

The anaerobic digestion of organic matter in sugar-beet mud

M. G. Keizer, F. A. M. de Haan, J. J. C. Blom and J. W. M. Knaapen

Department of Soil science and Plant nutrition, Agricultural University, Wageningen, the Netherlands

Accepted: 8 January 1981

Key words: anaerobic digestion, sugar-beet mud, volatile fatty acids, methane production, bad odour

Summary

Storage of sugar-beet mud in the traditional way, viz direct dewatering after pumping the slurry in storage basins, may cause bad odour nuisance because of digestion of organic substances. In order to prevent these bad odour problems one can keep the mud submerged during the digestion period. No production of malodorous compounds was observed in this way, also not when after one year of submersion the basins were dewatered and the mud began to ripen. Experiments were performed with small mud fields, barrels and pots in order to gain insight in the anaerobic digestion processes and to develop methods to optimize these. During these experiments the mud was subjected to a number of different treatments. It was found that no essential difference exists between the digestion process in dewatered and submerged mud. The digestion rate of the produced volatile fatty acids (VFA) depends on the growth rate of the bacteria responsible for the conversion of VFA into CH_4 and CO_2 . This growth rate seems to be controlled in the pot experiment by the amount of available $\text{NH}_4\text{-N}$. Treatment of the mud, for instance inoculation with old ripened mud or addition of carbonates, is only successful when the environmental conditions in the mud are very unfavourable for methane production (high VFA concentrations and low pH).

Introduction

The washing water of a beet sugar factory contains soil particles, beet pieces and root remnants. It also has a substantial concentration of sugar. This water is led to a sedimentation basin and after sedimentation the remaining slurry (ca. 15 % dry solids on weight basis) is pumped into mud storage fields (Fig. 1). The decanting water of sedimentation basin and mud storage fields is re-used and at the end of the cam-

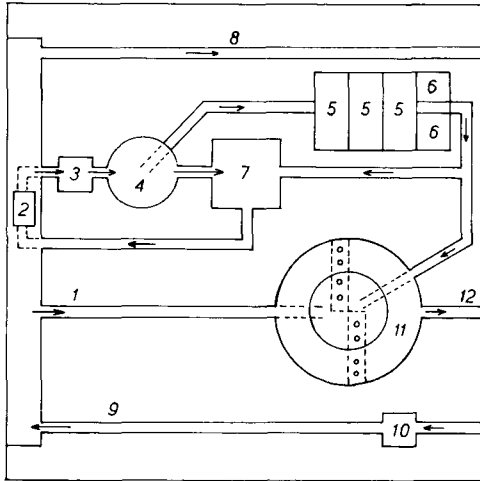


Fig. 1. Water flow diagram of a sugar-beet factory. 1, sewage water; 2, beet washing and transporting water; 3, sieve installation; 4, sedimentation basin; 5, mud storage fields; 6, after-sedimentation basins; 7, storage pond; 8, cooling water; 9, intake river water; 10, dirt catcher; 11, sewage water treatment; 12, discharge of purified water. (From Suiker Unie, 1978a.)

paign – after biological sewage water treatment – discharged (de Vletter, 1980; Suiker Unie, 1978a).

During the ripening of the mud in the dewatered mud storage fields, the organic matter in the mud is digested by micro-organisms anaerobically and eventually also aerobically. Dewatering of the mud is promoted by making ditches and by plant growth (Willet et al., 1976).

The dewatered mud can give bad odour nuisance probably caused by incomplete anaerobic and aerobic digestion. This may happen particularly at somewhat higher temperatures (in April/May) by a sudden release of many volatile fatty acids (VFA) and possibly other malodorous compounds.

Incomplete aerobic digestion can occur due to insufficient aeration of the top-layer of the mud. Formation of a thick crust at the surface may play a role. This crust might be produced by certain micro-organisms. Beneath the crust accumulation of malodorous compounds can occur which will release when the crust breaks open.

At the beet sugar factory in Puttershoek the bad odour gave so many problems that measures had to be taken to prevent further nuisance. Here a new method was developed to treat the sugar-beet mud: the ‘submerged mud’ method (Suiker Unie, 1978b). At the end of the campaign the mud is not dewatered immediately, but kept under a water layer of about ½ m, thus admitting only anaerobic digestion in the mud. In the water layer aerobic digestion of VFA, diffused from the mud into this water, takes place. To prevent bad odour the water layer has to be refreshed regularly to keep the oxygen content at a sufficient level. After complete digestion the water is discharged and the mud can start to ripen.

