CHEMICAL ENGINEERING TRANSACTIONS
VOL. 32, 2013
A publication of AIDIC
The Italian Association of Chemical Engineering
Online at: www.aidic.it/cet
Chief Editors: Sauro Pierucci, Jiří J. Klemeš

ISBN 978-88-95608-23-5; **ISSN** 1974-9791
DOI: 10.3303/CET1332167

Dark Fermentation Optimization by Anaerobic Digested Sludge Recirculation: Effects on Hydrogen Production

Marco Gottardo*[a], Cristina Cavinato [a], David Bolzonella[b], Paolo Pavan [a]

[a]University Ca'Foscari of Venice, Department of Environmental Science, Informatics and Statistics, Calle Larga Santa Marta, Dorsoduro 2137 – 30123, Venice, Italy
[b]University of Verona, Department of Biotechnology, Strada le Grazie 15 – 37134 37134, Verona, Italy.
marco.gottardo@unive.it

The addition of small amounts (5 – 10 %) of H_2 to rich CH_4 biogas improves the quality of gas combustion while reducing CO_2 emissions. For this reason, many researchers have recently been focused to optimize the two phase anaerobic digestion process (dark fermentation, DF and anaerobic digestion, AD) to concurrently produce H_2 and CH_4 from biowaste and organic residues. In this paper the results of a two phases thermophilic AD process treating biowaste for H_2 and CH_4 production are presented: in this process, nor physical neither chemical pre-treatment of inoculum, was used, but recirculation of anaerobic digested sludge to the DF reactor was exploited to control the pH in the optimal H_2 production range of 5-6. The experiment was carried out in bench scale using a stirred reactor (CSTR), applying an organic loading rate of 16 KgTVS/m^3d, and an hydraulic retention time of 3 days in DF phase. Four different recirculation conditions were tested keeping constant the ammonia concentration (about 500 mg/L) through a separation process by evaporation. The aim was to investigate the influence of ammonia in the biological process of hydrogen production via dark fermentation and to research the recirculation condition that allow to reach the best yields and a stable process. The optimal hydrogen production (SHP of 0.03 m^3/Kg TVS fed, 30 % H_2 in the off – gas) was found using a recirculated sludge ratio of 0.66. This allowed for a stable pH value around 5.5.

1. Introduction

Since the introduction of anaerobic digestion (AD) of biowaste in the beginning of the nineties, the adoption of the technology has consistently grown. Considering that twenty years ago only a handful of digesters were running on biowaste or municipal solid waste and that at the end of 2010 almost 200 plants were running in Europe, one can not dispute that AD is a mature technology. A step forward of the common anaerobic digestion process of biowaste, that has gained interest among the researchers, is the two - stage approach finalized to the production of hydrogen in the first phase reactor and methane in the second one (Martinez – Perez et al., 2007). Obtained gases can be used separately or mixed together, with an average percentage composition of 10 % H_2, 30 % CO_2 and 60 % of CH_4, to obtain a second generation biofuel that can be of great interest in the transportation sector. Studies conducted by Porpatham et al. (2007) found that adding 10 % of hydrogen in biogas improves the efficiency in the combustion, with a consequent improvement in thermal efficiency and power output. Moreover, a drastic reduction of hydrocarbons (HC) emissions and no significant increase in NO level was observed. The hydrogen production in first phase reactor occurs by fermentation of carbohydrate-rich substrates, and is carried out by anaerobic bacteria belonging to the species such as Enterobacter, Bacillus and Clostridium. This fermentative process, named dark fermentation, produces a mixed biogas containing primarily hydrogen and carbon dioxide (Levin et al., 2004); to optimize the hydrogen production by dark fermentation is necessary to optimize the activity of enzyme hydrogenase: recent studies have shown that to maximize the yield of hydrogen production the pH should be maintained in the range 5 - 6.5, with an optimum value at 5.5 (Valdez – Vazquez et al., 2009). To buffer the pH in the first phase reactor, some authors (e.g. Antonopoulou et al., 2008) have used chemicals like sodium hydroxide and potassium

