

Immobilized Cells in Anaerobic Digestion for Methane from Poultry Manure

THOMAS L. STINEMAN^{*,†}

*Research Biotechnology Associate, Research & Development Division,
The Kroger Co., 1212 State Avenue, Cincinnati, Ohio 45204*

AND

RALPH A. MESSING

*Senior Research Associate, Research & Development Division, Corning
Glass Works, Corning, New York 14831*

Received July 8, 1983; Accepted September 19, 1983

Abstract

The two-stage immobilized microbe waste processor designed for sewage treatment by Messing has been modified to process poultry manure.

The Messing reactor of 120-mL volume was modified and scaled up to a 4-L volume.

Three different carrier materials have been investigated. Temperatures for each of the two stages were examined, and residence time as well as feed concentration were explored.

Analytical data has been computer analyzed using multiple variable correlations and the results of this analysis have indicated directions for optimization.

Index Entries: Immobilized microbes; methane, digestion for; digestion, for methane; two-stage reactor; carriers, controlled pore; anaerobic, digestion, for methane from poultry manure; continuous reactor technology, in methane digestion; cell, immobilized, in anaerobic digestion for methane; poultry manure, digestion for methane from; manure, digestion for methane from poultry.

*Author to whom all correspondence and reprint requests should be addressed.

†Thomas L. Stineman, Fermentation Operation Manager, G.B. Fermentation Industries, Inc., Box 5000, Kingstree, South Carolina 29556.

Introduction

It has been shown that the dimensions of microbes and the size of the pore diameters of inorganic supports will allow one to predict the efficiency of cell accumulation and growth inside inorganic supports (1-4). These studies demonstrated that cells could accumulate in high levels inside the pores, be grown continuously, and the immobilized cell supports be used in a two-stage reactor system for the anaerobic digestion of sewage. It was further demonstrated that this immobilized reactor for sewage was high-rate and produced high BTU gas.

The objectives of this study were to scale up the two-stage sewage reactor and examine its efficiency on a waste stream other than sewage, i.e., poultry waste from laying hens.

To accommodate the particulate material found in poultry manure, it was necessary to modify the immobilized cell sewage reactor. This was done by locating the effluent port on the second stage (the anaerobic stage) at the bottom of the column.

In addition to the scale up and modification of the reactor, information was desired on a variety of carrier supports. Optimal temperatures, residence time, and feed concentration for the system on poultry manure were gathered.

Materials and Methods

The reactor utilized for these studies was a scaled-up version of the Messing two-stage reactor (4) for processing sewage waste. The reactor consists of two stages, a level controller, two pumps, and a check valve. The first stage, the hydrolytic-redox stage, is vertically mounted. The second stage, the anaerobic stage, is horizontally mounted and connected by 1/2-in. tubing to the first stage. The modification of the Messing reactor for poultry manure was to put the discharge on the end, at the bottom of the second stage to allow for passage of large particles and to prevent plugging. The reactor is diagrammatically represented in Fig. 1. A liquid level controller is mounted on the top of the second stage, attached to a gas pump, and activates that pump when the level is low to remove gas buildup. A feed pump is employed to pump through both stages and permit pressure on a check valve mounted to the discharge line of the second stage. Both stages employ porous, inorganic material for immobilizing the microbes.

Both stages consisted of custom-made, water-jacketed, glass columns. The sizes of the columns were approximately 5.0 cm internal diameter \times 112 cm long. Temperature was maintained and controlled by means of a separate Lauda Brinkman circulating water bath attached to each stage. The liquid level controller, mounted at the top of the second (anaerobic) stage, was a Lab Monitor III. Both the feed pump and the gas pump were Masterflex tubing pumps. The check valves used for all of the systems were Circle-Seal, brass, 3 psi, 1/2-in. diameter check valves.

The poultry manure was all from laying hens supplied by Egg City of California, Moorpark, California. The manure was stored at 4-6°C. Prior to use, the manure was diluted with water, ground with a Waring blender, and then filtered through