During anaerobic digestion several malodorous compounds can be broken down as was shown for pig waste (Spoelstra, 1978; van Velsen, 1977). The process can be divided into three phases: hydrolysis, acid formation and methane production. The last phase comprises also an acetogenic phase converting VFA into suitable sub-

strate for methane bacteria (Bryant, 1976; Lettinga, 1977). With the methane production the final elimination of organic matter takes place by formation of CH_4 and CO_2 . Some important characteristics of methane bacteria are: strictly anaerobic, substrate specific, slow growth, pH sensitive (optimum at pH 6.7-7.3) and sensitive to toxic compounds (Lettinga, 1977).

The anaerobic digestion in the mud was studied in order to find the main parameters governing the processes involved, and to develop methods for influencing these processes by changing the environmental conditions (Blom, 1978; Knaapen, 1979).

For application in practice it is useful to know how fast digestion in the mud proceeds in order to establish the moment that dewatering of the mud can start without getting bad odour nuisance.

Methods and materials

The investigations included experiments with pots, small experimental fields, barrels and also measurements in a mud storage field. Experiments started November 1977

Table 1. The different treatments of pots, experimental fields, barrels and mud storage field.

Exp. No	Number of pots or fields	Treatment
<i>Pot experiment</i>		
1	3	fresh mud (FM); 15 °C; submerged; no water refreshment (NWR)
2	2	FM + 10 % old mud (OM); 15 °C; submerged; NWR
3	3	FM + 25 % OM; 15 °C; submerged; NWR
4	3	100 % OM; 15 °C; submerged; NWR
5	2	FM + 10 mg sewage sludge/kg wet mud; 15 °C; submerged; NWR
6	2	FM; 15 °C; submerged; with water refreshment
7	3	FM; 15 °C; dry (no overlying water)
8	2	FM + 25 % OM; 15 °C; dry (no overlying water)
9	2	FM + 0.75 g CaCO_3 /kg wet mud; 15 °C; submerged; NWR
10	2	FM + 1.70 g CaCO_3 /kg wet mud; 15 °C; submerged ; NWR
11	2	FM + (1.7 g CaCO_3 + 10 mg sewage sludge)/kg wet mud; 15 °C; submerged; NWR
12	2	FM; 5 °C; submerged; NWR
13	2	FM + 1.70 g CaCO_3 /kg wet mud; 5 °C; submerged; NWR
14	2	FM + 1.52 g NH_4Cl /kg wet mud; 15 °C; dry (no overlying water)
15	2	FM + 3.02 g KNO_3 /kg wet mud; 15 °C; dry (no overlying water)
<i>Field experiment</i>		
1	1	fresh mud; dry (no overlying water)
2	1	FM + 1.5 g CaCO_3 /kg wet mud; dry (no overlying water)
3	1	FM + 1 % old mud (wet weight %); dry (no overlying water)
4	1	FM + 1.5 g CaCO_3 /kg wet mud + 1 % OM; dry (no overlying water)
Barrels No 1-4:		mud from fields 1-4; submerged; NWR
Mud storage field:		fresh mud + 1.5 g CaCO_3 /kg wet mud; submerged; with water

and lasted about one year. The pots used had a content of ca. 2.5 litres (with 3 kg wet mud per pot), with a mud layer thickness of ca. 0.15 m; experimental fields were ca. 10 m × 10 m, with a mud layer thickness of ca. 1.5 m; barrels had a content of ca. 100 litres, with a mud layer thickness of ca. 0.7 m. The surface area of the mud storage field was ca. 2 ha, with a mud layer thickness of ca. 1.5 m.

Environmental conditions

Changes of environmental conditions in the mud (fresh mud of the campaign 1977) were obtained by the following treatments (for details see Table 1).

Inoculation with old mud (ripened mud, of the campaign 1976) and with anaerobic sewage sludge. The fermentation rate may be influenced by adding already adapted bacteria. In practice inoculation with more than 1 % old mud is not feasible. In the pot experiment much more old mud was added (10 and 25 %, wet weight %) so that here differences in composition of old and fresh mud may play a role as well.

Dry mud and submerged mud, with or without refreshment of the overlying water layer. This treatment influences the anaerobic and aerobic digestion in mud and water. With the dry method formation of the surface crust can prevent aeration of the top-layer of the mud and cause bad odour nuisance. Refreshment of overlying water promotes rinsing of VFA and other malodorous compounds. With this refreshment the O₂ content of the overlying water remains high enough for aerobic digestion. Anaerobic digestion in the water may give bad odour.

Addition of CaCO₃. This treatment was performed in order to prevent decrease of the pH, caused by strong VFA production. Methane bacteria are very sensitive to low pH values. In practice already 1.5 g CaCO₃/kg wet mud was added (only in 1977).

Different temperatures. Temperature influences digestion course and fermentation rate. Temperatures of the mud in the fields varied roughly between 0 and 15 °C. In the pot experiment temperature was kept constant at 15 °C or 5 °C.