Please cite this article as: Gottardo M., Cavinato C., Bolzonella D., Pavan P., 2013, Dark fermentation optimization by anaerobic digested sludge recirculation: effects on hydrogen production, Chemical Engineering Transactions, 32, 997-1002 DOI: 10.3303/CET1332167

hydroxide, while other authors (e.g. Lee et al., 2010) recirculated the second phase sludge to support the dark – fermentation with buffering agents. Recent studies carried out by Cavinato et al. (2011) at pilot scale have demonstrated the possibility to produce hydrogen in a two-phase thermophilic anaerobic digestion without the use of external pH control, but using the recirculation of the liquid fraction of anaerobic digestion effluent: the highest hydrogen yield (about 51 LH_2/kgTVS fed) was achieved at 16 kgTVS/m^3d of organic loading rate (OLR), a hydraulic retention time (HRT) of 3.3 for dark fermentation. The production of hydrogen by dark fermentation of biowaste without the use of external pH control was tested in a long term run too; the results of this work have evidenced that the hydrogen production rate decrease in correspondence with the ammonia concentration increase in the system, due to the action of sludge recirculation (Cavinato et al., 2012). Starting from this evidence, the aim of this work was to individuate the sludge recirculation ratio that allows to keep a stable biological process while maximizing the hydrogen yield.

2. Material and Method

2.1 Experimental set – up

A stirred reactor (CSTR) with 4.5 L of working volume was used for the experiment; the reactor was heated by hot water recirculation system and maintained at 55 °C. The feeding system was semi – continuous, arranged once per day. The organic waste was reduced in size using a grinder, mixed with tap water and liquid fraction of sludge recirculation from a pilot digester and then fed to the reactor. The experimental test was divided in four periods (runs) distinct from each other to different sludge recirculation ratio: in the first run was used a sludge recirculation ratio of 0.33, in the second run the sludge recirculation ratio was 0.42, in third run the sludge recirculation ratio was 0.66 and then in the fourth run was used a sludge recirculation ratio of 1. The OLR and HRT were maintained in all the runs at about 16 KgTVS/m^3d and 3 d, respectively. Table 1 shows the operational conditions applied to the reactor during the experimental test.

Table 1: Operational conditions applied during the experimental test

		Run I	Run II	Run III	Run IV
HRT	d	3	3	3	3
OLR	Kg/m^3d	16	16	16	16
Sludge recirculation ratio (α)		0.33	0.42	0.66	1

The recirculated sludge used was collected from a pilot digester and then, before of its use, treated with separation process by evaporation unit in order to maintain the ammonia concentration to about 500 mg/L. The evaporation process was performed by means of pilot vacuum heat pump evaporator EVALED R150v3 Veolia Water.

2.2 Substrate and Inoculum

The reactor was inoculated with biowaste and water and then regularly fed with separately collected organic biowaste coming from the municipality of Treviso, and with sludge recirculated and water in order to reach the required volume. The typical composition of the collected waste is shown in table 2.

Table 2: Composition of treated biowaste (WW = Wet Weight; DW = Dry Weight)

Waste Class	% WW	% DW
Fruit and vegetable	38 – 46	30 – 38
Other food waste	13 – 16	12 – 19
Paper and cardboard	13 – 18	15 – 19
Plastic	5 – 10	7 – 14
Inerts	3 – 9	14 – 19
Unclassified materials	10 – 20	13 – 25

As shown in table 2, the food waste collected has a relevant content of fruit and vegetables (38 – 46 % on wet weight basis), and a content of other food waste (such as meat, pasta, etc.) of 13 – 16 % on wet weight.