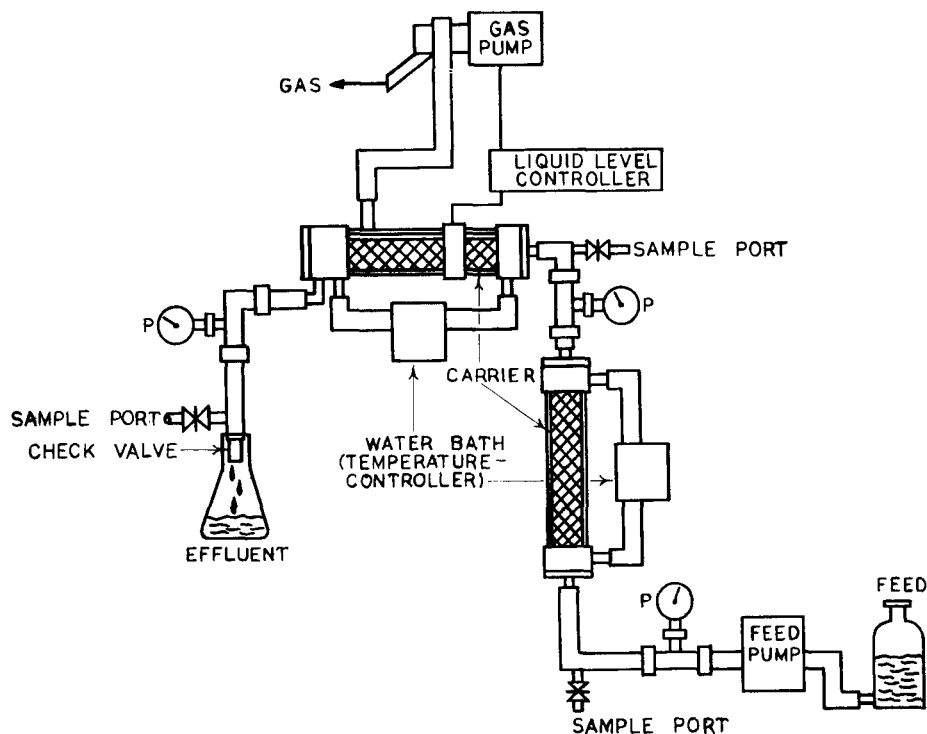


Fig. 1. Poultry manure processor.

burlap to remove coarse particulate matter. The filtered manure was adjusted to the desired feed concentration with water and gently agitated on an orbital shaker to ensure constant feed of suspended solids. The pH of the feed manure was not adjusted and was between 6.8 and 7.3.

Four systems were used and a total of three separate carriers were employed. Two of the carriers were supplied by the Manville Products Corporation, Zelienople, Pennsylvania. These two carriers were insulating bricks size $9 \times 4 \times 2\frac{1}{2}$ in. They were identified as a Johns-Manville 23SL firebrick (JM 23SL) and Johns-Manville Microbiological Reactor Media (JM MRM) brick. Both bricks were cut in half lengthwise, turned on a lathe to 4.9 cm diameter, and then had four $\frac{1}{4}$ in. holes drilled through them lengthwise. These bricks are composed of alumina, silica, and calcium oxide, respectively. The third carrier was supplied by the Aluminum Company of America, Alcoa Center, Pennsylvania. This carrier was an alumina bead, $\frac{1}{2}$ -in. in diameter, and identified as Alcoa Controlled-Pore Alumina #P2311-75 (Alcopal) bead. The Alcopal carrier was used as is. The reactors were charged with 1640 grams of JM 23SL in each of two reactors, 1650 grams of JM MRM in one system, and 2450 grams of the Alcopal bead in the fourth processor. The pore dimensions of the carriers were determined by Mercury Intrusion Porosimeter Analyses and can be seen in Fig. 2. The JM 23SL brick has an average pore diameter of $9\text{ }\mu\text{m}$, pore distribution from 2 to $25\text{ }\mu\text{m}$, pore volume of 0.84 cc/g and a porosity of 67%. The JM MRM has an average pore diameter of $13\text{ }\mu\text{m}$, a pore distribution from 2 to $25\text{ }\mu\text{m}$, a pore volume of 1.22 cc/g and a