Addition of NH₄-N or NO₃-N. Nitrogen addition may cause:

- reduction of C/N. Crust formation is possibly caused by micro-organisms which grow well at high C/N. In the mud C/N is probably high due to the high content of cellulose and sugar. Thus, a lower C/N could prevent formation of the crust by these organisms.
- raise of redox potential with NO₃-N, which will reduce methane formation. Methane production requires a redox potential below 150-220 mV (Takai & Kamura, 1966).
- increase of the growth rate of the bacteria by NH₄-N nutrition.

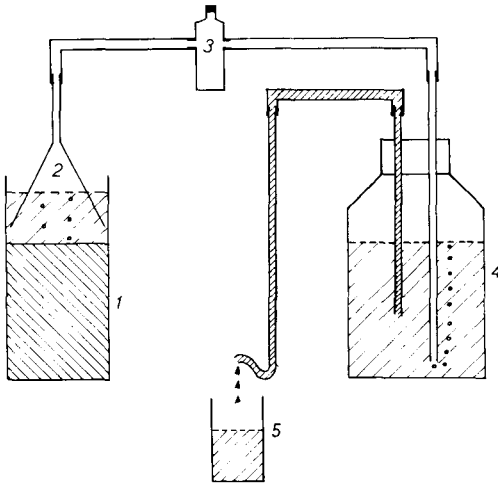


Fig. 2. Schematic representation of the gas collection system in the pot experiment. 1, pot with submerged sugar-beet mud; 2, fyunnel; 3, sampling device for qualitative gas measurement; 4, bottle of Mariotte filled with acid; 5, beaker to collect removed acid.

Environmental characteristics

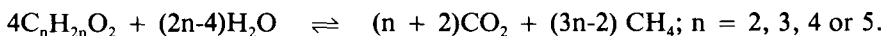
Gas production. Gases were collected in an adapted bottle of Mariotte. Only pots and barrels with overlying water offer a good opportunity for gas collection. In the pot experiment this was performed by placing a funnel up side down in the overlying water (Fig. 2). The same system was used with the barrels. Gas collection of the barrels did not function well because of great temperature variations in the open air, which caused volume changes in the bottle of Mariotte.

Gas composition. In the pot experiment gas samples were taken with an injection needle after 195, 227 and 302 days. Analysis was performed by means of gas chromatography (CH_4 , CO_2 , air).

Volatile fatty acids (VFA). VFA were analysed by means of gas chromatography as acetic acid (C_2), propionic acid (C_3), butanoic acid (C_4), iso-butanoic acid ($i\text{-C}_4$), valeric acid (C_5) and iso-valeric acid ($i\text{-C}_5$). Analysis was performed in the supernatant after centrifuging the mud samples during 10 min at 18000 rev/min. Overlying water was filtered prior to analysis.

Calculation of CH_4 production. Methane production was calculated:

- from VFA concentration according to the following equation:



The amount of VFA is found as the difference between the maximum amount of VFA in a certain period and the amount at the end of that period.

- from gas production and gas composition. Correction on the produced amount

of gas was necessary because of acid evaporation ($5 \text{ cm}^3/\text{day}$) and gas side losses (factor 1.19).

pH. The pH of overlying water and supernatant was measured with a glass electrode and KCl-saturated calomel electrode. pH-KCl was also determined in the centrifuged mud.

Redox potential. Redox potentials were measured after an incubation time of 2 days by using a Pt electrode and a KCl-saturated calomel electrode. Measurements were performed only in pots with overlying water.

Organic matter. The organic matter content was measured routinely by loss on ignition (weight difference between 105 and $600 \text{ }^\circ\text{C}$). At the start of the experiments C determinations according to Kurmies were also performed (Houba et al., 1974). The method based on loss on ignition is less accurate because of errors due to the presence of carbonates and VFA.

Carbonates. Carbonates were determined according to Van Wesemael (Houba et al., 1974).

Bad odour. Determination of bad odour by personal observation only.

Sampling

Sampling of the pots was performed about once a month by means of an adapted pipette. The samples amounted to about 0.7 % of the total volume. Barrels were sampled every 14 days by means of a metal pipe. Sampling depth was about 35 cm above the bottom. The experimental fields and the mud storage field were sampled with the use of a block shaped metal box with valves at top and bottom. Samples from different places of each field were mixed. Sampling of the experimental fields was repeated every 14 days at a depth of 0-10 cm and 60-80 cm. The mud storage field was sampled once a month at a depth of about 1 m.

All samples were subjected to determination of: VFA, loss on ignition, pH-KCl, pH-supernatant and watercontent.

Inaccuracy of the determinations was caused mainly by great heterogeneity of the mud.

Results and discussion

Experimental results are presented in Tables 2, 3, 4 and 5 and in Fig. 3, 4, 5 and 6. Only average values are given. Experimental points are not shown in Fig. 5 for practical reasons. Also, no correction was made in Fig. 5 for acid evaporation and gas side losses. Experimental results of the barrels are not presented because they agree very well with the results of the lower layer of the experimental fields. None of the treatments produced a real bad odour.

