °C	SRT Days	OLR kgVS/m^3 · d	ORR kgVS/m^3 · d	%VS$_r$	OLR kgCOD/m^3 · d	ORR KgCOD/m^3 · d	%COD$_r$	pH	Biogas m^3/m^3 · d	Ac./Alk.*	Acetic Acid mg/L	Total Acidity mg Acetic Acid/L
Mesophilic, 35°C	Mean value	35.2	27	1.26	0.67	54.3	2.47	1.37	52.8	7.5	0.36	0.09	515	830
	Final value	35.2	27	1.22	0.69	55.0	2.39	1.36	56.1	7.5	0.38	0.09	625	876
Increasing 35 to 43°C	Mean value	41.4	27	1.30	0.75	56.6	2.70	1.55	56.8	7.5	0.14	0.23	1873	4800
	Final value	43.7	27	1.17	0.58	49.2	2.74	1.49	53.9	7.2	0.12	0.46	3936	5456
Maintenance at 45°C	Mean value	45.3	92	1.06	0.57	54.6	1.94	1.02	48.7	7.3	0.06	0.46	4194	5824
	Final value	45.8	—	—	—	—	—	—	—	7.7	0.07	0.46	3363	5851
Increasing 45 to 50°C	Mean value	48.3	—	—	—	—	—	—	—	7.9	0.12	0.44	6213	9440
	Final value	49.5	—	—	—	—	—	—	—	7.7	0.16	0.42	4977	8015
Maintenance at 50–52°C	Mean value	51.4	65	0.58	0.36	62.7	1.02	0.58	54.1	7.7	0.11	0.20	1588	4559
	Final value	54.5	52	0.81	0.61	74.9	1.41	0.91	63.6	7.7	0.10	0.30	1613	4503
Maintenance at 55°C	Mean value	54.7	54	0.70	0.36	53.7	1.48	0.81	50.8	7.6	0.14	0.39	2603	5420
	Final value	54.9	52	0.69	0.37	56.3	1.45	0.79	51.5	7.7	0.16	0.45	3772	6583
Thermophilic, 55°C	Mean value	55.0	27	1.48	0.76	53.3	2.48	0.96	35.6	7.8	0.32	0.25	566	3163
	Final value	55.0	27	1.49	0.84	53.5	2.43	0.78	31.9	7.8	0.32	0.24	489	2950

Acidity to alcalinity ratio.[12]

Table 4. Individual VFA Levels (acetate, propionate, n-butyric and Total Acidity, as mg Acetic acid/L) in the Effluent of the Reactor at Mesophilic and Thermophilic Conditions

Stage	Value	T°C	Acetic Acid	Propionic Acid	n-Butyric + n-butyric Acids	Total Acidity mg Acetic Acid/L
Mesophilic	Mean value	35.2	515.4	103.3	115.8	829.9
35°C	Final value	35.2	625.5	111.6	114.0	875.7
Thermophilic	Mean value	54.7	566.2	1400.6	209.1	3163.3
55°C	Final value	54.9	489.2	1366.6	307.1	2950.5

study was increased at a much slower rate than 0.6°C/d. Furthermore, the digester was operated at a variable detention time during the temperature adjustment, as shown in Table 3, in order to maintain the stability of the digestion process.

Once the mesophilic conditions had been reached, the temperature of the digester was raised in 21 days from 35 to 43°C (rate of 0.38°C/d). The SRT was maintained at 27 days. During that period the mesophilic microorganisms were active. Hence, the ORR was 0.75 kgVS/m^3·d (1.55 kgCOD/m^3·d), and the volatile solids destruction efficiency and organic removal efficiency were 56.6% and 56.8%, respectively. The biogas generation decreased until it reached 0.12 m^3/m^3·d at the end of the stage. At this time, the volatile fatty acid content increased sharply from 875.7 mg acetic acid/L to about 5455.6 mg acetic acid/L. The acidity/alkalinity ratio was 0.46 and the pH was 7.2.

At this stage the experiment suffered from problems associated with maintaining the pH values within the operational level (in the approximate range 6.5 – 8.5). Adjustment of the pH of the feedstock with alkali (sodium hydroxide) was found to be sufficient to stabilize the operational pH in the range 7.2 – 7.5.