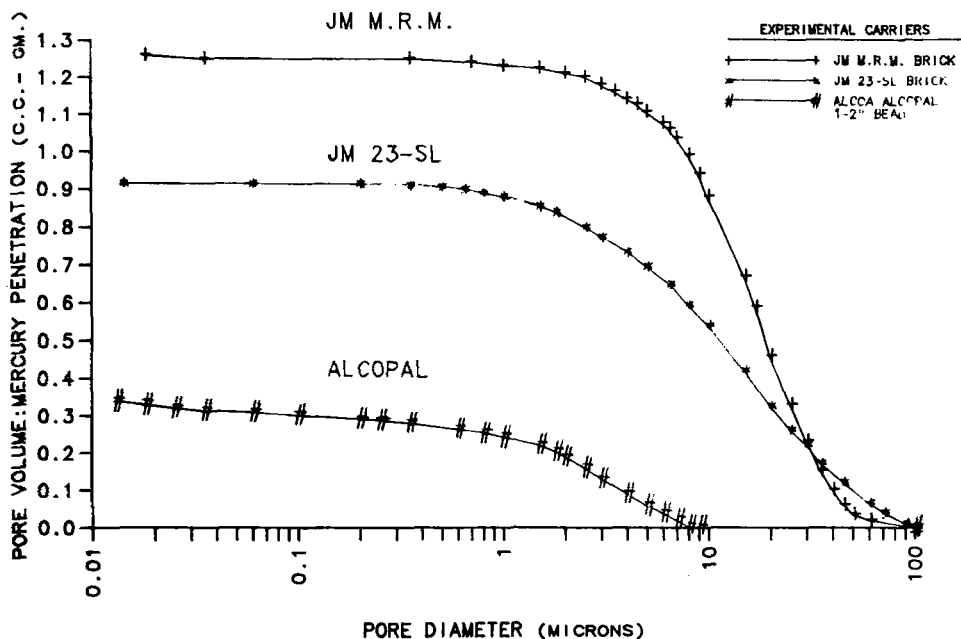


Fig. 2. Carriers for cell immobilization. Mercury intrusion porosimeter analyses on experimental carriers.

porosity of 75%. The Alcopal bead has an average pore diameter of 2 μm , pore distribution from 0.1 to 9 μm , a pore volume of 0.25 cc/g, and a porosity of 36%.

The system volume before the carrier material was packed was 4350 mL including tubing. The fluid volumes of the reactors were measured after the carriers were packed and two were 2850 mL, while two were 3000 mL.

Gas determinations were performed with a Perkin-Elmer Sigma 1 Gas Chromatograph equipped with a Sigma 10 data system and a thermal conductivity detector through 6 ft \times 1/8-in. stainless-steel columns packed with 100–120 mesh Carbosieve 5 (Supelco, Bellefonte, PA.) initial temperature 50°C; temperature program, hold initial 4 min, then 20°C/min to 175°C, hold final temperature 20 min. The flow helium was at 40 mL/min, the injector at 300°C, the detector at 300°C, and the detector voltage at 150 mA. Total carbon determinations were performed with an Oceanography International Model 525 Carbon Analyzer equipped with Horiba Infrared Gas Analyzer. COD analyses were performed with an EPA certified procedure, the Ampule Method, employing a Perkin-Elmer Model 200 spectrophotometer for colorimetric determinations.

The seeding of two of the reactors, one JM 23SL and the Alcopal, was accomplished by obtaining sludge from a digester operating at the city of Cincinnati Gest Street Sewage Treatment Plant, mixing with fresh poultry manure and circulating through the reactors for 1 d followed by dilute poultry manure. One reactor, JM MRM, was seeded by circulating effluent from an operating digester for one day before initiating feed with dilute poultry manure. One of the JM 23SL reactors was seeded with dilute poultry manure only. All reactors were run for 40–50 d before results were considered for final analyses.

Results and Discussion

A matrix of experiments was run utilizing three different carrier materials, residence times from 16 to 55 h, temperature for stage 1 between 20 and 40°C, temperature for stage 2 between 20 and 35°C, and feed concentrations from 1,000 to 15,000 ppm Total Carbon (TC).

Initially, the feed concentrations of manure were monitored using Chemical Oxygen Demand (COD). We found, however, that the reproducibility of COD was poor. It is believed that this was caused by the high level of suspended solids in the feed solution. The Total Carbon (TC) was found to be a more satisfactory indicator of the feed levels for the reactors. COD was monitored only as a rough indicator. Some COD results can be seen in Table 1. It was found that reductions in COD ranged from 67 to 22%, depending on the feed concentration. At approximately 5000 ppm COD, the reductions were the greatest, and then fell as the concentration was increased to 35,000 ppm.

The system performance in relationship to plugging and pore diameter over time was of great interest. The only plugging problems encountered were at the entrance to the 1st stage. This was a result of problems with tubing, rather than the system design itself. The inlet and outlet pressures were monitored; representative data is summarized in Table 2. The largest pressure drop was seen across the 1st stage. It was approximately 0.80 psi on the average, while the pressure drop across the hor-

TABLE 1
Chemical Oxygen Demand

Feed	Outlet	% Change
5 800	1870	-67.7
5 200	2170	-58.3
15,080	8340	-44.7
16,200	8500	-47.5
43,680	33,800	-22.6
35,750	27,750	-22.4

TABLE 2
Pressure Drop Information

Pressure 1, ^a psi	Pressure 2, ^b psi	Pressure 3, ^c psi
2.89	1.80	2.10
2.49	1.80	2.00
3.29	2.80	2.50
2.29	1.75	2.25
2.89	1.80	2.00
2.79	1.90	2.10

^aFeed pressure into bottom of Stage #1 adjusted for head pressure.