ANAEROBIC DIGESTION OF ORGANIC MATTER IN SUGAR-BEET MUD

Table 2. Some characteristics of the sugar-beet mud from the pot experiment.

Characteristic	Old mud (1976) at day 0	Fresh mud (1977) at day 0	Fresh mud (1977) at day 374
Water (%)	45	75	70 (submerged)
		75	58 (dry)
Organic matter (%)	9.7	18.5	12
Total VFA (g/l)	0.6	6.7	0.3
pH-KCl	7.0	6.7	6.6
pH-supernatant	ca. 7.4	ca. 5.7	7.2
carbonates (%)	4.5	3.3	ND
N-total (%)	0.4	0.6	ND
C (%)	3.3	ca. 7	ND

ND = not determined.

Pot experiment

Some general data of the mud used in the pot experiments are presented in Table 2.

Submerged mud (treatment 1). From Fig. 3a, line 1 it is clear that production of VFA took place specially during the first 40 days. Fig. 4 shows that after 40 days still only i-C₄, C₅ and i-C₅ were produced in relatively small amounts. Till day 180 the total VFA concentration remained rather constant. During that time period there hardly was any gas production (Fig. 5, line 1). Retardation of hydrolysis or of

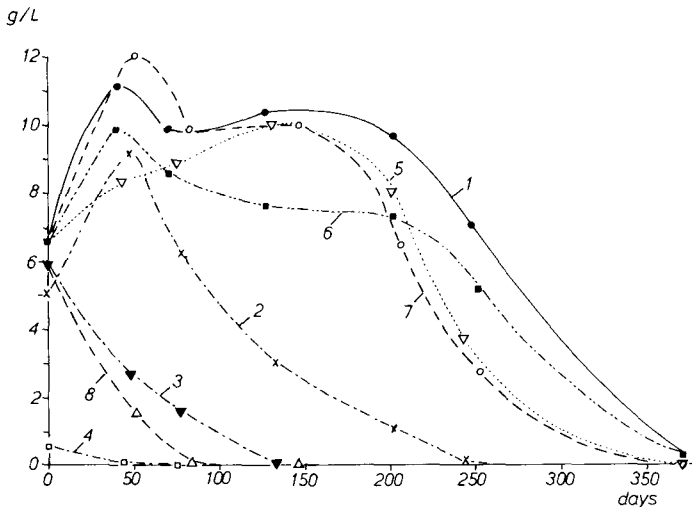


Fig. 3a. Total VFA concentration (g/l) in the pot experiment as a function of time. Treatments 1 (●), 2 (×), 3 (▼), 4 (□), 5 (▽), 6 (■), 7 (○), 8 (△).

acid formation after 40 days was not expected because of the relatively high pH (pH-supernatant = 5.7) and relatively low total VFA concentration (ca. 11 g/l).

Presumably all easily decomposable organic matter was transformed to VFA du-

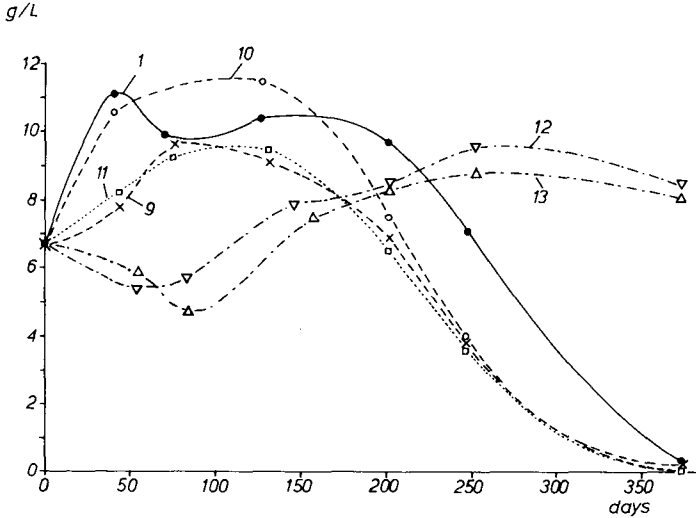


Fig. 3b. Total VFA concentration (g/l) in the pot experiment as a function of time. Treatments 1 (●), 9 (×), 10 (○), 11 (□), 12 (▽), 13 (Δ).