The temperature of the digester was then raised from 43 to 45°C, and the system was operated at 45°C during 32 days. SRT was gradually increased until feeding was suppressed due to the fact that all operating parameters indicated that the digester was unstable. The volatile fatty acid content increased to approximately 5800 mg acetic acid/L and the acidity/alkalinity ratio was 0.46. The gas generation decreased and the production at 45°C was 0.07m^3/m^3·d at the end of this stage.

The temperature was raised from 45 to 50°C (without feeding) during 11 days. The biogas generation rate increased from 0.07 to 0.16m^3/m^3·d, but the system reached higher VFA concentrations (9440 mg/L). An example in the literature indicates that the temperature range between 43 and 50°C would be unstable.[8]

The temperature of the system was maintained at 50 – 52°C for the next 42 days. The imposed SRT was 65 days, and this was decreased to 52 days at the end of this stage. The volatile acid content decreased from 5,800 to 4,500 mg acetic acid/L. During the preceeding weeks, the acidity/alkalinity ratio was about 0.2 – 0.3. However, gas production was reduced during the preceding weeks (0.11 – 0.10 m^3/m^3·d).

The upper temperature limit was 55°C, which is the optimal value for thermophilic digestion. This temperature adjustment was started after 197 days of the experiment and was changed at a rate of 0.13°C/d. At this point, the ORR was 0.36 kgVS/m^3·d at SRT: 54 days. The volatile solids destruction efficiency and organic removal efficiency were 54 and 51%, respectively. The biogas generation increased until it reached a value of 0.16 m^3/m^3·d. The volatile fatty acid content increased to about 6,500 mg acetic acid/L at the end of this period.

The acidity/alkalinity ratio reached 0.39 and the pH reached 7.7.

As can be observed, the acetic acid level increased between 1873 and 4977 mg/L as the temperature increased from 35 to 50°C and then dropped sharply at 55°C (2603 mg/L). VFA concentration is usually higher in thermophilic digestion. This situation has been observed by several authors in systems operating with anaerobic digestion thermophilic sludge.[13,14] Reusser and Zelinka[15] report that, after operating at 15 days SRT, the VFA concentration was 2,558 mg of acetic acid/L.

It is generally accepted that the experimental start-up period for a stable thermophilic operation is long, that is, up to one year, before gas production reaches an acceptable level.[16]

Operation under thermophilic conditions (27 days SRT).

The experimental protocol was defined to examine the effect of increasing the organic loading rate on the efficiency of the digester, and to report on its steady-state performance at 27 days SRT.[17] The attainment of the steady state was verified after an initial period (three times the SRT) by checking whether constant effluent characteristic values had been attained (VS destruction, COD removal, volumetric methane production rate and individual VFA levels). Sodium carbonate was added at concentration of 2N to maintain the digestion at the optimum pH for anaerobic thermophilic digestion.

At 55°C and 27 SRT, the organic loading rate was 1.48 kgVS/m^3·d or 2.48 kgCOD/m^3·d. Under these conditions, the volatile solids destruction efficiency and organic removal efficiency were 53.3% and 35.6%, respectively, and the volumetric methane production rate was 0.32 m^3CH_4/m^3·d.

The individual volatile acid concentrations (Table 4) were in the range 400 – 600 mg acetic acid/L and the propionic acid concentration was very high, at approximately 1400 mg/L. The total VFA concentration was approximately 3000 mg acetic acid/L. The pH was maintained at 7.8, and the acidity/alkalinity ratio decreased until it stabilized to constant values in the range 0.25 – 0.30 mg acetic acid/mg calcium carbonate (very high for operation under thermophilic conditions).

Comparison of mesophilic and thermophilic processes.

The mesophilic and thermophilic performance parameters of the digester at 27 days SRT are shown in Table 3. As can be seen, the efficiency of both systems was the same (53% volatile solids reduction). On an industrial scale and under mesophilic conditions, 40% volatile solids reduction is an acceptable value in the performance of a sludge digestion process.[17]

The COD removal of the mesophilic sludge was higher than that of the thermophilic sludge at 27 days solids retention time (52.8% COD_r vs. 35.6%COD_r), and this is due to the high VFA concentrations in the effluent. Biogas generation was always similar under the two sets of conditions — although it was only marginally higher than thermophilic production (0.32 m^3/m^3·d vs. 0.36 m^3/m^3·d). In both digesters, the methane fraction average was close to 60% and fell in the range between 57.7% and 64.5%.