^bOutlet pressure of Stage #1 and inlet pressure of Stage #2.

^cOutlet pressure of Stage #2.

izontal anaerobic stage was nominal. The pressure drop across the entire system was on the average less than the drop across the vertical hydrolytic-redox stage. After approximately 6 months continuous operation, two systems were taken apart, one of the JM 23SL carriers and the Alcopal carrier system. About 50 g of carrier was removed from the systems for analysis. This used carrier material was dried at 100°C for 3 h, and porosimeter analysis was then performed on it as well as on the original starting material. There was no appreciable change in the pore diameter, as seen in Fig. 3. There was an approximate 15% decrease in the pore volume for both materials. This is explained by the fact that the organic and cell matter inside the pores could not be removed. This matter would reduce the pore volume and not necessarily represent any pore reductions with regard to operation.

At the time of startup of two of the systems employed, work was done to determine the difference between seeding the digester versus not seeding. One reactor was seeded with a mixture of effluent from an operating digestion system and dilute manure. Another reactor was started with dilute manure only. These mixtures were circulated for 24 h, and then regular poultry manure solutions were employed. The results of this work are seen in Fig. 4. The reactor which was seeded showed better productivity initially than the unseeded system. Gas production started in 5 d in the seeded system, whereas the unseeded system took 7 d longer to produce measureable amounts of methane. The seeded reactor reached steady-state in 25 d, though the reactor started on manure only did not attain steady-state until the 40th d. Results indicate that either method of startup is acceptable after 50 d of operation. Steady-state might be reached in a shorter period of time if operating digester effluent was circulated for longer periods (5–10 days) when starting a new digester. However, fresh manure may be employed if no digester effluent is available.

After 9 months of continuous operation on two systems and 6 months operation on the other two, the data was computer analyzed using multiple variable correla-

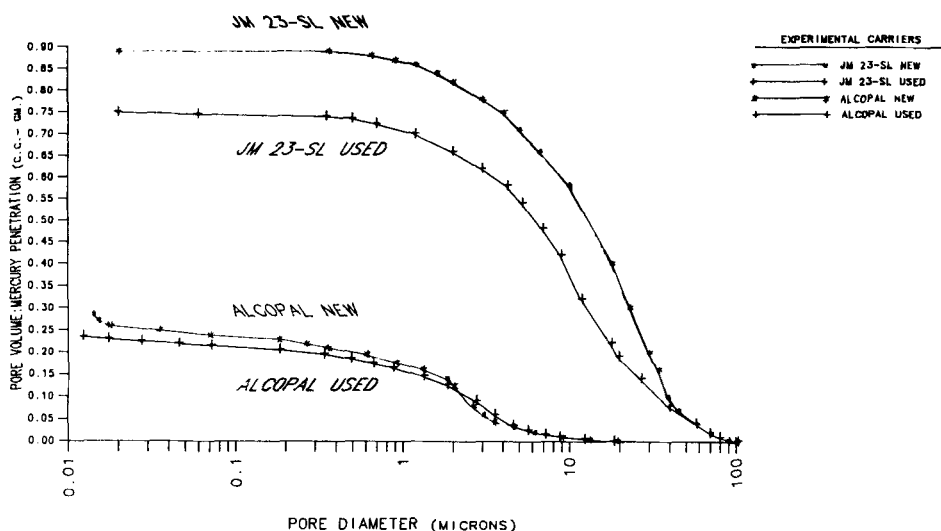


Fig. 3. Carriers for cell immobilization. Mercury intrusion porosimeter analyses of new vs used carriers (6 months continuous).