2.3 Analytical methods

The effluent of the reactor was monitored 2/3 times per week in terms of total and volatile solids content, chemical oxygen demand, TKN and total phosphorus. The process stability parameters, namely pH, volatile fatty acid content and speciation, total and partial alkalinity and ammonia, were checked daily. All the analyses, except for VFAs, were carried out in accordance with the Standard Methods (APHA–AWWA–WEF). Volatile fatty acids content was monitored using a gas chromatograph (Carlo Erba instruments) with hydrogen as gas carrier, equipped with a Fused Silica Capillary Column (Supelco NUKOLTM, 15 x 0.53 x 0.5 µm film thickness) and with a flame ionization detector (200 °C). The temperature during the analysis started from 80 °C and reaches 200 °C trough two other steps at 140 and 160 °C, with a rate of 10 °C/min. The analyzed samples were centrifuged and filtrated on a 0.45 µm membrane. Gas production was monitored continuously by a gas flow meter (Ritter Company, drum-type wet-test volumetric gas meters), while the hydrogen content was measured by a gas-chromatograph (GC Agilent Technology 6890 N) equipped with the column HP-PLOT MOLESIEVE, 30 x 0.53 mm ID x 25 um film, using a thermal conductivity detector and argon as gas carrier.

3. Results and discussion

The main chemical – physical characteristics of the food waste and anaerobic sludge after treatment with evaporator unit, used as fed in four Runs, are reported in table 3 and 4.

Table 3: Characterization of the food waste after pre-treatment step

Parameters	Units	Average	SD	Max	Min
TS	g/Kg_{WW}	238	29.3	323.8	232.6
TVS	g/Kg_{WW}	233.6	21.9	261.3	191.3
COD	g/Kg_{WW}	945.8	150.4	1,126.5	658.6
TKN	g/Kg_{WW}	19.4	9.2	29.8	8.5
P_{TOT}	g/Kg_{WW}	7.4	3	11.1	3.4

Table 4: Characterization of anaerobic sludge after treatment with evaporator unit

Parameters	Units	Average	SD	Max	Min
TS	g/Kg	12.6	3.8	19.3	5.3
TVS	g/Kg	7.3	2.8	12.5	2.6
COD	g/Kg_{WW}	684.8	110.8	876.1	509.4
TKN	g/Kg_{WW}	40.2	3.2	43.3	34.4
P_{TOT}	g/Kg_{WW}	10.2	2	15.2	8.3
pH		8.5	0.3	9.1	8
VFA	mgCOD/L	331.6	230.1	852.8	0
Alkalinity (pH = 6)	$mgCaCO_3/L$	2,162.3	312.6	2,754.5	1,646.7
Alkalinity (pH = 4)	$mgCaCO_3/L$	3,236.9	485.7	3,920.4	2,335.3
NH_4^+	$mgN\text{-}NH_4^+/L$	562.8	60.6	674.6	422.1

As shown in table 4, the use of the evaporation unit allowed for a good control of the ammonia concentration around the value of 500 $mgN\text{-}NH_4^+/L$.

Among the four conditions applied the Run I and Run IV give the worst results with a not significant hydrogen production as described below. As mentioned above pH is an important parameter involved in the biohydrogen generation process: to optimize the activity of enzyme hydrogenase is necessary to maintain the pH in range between 5 and 6.5. During Run I (α = 0.33), pH in the first phase reactor was maintained in range between 4.2 and 4.6; in correspondence of these values, it was not observed (a considerable) H_2 production. This low pH value could be explained by the low alkalinity in the system, that reached a maximum of about 1,100 $mgCaCO_3/L$ and then stabilized at 800 $mgCaCO_3/L$. This value of alkalinity was not sufficient to buffer the acids produced from fermentative process (like VFA , that reached a value of 6 gCOD/L).

During Run IV (α = 1), pH system remained in range between 6.7 and 7 after seven days. In correspondence of these conditions, was not observed hydrogen production, instead was observed a small production of methane.

The other two runs, II and III, working with sludge recirculation ratio of 0.42 and 0.66 respectively, showed an interesting H_2 production; the specific hydrogen production, for the Run II and III is showed in figure 1.