It has often been suggested that operation at thermophilic temperatures and low SRT will provide better breakdown of organic material and thus generate more methane.[6] However, in the study reported here, at SRT 27 days, the rate of volumetric methane production rate in the mesophilic digester was higher than that found in the thermophilic unit (0.25 m^3/m^3·d vs. 0.19 m^3/m^3·d). Similarly, methane production activity (as m^3 of methane produced per gram of VS destruction) of the mesophilic system was clearly superior to that in the thermophilic digester (0.40 $m^3CH_4/kgVS_r$ and 0.27 m^3CH_4/m^3VS_r, respectively). Acetate will be the main precursor of methane in both mesophilic and thermophilic systems, and this material accounts for approximately 65 – 80% of the methane produced, with hydrogen/carbon dioxide making up the rest.[18]

The performance of both processes based on these traditional parameters was normal. One of the major criticisms concerning the use of thermophilic digestion is that the final effluents contain higher concentrations of volatile fatty acids than those from mesophilic digesters. The data here support this trend (at 27 days SRT, total acidity was approximately 3000 mg/L under thermophilic conditions vs. 830 mg/L under mesophilic conditions). As can be seen from the results in Table 4, the individual mesophilic volatile acid concentrations were always in the range 100 – 600 mg/L. Under thermophilic conditions, the propionic acid concentration was approximately 1400 mg/L vs. 100 mg/L under mesophilic conditions. Overall, the individual VFA levels were consistently lower than those in the thermophilic unit. High levels of total VFA in thermophilic processes have also been reported by Dinsdale et al.[19]. These authors operated continuous thermophilic studies on coffee waste for long periods and achieved stable digestion at a variety of loading rates. The activity of thermophilic bacteria was higher than that of mesophilic bacteria; however, thermophilic bacteria tend to remove propionic acid more slowly than mesophilic bacteria.[20]

The pH of the thermophilic sludge was kept at a constant level in the range 7.6 – 7.8, while the pH of the mesophilic sludge was maintained above 7.5. Thermophilic bicarbonate alkalinity was also slightly higher than the mesophilic alkalinity (14000 vs. 12500 mg $CaCO_3$/L). The high alkalinity level indicates that the various bacterial groups are in balance. The system is well buffered under both sets of conditions.

The mesophilic acidity/alkalinity ratio was very low (0.09 vs. 0.25 under thermophilic conditions).

Conclusions

The results of this research confirm that the transition from a mesophilic digester to a thermophilic system is possible if temperature changes are made carefully. The experimental protocol proposed for conversion from mesophilic (35°C) to thermophilic (55°C) operation in a municipal sludge digester is as follows:

1. Increase the temperature slowly from 35 to 43°C in 2.5°C increments and maintain each new temperature for at least 4 days while operating at the same organic loading speed as used in the optimum mesophilic operating conditions (that is, at 35°C).
2. Increase the temperature straight form 43 to 50°C, without feeding the system, until degradative activity is observed in the digester. Carry out and stabilize the same organic feed rate as in the previous stage.
3. Once again increase the temperature of the system in maximum increments of 2.5° C, leaving the system to stabilize after each temperature increase, until a final temperature of 55 °C is reached. During this process the same organic loading rate as in previous stages should be used.

As far as the performance of both mesophilic and thermophilic processes operating at 27 days retention time are concerned, the experimental results indicate that the mesophilic ORR was 0.67 $kgVS/m^3$·d (1.37 $kgCOD/m^3$·d), and the methane yield was 0.40 $m^3CH_4/kgVS_r$. Under thermophilic conditions, the ORR was 0.76 $kgVS/m^3$·d (0.96 $kgCOD/m^3$·d) and the methane yield was 0.27 $m^3CH_4/kgVS_r$.