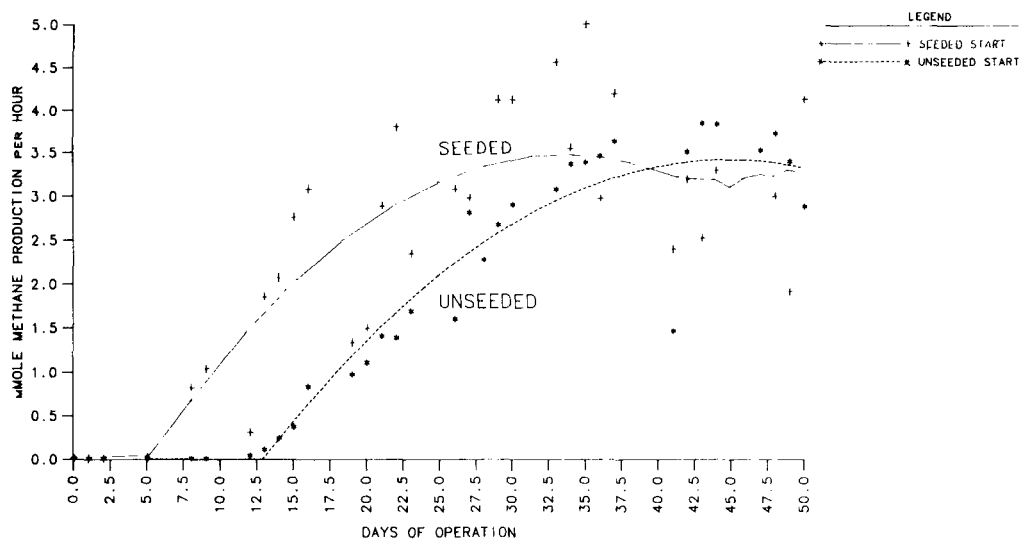


Fig. 4. Digester startup (seeded vs. unseeded start). Methane production, mmol/h.

tion techniques. Separate analyses were done for biogas production per hour and for the methane content of the biogas produced. The intent was to optimize all variables including any multivariable interactions for these two parameters.

The biogas production is related to the carrier material used as seen in Fig. 5. The JM MRM brick had the highest biogas productivity of the carrier materials used. This carrier also had the highest pore volume of the three carriers tested (Fig. 2). It is consistent with earlier work done by Messing et al. (1-3) that the greatest accumulation of cells would occur in the carrier with the greatest pore volume in the correct range of pore diameter. The current results seem to indicate that as the pore volume increased, so does the gas productivity, probably as a result of cell loading. The second best productivity was with the JM 23SL brick, which had pore volumes 40% less than the JM MRM. The gas production for the JM 23SL is also

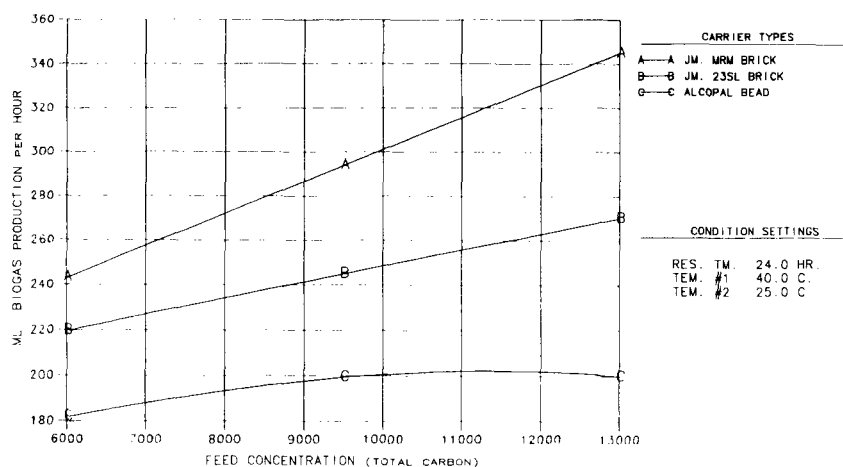


Fig. 5. Milliliters of biogas production per hour, related to feed concentration and carrier type.

from 10 to 30% less than the JM MRM. Results also seem to indicate that as the feed concentration increases, the effect is greater on the JM MRM than the other two materials. This seems to relate to porosity of that material and microbial density within the carrier in relation to the other materials tested. Gas production rates increase on this carrier at a faster rate than on the other materials tested.

Stress on cell loading would be greater at the higher feed concentrations.

Figure 6 demonstrates the relationship between residence time and feed concentration (TC). Gas productivity increases with residence time up to 23 h, falling steadily after that point. Another increase was found in productivity after 50-h residence time, but this was not pursued. It is believed that the most easily digested portion of poultry manure occurs in the earliest time period and further digestion only occurs after considerably longer periods of time. The amount of gas production is directly related to the carbon concentration in the feed (Fig. 6). Residence times exhibited with poultry manure are much longer than those encountered with sewage (4), but are consistent with the fact that COD concentrations worked with were much greater than the earlier work with sewage.