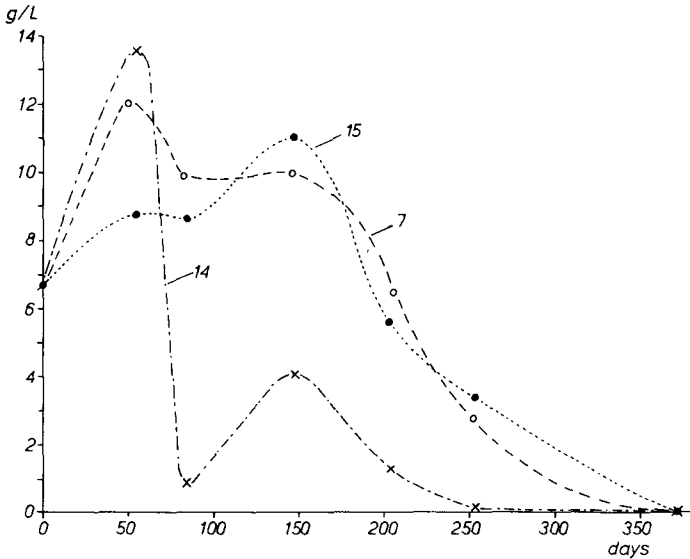


Fig. 3c. Total VFA concentration (g/l) in the pot experiment as a function of time. Treatments 7 (○), 14 (×), 15 (●).

Table 3. Amount of CH₄ calculated from VFA measurements and from gas production and % CH₄ in the gas mixture.

Treatment No (Table 1)	Time period (days)	CH ₄ from VFA concentration (mmol)	CH ₄ from gas production (mmol)	CH ₄ in gas mixture (%)
1	40-320	420	350	78
2	40-250	340	330	84
3	0-250	240	310	84
3	40-250	120	180	84
4	0-250	25	130	81
5	0-320	430	400	79
5	40-320	430	370	79
6	40-320	450	270	77

ring the first 40 days. This also follows from the reasonably good agreement between the amounts of mmol CH₄ calculated from VFA concentrations and from measured amounts of gas (cf. Table 3).

Gas production proceeds well after about 180 days (Fig. 5, line 1). Methane formation hardly occurred during the first 160 days, probably by a too low pH (at day 0 pH-supernatant = 5.7 and at day 160 pH-supernatant = 6.0) combined with a high total VFA concentration (> 10 g/l). The redox potential was found to be low enough (< 150 mV) to make methane formation possible from the beginning. In spite of better environmental conditions after 160 days, such as higher pH and lower total VFA concentration, the exponential growth period was rather short; presumably because of lack of nutrients. After 180 days gas production remained at a constant level of ca. 55 cm³/day. From Fig. 4 it appears that C₂ and C₄ were digest-

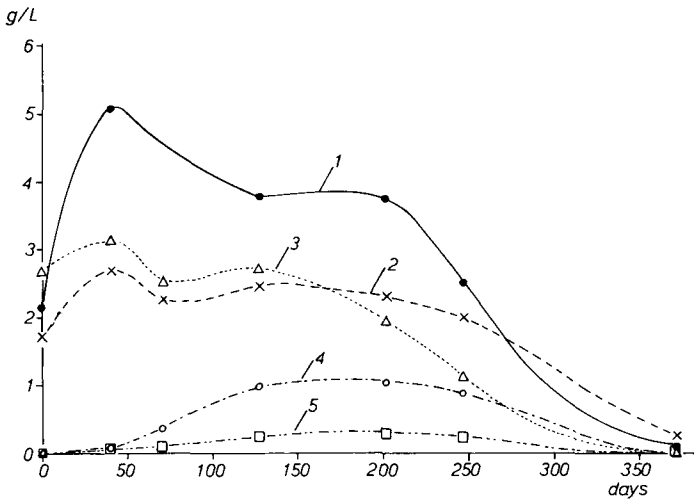


Fig. 4. VFA concentration (g/l) of the submerged mud treatment (No 1) in the pot experiment as a function of time. 1, C₂ (●), 2, C₃ (×), 3, C₄ (Δ), 4, i-C₄ (○), 5, C₅ and i-C₅ (□).

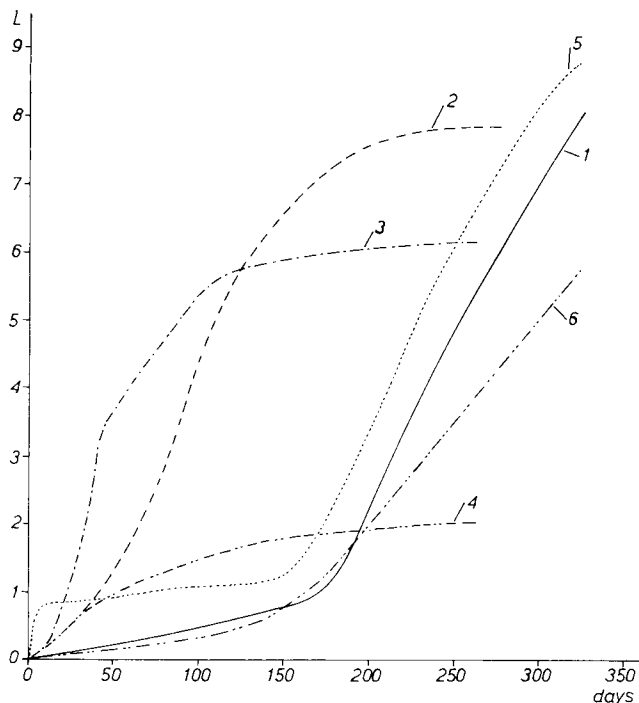


Fig. 5. Cumulative amount of gas (l) produced in the pot experiment as a function of time. Treatments 1, 2, 3, 4, 5 and 6.