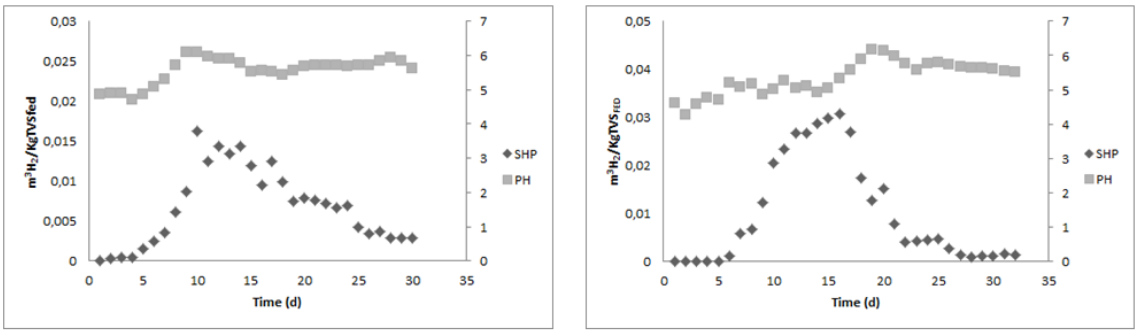

Figure 1: Specific Hydrogen Production and pH in Run II (A) and Run III (B)

The trend of the hydrogen production, expressed as specific hydrogen production (SHP), in both of these runs, showed an initial low hydrogen production, due to the lag time, then a fast increase until a maximum value (0.016 m^3H_2/KgTVS fed in Run II and 0.03 m^3H_2/KgTVS fed in Run III); after few days the production decreased.
The SHP versus ammonia concentration for Runs II and III is shown in figure 2, and it is possible to observe the occurrence of inhibition action in the biological process. In literature some studies shown the inhibitory effect of ammonia concentration on the fermentative hydrogen production (e.g. Cavinato et al., 2012).

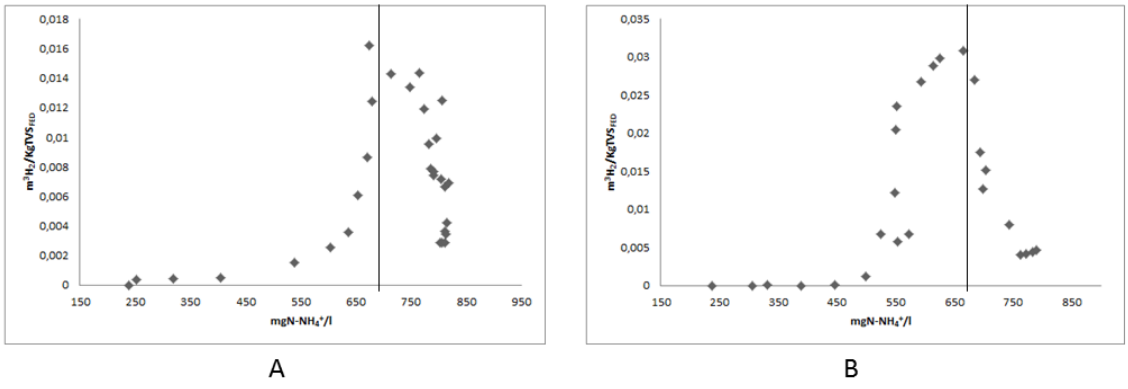

Figure 2: Specific Hydrogen Production versus ammonia concentration in Run II (A) and Run III (B)

The hydrogen yields increased in both Runs with the increasing of ammonia concentration until the same threshold value (about 670 mgN-NH_4^+/L), beyond which it decrease sharply. The increase of ammonia concentration, until a value close to 670 mgN-NH_4^+/L, allows an increase in the hydrogen production as it support the dark fermentation with alkalinity, but exceeded this value the ammonia could act an inhibitor of the biological process of hydrogen production.
In table 5, the characterization of effluents during Run II and III, and the corresponding gas yields, in the condition of maximum hydrogen production, are shown; in this table are also reported the total volatile fatty acid concentrations (VFA) and particularly acetic and butyric acid concentration, since they are the predominant by – products generated.