A strong relationship was found between the temperature of the anaerobic stage and feed concentration. As the temperature of stage 2 is increased, it has effect at the higher feed concentrations (Fig. 7). Essentially, at feed concentrations up to 12,000 (TC), the temperature of the anaerobic stage impacts strongly above 28.5°C. The temperature of stage 2 must be kept lower than this if higher feed concentrations are to be attempted.

In order to optimize any methane digestion system, it is not only important to improve the biogas output, but also to attempt to improve the methane content and resulting BTU level of the gas. Figure 8 shows the comparison of methane production in relationship to the carrier used and the feed concentration of poultry manure. As was seen earlier, the carrier with the highest pore volume, JM MRM, exhibits the highest levels of methane production. The other two carriers, Alcopal

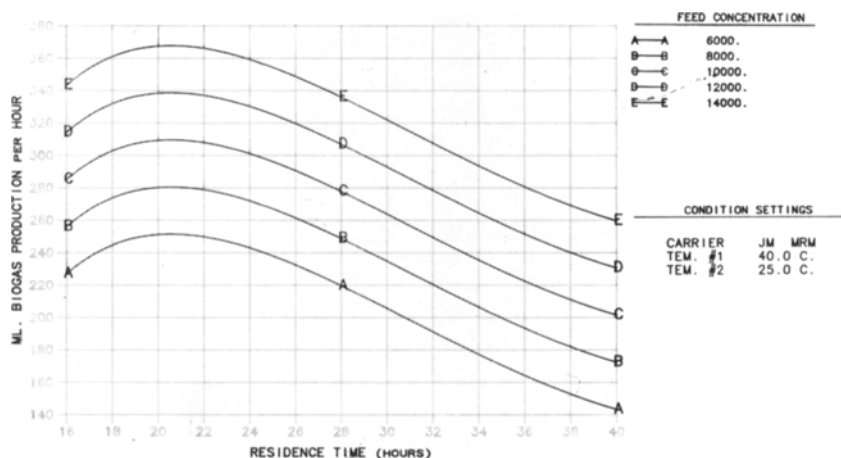


Fig. 6. Milliliters of biogas production per hour, related to residence time and feed concentration.

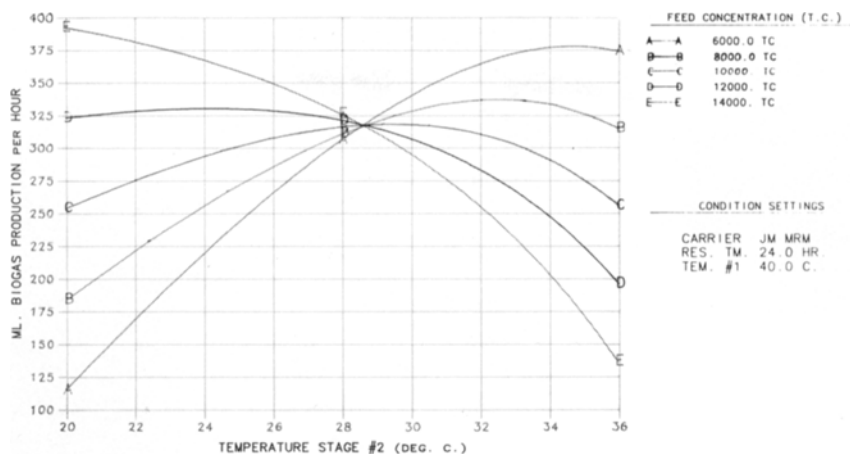


Fig. 7. Milliliters of biogas production per hour, related to temperature stage #2 (degrees C,) and feed concentration (total carbon).

and JM 23SL, exhibit lower methane production as the feed concentration increases. These two carriers actually decrease in methane content when feed concentration is increased. The Alcopal carrier actually outperforms the JM 23SL carrier, which is not consistent with the porosity characteristics of the two materials. At lower feed concentrations, JM 23SL and Alcopal have higher methane production than JM MRM, but lose their performance above 6000 ppm (TC) in the feed.

As residence time increases above 14.0 h, so does the methane content. There is a relationship between residence time and feed concentration for methane production as is illustrated in Fig. 9. As feed concentration increases above 5000 ppm (TC), it becomes important to increase residence time. As throughput through the system increases in terms of carbon, methane content of the gas also increases if feed rate of carbon is decreased.