ed first. C_3 was digested after the other VFA had almost disappeared. Obviously propionic acid is an unfavourable substrate.

Inoculation with old mud (treatments 2, 3, 4). Addition of large amounts of old mud to fresh mud had a stimulating influence on anaerobic digestion (Fig. 3a and 5, lines 2, 3). Methane production started immediately and maximum CH_4 production was greater: about $75 \text{ cm}^3/\text{day}$ with 10 % OM and about $100 \text{ cm}^3/\text{day}$ with 25 % OM. The first may have been caused by adding adapted bacteria in combination with improvement of environmental conditions (higher pH, lower total VFA). The latter may have been caused by more available nitrogen and other nutrients in the old mud. Since gas production occurred simultaneously with VFA production at treatments 3 and 4, the amount of mmol CH_4 calculated from VFA data is much lower than that from the gas data (cf. Table 3). From the calculations for treatment 3 one can deduce that during the first 40 days a large amount of VFA was produced.

Inoculation with anaerobic sewage sludge (treatment 5). Addition of anaerobic sewage sludge caused strong methane production after a few days (Fig. 5, line 5 and Table 3). CH_4 production, however, lasted only a short time, presumably because of unfavourable environmental conditions (low pH and high total VFA). Methane pro-

duction started again after some 150 days and remained rather constant (ca. 55 cm³/day). Total VFA concentration was lower as in treatment 1, due to the early start of CH₄ production (Fig. 3a, line 5). This can also explain the start of the CH₄ production at 150 days in stead of 160 days.

Water refreshment (treatment 6). Refreshment of overlying water caused rinsing of VFA from the mud (Fig. 3a, line 6). CH₄ production did not start earlier as a result of this rinsing (Fig. 5, line 6). Methane production was much lower than without refreshment (ca. 30 cm³/day), presumably caused by elimination of available nitrogen with the refreshments. The calculated amount of mmol CH₄ from VFA data is here higher than from gas data because much of the VFA is removed after production by the refreshments (cf. Table 3).

Dry mud (treatments 7, 8). Total VFA digestion in the dry pots appeared to be faster and also started earlier than in pots with overlying water (Fig. 3a, line 7, 8). This may have been caused by a higher pH at the beginning and presumably also by more available nitrogen. Digestion certainly was anaerobic because of the formation of a surface crust. The digestion process was completely comparable with that of treatment 1 (Fig. 4).

Addition of CaCO₃ (treatments 9, 10, 11). With addition of CaCO₃ methane production started about 30 days earlier than in treatment 1, probably by raising otherwise too low pH values (Fig. 3b, line 9, 10).

Gas production remained constant after 150 days (ca. 55 cm³/day). Apparently the carbonates already present in the mud had a too low reactivity to buffer the pH sufficiently and keep it at a sufficiently high level. The combination of inoculating sewage sludge and adding CaCO₃ yielded results as expected from the separate treatments (Fig. 3b, line 11).

Low temperature (treatments 12, 13). Temperature appeared to be the most critical environmental condition. At 5 °C no CH₄ was produced and hydrolysis and acid formation proceeded much slower than at 15 °C (Fig. 3b, line 12). In the beginning total VFA concentration declined possibly by diffusion of VFA in the overlying water. Addition of CaCO₃ seemed to have hardly any influence at this low temperature (Fig. 3b, line 13).

Addition of nitrogen (treatments 14, 15). From Fig. 3c, line 14 it appears that with NH₄ nutrition VFA are digested about 80 days earlier than without nitrogen nutrition or with NO₃ nutrition. The digestion rate of VFA is here about 30 % higher than at treatment 7 or 15. Although the strong decline in total VFA concentration between 50 and 100 days (Fig. 3c, line 14) could have been caused by local strong CH₄ production due to the heterogeneity of the mud, it is also possible that this decline was a result of the NH₄ nutrition. NH₄ may induce increased activity of bacteria which may play a role in the different breakdown processes. Any influence of N nutrition on crust formation was not observed. It is questionable whether micro-or-

ganisms are important in the formation of the surface crust. Diffusion of Fe (II) to the surface and precipitation of Fe (III) can also be responsible for crust formation.