Table 5: Characterization of effluent and gas yields of Run II and Run III

Parameters	Units	Run II	Run III
Characterization of effluent			
TS	g/Kg	71 ± 0.4	44 ± 4
TVS	g/Kg	48 ± 0.2	33 ± 4
COD	g/Kg_{WW}	67 ± 0.3	46 ± 2
TKN	g/Kg_{WW}	2 ± 0.1	1.1 ± 0.1
P_{TOT}	g/Kg_{WW}	1 ± 0.1	0.22 ± 0.02
pH		5.8 ± 0.2	5.5 ± 0.2
VFA	mgCOD/L	11,156 ± 1,372	15,352 ± 1,395
Acetic Acid	mgCOD/L	1,066 ± 100	3,344 ± 696
Butyric Acid	mgCOD/L	3,442 ± 496	5,747 ± 996
Alkalinity (pH = 4)	$mgCaCO_3/L$	4,768 ± 135	3,483 ± 463
NH_4^+	$mgN\text{-}NH_4^+/L$	677 ± 3	675 ± 14
Gas yields			
Gas Production (GP)	L/d	2.4 ± 1.5	3 ± 0.2
Specific Gas Production (SGP)	$m^3_{biogas}/KgTVS_{fed}$	0.033 ± 0.01	0.094 ± 0.05
Gas Production Rate (GPR)	$m^3_{biogas}/m^3_{reactor}d$	0.48 ± 0.03	0.84 ± 0.06
H_2	%	35 ± 8	40 ± 10
Specific Hydrogen Production (SHP)	$m^3_{Hydrogen}/KgTVS_{fed}$	0.014 ± 0.002	0.03 ± 0.002

In terms of hydrogen production, when the ammonia concentration is close to the threshold value, the Run III is more productive (0.03 m^3/KgTVS fed with H_2 percentage of 30 %) than Run II. This can be explain by the fact that operating with the greater recirculation ratio allows to have a higher concentration of bacteria in the reactor.

4. Conclusion

A dark fermentation process optimized for the hydrogen production was tested in four different recirculation conditions. The control of ammonia in the recycling flow was fundamental to keep the ammonia concentration in the first stage and preserve H_2 production.

From this study, the ammonia concentration greater than a threshold value of approximately 670 $mgN\text{-}NH_4^+/L$ showed inhibition towards the biological process of hydrogen production; in terms of hydrogen production the best condition was found feeding 40 % of AD sludge and 60 % of water and food waste, leading to a SHP of 0.03 m^3/kgTVS fed, with H_2 percentage in the final product of 30 %. However, this condition is not maintained stable for continuous accumulation of ammonia in the system due to the sludge recirculation.

Reference

Antonopoulou G., Gavala H.N., Skiadas I.V., Angelopoulos K., Lyberatos G., 2008, Biofuels generation from sweet sorghum: fermentative hydrogen production and anaerobic digestion of remaining biomass, Bioresources Technology 99, 110-119.

Cavinato C., Bolzonella D., Fatone F., Cecchi F., Pavan P., 2011, Optimization of two-phase thermophilic anaerobic digestion of biowaste for hydrogen and methane production through reject water recirculation Bioresource Technology 102, 8605-8611.

Cavinato C., Giuliano A., Bolzonella D., Pavan P., Cecchi F., 2012, Bio – hythane production from food waste by dark fermentation coupled with anaerobic digestion process: A long – term pilot scale experience, International Journal of Hydrogen Energy 37, 11549-11555.

Lee D.Y., Ebie Y., Xu K.Q., Li Y.Y., Inamori Y., 2010, Continuous H2 and CH4 production from high-solid food waste in the two-stage thermophilic fermentation process with the recirculation of digester sludge, Bioresource Technology 101, S42-S47.

Levin D.B., Pitt L., Love M., 2004, Biohydrogen production: prospects and limitations to practical application, International Journal of Hydrogen Energy 29, 173-185.

Martinez – Perez, N., Cherryman, S.J., Premier, G.C., Dinsdale, R.M., Hawkes, D.L., Hawkes, F.R., Kyazze, G., Guwy, A.J., 2007, The potential for hydrogen-enriched biogas production from crops: scenarios in the UK, Biomass & Bioenergy 31, 95–104.

Porpatham, E., Ramesh A., Nagalingam B., 2007, Effect of hydrogen addition on the performance of a biogas fuelled spark ignition engine, International Journal of Hydrogen Energy 32, 2057-2065.

Valdez – Vazquez I., Poggi – Varaldo H.M., 2009, Hydrogen production by fermentative consortia Renewable and sustainable energy Reviews 13, 1000 – 1113.