Earlier work by one of the authors with sewage (4) indicated that temperatures lower than 40°C were optimum for stage 1 (hydrolytic-redox stage). In this work

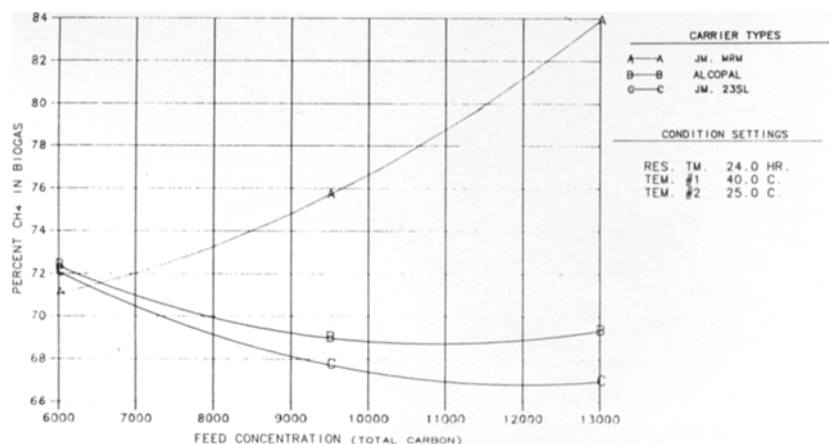


Fig. 8. Percent CH₄ in biogas, related to feed concentration and cell carriers.

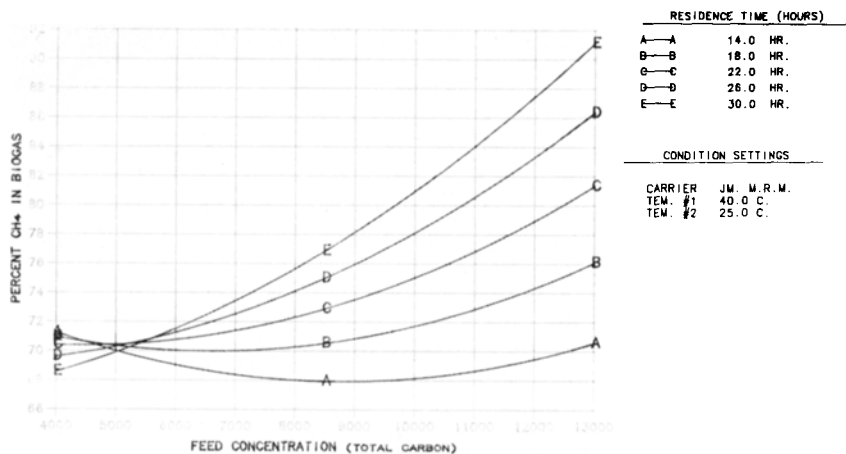


Fig. 9. Percent CH_4 in biogas, related to feed concentration and residence time.

with poultry manure, it was found that the rate of gas production increased as the temperature of stage 1 increased. In contrasting the percentage of methane in the gas produced, it appears that the ideal temperature for the hydrolytic–redox stage interacts with the temperature of the anaerobic stage (Fig. 10). In order to allow for a higher stage 1 temperature to increase gas production, it is necessary to lower the temperature of the anaerobic stage to below 25°C to increase the methane content of that gas. This may occur because of the solubility of carbon dioxide at the lower temperatures and its conversion to methane. As the temperature of the hydrolytic–redox stage is increased, methane content decreases unless the temperature of the anaerobic stage is lowered. Gas analyses showed a higher content of carbon dioxide at temperatures above 25°C for stage 2 and a lower methane content when the temperature of the first stage was raised.

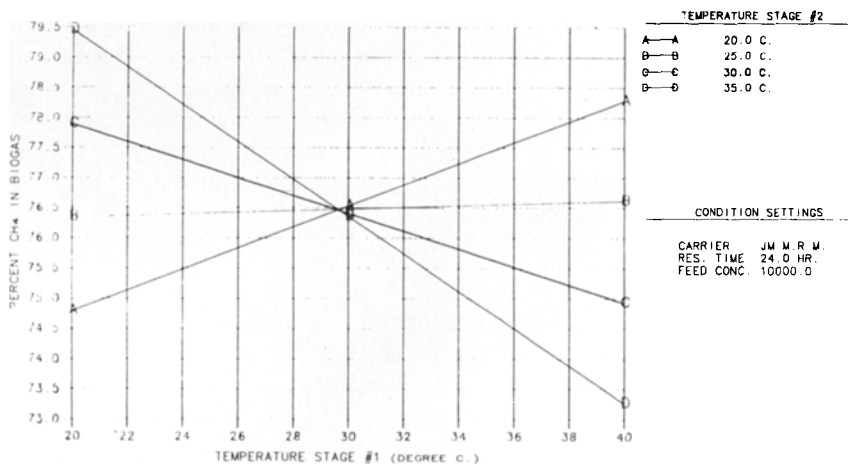


Fig. 10. Percent CH_4 in biogas, related to temperature stage #1 and temperature stage #2.