NO₃ nutrition did not have much influence on the overall breakdown processes (Fig. 3c, line 15). The composition of the VFA, however, differed from the treatments without N nutrition and with NH₄ nutrition: less C₃ and C₄, even a decline of C₄ in the first 50 days, and more C₂ (cf. Fig. 4). Obviously addition of NO₃ stimulated the digestion of C₄ and the formation of C₂. The increase of C₂ can be caused by conversion of C₃ and C₄ in C₂ or by a different breakdown process of the easily decomposable organic matter.

A raise of the pH-supernatant by denitrification (ca. 6.2 without N and ca. 6.4 with NO₃ nutrition) should cause an earlier start of the CH₄ production. This did not occur because of probable retardation of the CH₄ production due to an increase of the redox potential.

Field experiments

The strong heterogeneity of the mud caused large scattering of the results obtained in the experimental fields. Characteristics of the mud in the experimental fields at two different times are given in Table 4. The organic matter decomposition (the course of total VFA concentration at two depths) is presented in Fig. 6. After about 160 days the fields show a decline in total VFA concentration. Presumably before that time temperature was too low to make CH₄ production possible. In the top layer (0-10 cm) methane production started about 20 days earlier than in the lower layer (60-80 cm), due to increased temperature in the top layer. During the first 60 days total VFA concentration declined strongly in the top layer probably caused by rinsing with rain water. In this field experiment addition of CaCO₃ or inoculation with old mud showed no significant influence.

Table 4. Some characteristics of the sugar-beet mud from the field experiments (average values of the 4 fields).

Characteristic	At day 0		At day 272	
	0-10 cm	60-80 cm	0-10 cm	60-80 cm
Water content (%)	71	60	24	46
Organic matter (%)	12.6	10.6	10.3	9.4
Total VFA (g/l)	4.1	4.1	< 0.1	< 0.1
pH-supernatant	ca. 7.0	ca. 6.6	7.8	7.4
Carbonates (%)	5.7	5.7	ND	ND
N-total (%)	0.4	0.4	ND	ND

ND = not determined.

ANAEROBIC DIGESTION OF ORGANIC MATTER IN SUGAR-BEET MUD

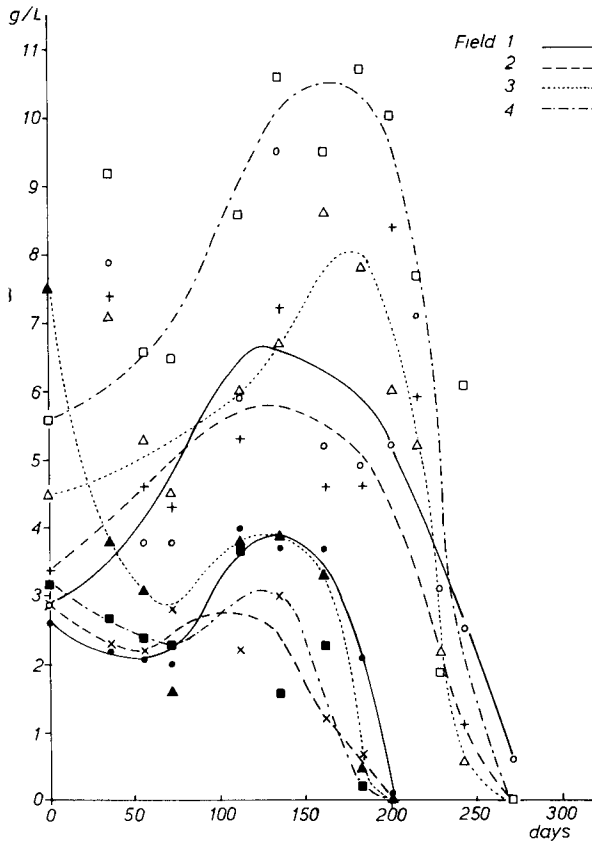


Fig. 6. Total VFA concentration (g/l) in the experimental fields as a function of time. Top layer (0-10 cm): fields 1 (o), 2 (+), 3 (Δ), 4 (\square). Lower layer (60-80 cm): fields 1 (\bullet), 2 (\times), 3 (\blacktriangle), 4 (\blacksquare).

Table 5. Some characteristics of the sugar-beet mud from the mud storage field during the period June-December 1978. Data from Suiker Unie (1979a).

Characteristic	Date of sampling					
	6-6	5-7	3-8	4-9	16-10	26-11
Water (%)	66	66	65	61	64	62
Organic matter (%)	12.5	9.4	11.5	9.7	12.2	10.6
Total VFA (g/l)	2.5	1.4	0.2	0.06	< 0.01	0.02
pH-KCl	—	7.4	7.8	7.9	7.7	7.2
N-total (%)	0.16	0.15	0.16	0.16	0.18	0.16

Mud storage field

Table 5 presents the results of the monthly performed analyses in the mud storage field. The endpoint of the anaerobic digestion was reached after approximately one year. After dewatering in March 1979 ripening started immediately. Chemical ripening did not produce a bad odour (Suiker Unie, 1979). Apparently, as long as the overlying water has a sufficiently high O₂ content to maintain aerobic digestion the 'submerged mud' method can be applied successfully.