However, the results obtained indicate that there is no advantage in operation at anaerobic thermophilic temperatures when the SRT was 27 days (1.3 $kgVS/m^3$·days), as indicated by volatile solids breakdown and gas production.

Further experiments are required in this area to determine the optimal operating parameters for the thermophilic process.

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Literature Cited

1. Zabranska J, Dohanyos M, Jenicek P, Kutil J. Thermophilic process and enhancement of excess activated sludge degradability-two ways of intensification of sludge treatment in Prague central wastewater treatment plant. *Water Sci Technol*. 2000;41:265-272.
2. Ahn JH, Forster CF. A comparison of mesophilic and thermophilic anaerobic upflow filters. *Bioresource Technol*. 2000;73:201-205.
3. van Lier JB, Hulsbeek J, Stams AJM, Lettinga G. Temperature susceptibility of thermophilic methanogenic sludge: implications for reactor start-up and operation. *Bioresource Technol*. 1993;43:227-235.
4. Pérez M, Romero LI, Sales D. Steady state anaerobic thermophilic degradation of distillery wastewater in fluidized bed bioreactors. *Biotechnol Progress*. 1997;13:33-38.
5. Garber W, Ohara G, Colbaugh J, Raksit G. Thermophilic digestion at the Hyperion Treatment Plant. *J Water Poll Cont Fed*. 1975;47:950-961.
6. Smart J, Boyko BI. Full scale studies on the thermophilic anaerobic process. Research program for the abatement of municipal pollution under the provisions of the Canada-Ontario; 1978; Project n° 73-129.
7. Rimkus R, Ryan J, Cook E. Full scale thermophilic digestion at the west-southwest sewage treatment works, Chicago, Illinois. *J Water Poll Cont Fed*. 1982;54:1447-1457.
8. Peddie CC, Tailford J, Hoffman D. Thermophilic anaerobic sludge digestion-taking a new look at an old process. *10th Annual Residuals Biosolids Management Conference*. 1996;1:39-46.
9. Vázquez E, Mejía GM. Producción de fangos en la depuración anaerobia. *Tecnología del Agua*. 1996;148:137-141.
10. Zhang TC, Noike T. Influence of retention time on reactor performance and bacterial trophic populations in anaerobic digestion processes. *Water Res*. 1994;28:27-36.
11. Garber W. Operating experience with thermophilic anaerobic digestion. *J Water Poll Cont Fed.* 1982;54:1170-1175.
12. APHA (American Public Health Association, American Water Works Association, Water Pollution Control Federation). 17th ed. Standard

Methods for the examination of water and wastewater. Rhodes Trussell eds; Washington; 1989.
13. Moen G, Stensel HD, Lepistö R, Ferguson J. Effect of solids retention time on the performance of thermophilic and mesophilic digestion. *Wat Environ Res*. 2003;75:539-548.
14. Song YC, Kwon SJ, Woo JH. Mesophilic and thermophilic temperature co-phase anaerobic digestion compared with single-stage mesophilic- and thermophilic digestion of sewage sludge. *Water Res*. 2004; 38:1653-1662.
15. Reusser S, Zelinka G. (2001). Lab-scale comparison of anaerobic digestion alternatives. WEFTEC; 2001.
16. Ahring BK. Status on science and application of Thermophilic anaerobic digestion. *Water Sci and Technol*. 1994;30:241-249.
17. de la Rubia MA. Puesta en marcha y optimización de la degradación anaerobia termofílica de lodos de EDAR. Serv. Pub. Univ. Cádiz. ISBN: 84-7786-919-7. Spain.; 2004. PhD Thesis.
18. Tchobanoglous G, Burton FL. Metcalf & Eddy Wastewater Engineering: *Treatment, Disposal and Reuse*, McGraw-Hill, Inc; New York;1995.
19. Dinsdale RM, Hawkes FR, Hawkes DL. The mesophilic and thermophilic anaerobic digestion of coffee waste containing coffee grounds. *Water Res.*1996;30:371-377.
20. Mackie UI, Bryant MP. Metabolic activity of fatty acid-oxidizing bacteria and the contribution of acetate, propionate, butyrate, and CO_2 to methanogenesis in cattle waste at 40 and 60°C. *Appl Environ Microbiol*. 1981;41:1363-1373.

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