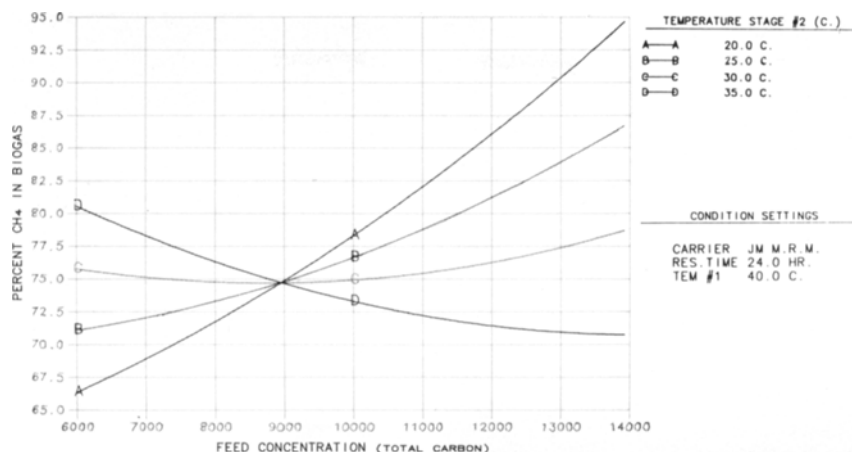


Fig. 11. Percent CH_4 in biogas, related to feed concentration and temperature stage #2.

It was necessary to lower the temperature of the anaerobic stage as the feed carbon concentration was increased above 9000 ppm (TC). As feed concentration increases above this level (illustrated in Fig. 11), the percent methane in the gas decreases at temperatures above 30°C in the anaerobic stage. At feed carbon concentrations above 10,000 ppm, the highest methane concentrations are achieved when the temperature of the anaerobic stage is at 20°C. This also seems to be an indicator of increased carbon dioxide solubility in the reactor stage 2 at temperatures at and below 25°C.

Summary and Conclusions

The two-stage sewage reactor of Messing can be modified to process poultry manure. This modified, scaled-up system was operated for over 9 months continuously. Carrier materials show no measurable amounts of destruction or pore modification. Optimization results indicate that if high carbon concentrations of poultry manure are to be used, the temperature of the hydrolytic–redox stage should be at or above 40°C, and the temperature of the anaerobic stage should be at or below 25°C to obtain the highest gas productivity with highest methane concentrations. At feed concentrations of 10,000 ppm (TC) poultry manure, residence times employed should be from 22 to 24 h. Of the carrier materials tested, JM MRM was found to be best for both gas production and high methane concentration. A carrier material with high porosity and pore diameter of 2–35 μm will outperform a carrier material with similar pore diameter and lower pore volume.

The reactors that were used had equal volumes for the hydrolytic–redox stage and the anaerobic stage. There is some indication that the size of the hydrolytic–redox stage should be smaller in relationship to the size of the anaerobic stage to improve performance. More extensive work will have to be completed to characterize this ratio.

Acknowledgments

We would like to thank Dr. Robert Williams, Vice President of the Research and Development Division of The Kroger Co. for his total support in this project. Without that support, the work could not have been accomplished.

Also, thanks are extended to Mr. Rick Norton for all of his contributions to the project, his support and enthusiasm over many long and tedious periods.

Much support was extended by many people at both The Kroger Co. and Corning Glass Works for analytical analysis and our thanks are extended.

We wish to thank Ms. Millie Russell for her efforts in the preparation of this manuscript.

References

1. Messing, R. A., and Opperman, R. A. (1979), *Biotechnol. Bioeng.* **21**, 49.
2. Messing, R. A., Opperman, R. A., and Kolot, F. B. (1979), *Biotechnol. Bioeng.* **21**, 59.
3. Messing, R. A., Opperman, R. A., and Kolot, F. B. (1979), *ACS Symposium Series* **106**, 13.
4. Messing, R. A. (1982), *Biotechnol. Bioeng.* **24**, 1115.