Evaluation. A comparison of the composition of the fresh mud used in the pot experiment, in the experimental fields, and in the mud storage field shows rather great differences (Tables 2, 4, 5). In the pot experiment the environmental conditions were much more unfavourable for methane production due to a greater amount of easily decomposable organic matter yielding more VFA and a lower pH. Treatments aimed at improvement of environmental conditions for bacteria involved in CH₄ production could, therefore, be successful in pot experiments and have no influence in field experiments.

Conclusions

This research provided insight into the parameters which control the anaerobic digestion of organic matter in sugar-beet mud and also into the fermentation process itself. Optimizing this process can be useful when environmental conditions for methane production are unfavourable, for instance in the case of high total VFA and low pH.

The following conclusions may be drawn.

- Fermentation can best be followed by measuring gas production, gas composition and volatile fatty acids concentration.
- The course of organic matter content of the mud cannot be followed by determining the loss on ignition because of errors introduced by carbonates and VFA.
- Temperature is a very critical environmental factor.
- Hydrolysis and acid formation proceed faster than CH₄ production.
- Redox potential in the mud is no limiting factor for CH₄ production.
- Addition of CaCO₃ and inoculation with old mud is only useful when a large quantity of VFA is produced (as in the pot experiment).
- NO₃ nutrition has hardly any influence on digestion rate.
- The growth rate of bacteria involved in CH₄ production (methane bacteria and/or acetogenic bacteria) depends mainly on the amount of available NH₄; at least in the pot experiment.
- Dry mud (no overlying water) has a higher digestion rate of VFA than submerged mud, presumably due to more available nitrogen.
- Refreshment of overlying water has a negative influence on fermentation rate, possibly through rinsing out of NH₄.
- Refreshment of overlying water is necessary to maintain a sufficiently high O₂ content for aerobic digestion in the water.

– The ‘submerged mud’ method has been applied successfully, without causing bad odour.

Acknowledgements

Facilities to make this research possible were provided by Suiker Unie Puttershoek and the departments of Microbiology and Water Purification, Agricultural University, Wageningen. The ‘submerged mud’ method was developed by Suiker Unie Puttershoek and Adviesburo Arnhem.

References

- Blom, J. J. C., 1978. Anaerobe afbraak van organische stof in bietentarra, deel I. Doctoraal verslag, Landbouwhogeschool, Wageningen.
- Bryant, M. P., 1976. The microbiology of anaerobic degradation and methanogenesis, with special reference to sewage. In: H. G. Schlegel & J. Barnea (Ed.), Microbial energy conversion. Pergamon Press.
- Houba, V. J. G., J. Ch. van Schouwenburg & I. Walinga, 1974. Method of analysis for soils, Soil Analysis II. M.Sc. course on soil science and water management, Agricultural University, Wageningen.
- Knaapen, J. W. P. M., 1979. Anaerobe afbraak van organische stof in bietentarra, deel II. Doctoraal verslag, Landbouwhogeschool, Wageningen.
- Lettinga, G., 1977. Anaerobe gisting en zuivering van afvalwater. Landbouwhogeschool, Wageningen.
- Spoelstra, S. F., 1978. Microbial aspects of the formation of malodorous compounds in anaerobically stored piggery wastes. Doctoral thesis, Agricultural University, Wageningen.
- Suiker Unie, 1978a. Schoon water op goede gronden, Brochure, Suiker Unie, Puttershoek.
- Suiker Unie, 1978b. Kwartaalbericht april 1978. Suiker Unie, Puttershoek.
- Suiker Unie, 1979a. Kwartaalbericht februari 1979. Suiker Unie, Puttershoek.
- Suiker Unie, 1979b. Kwartaalbericht april 1979. Suiker Unie, Puttershoek.
- Takai, Y. & T. Kamura, 1966. The mechanism of reduction in waterlogged paddy soil. *Folia Microbiol.* 11: 304-313.
- Velsen, A. F. M. van, 1977. Anaerobic digestion of piggery waste. 1. The influence of detention time and manure concentration. *Neth. J. agric. Sci.* 25: 151-169.
- Vletter, R. de, 1980. De bestrijding van de waterverontreiniging bij een suikerindustrie. *Pt/Procestechniek* 35: 1-9.
- Willet, J. R., N. P. Christen & A. W. van Nes, 1976. Bezinkvelden bij suikerfabrieken. *Pt/Procestechniek* 31: 483-490.