

Biogas production technology: An Indian perspective

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Biogas technology provides an alternate source of energy in rural India, and is hailed as an archetypal appropriate technology that meets the basic need for cooking fuel in rural areas. Using local resources, viz. cattle waste and other organic wastes, energy and manure are derived. Realization of this potential and the fact that India supports the largest cattle wealth led to the promotion of National Biogas Programme in a major way in the late 1970s as an answer to the growing fuel crisis. Biogas is produced from organic wastes by concerted action of various groups of anaerobic bacteria. An attempt has been made in this review on the work done by our scientists in understanding the microbial diversity in biogas digesters, their interactions, factors affecting biogas production, alternate feedstocks, and uses of spent slurry.

MICROBIAL conversion of organic matter to methane has become attractive as a method of waste treatment and resource recovery. This process is anaerobic and is carried out by action of various groups of anaerobic bacteria.

Three basic points about this process are:

- (i) that most of the important bacteria involved in biogas production process are anaerobes and slow growing;
- (ii) that a greater degree of metabolic specialization is observed in these anaerobic microorganisms; and
- (iii) that most of the free energy present in the substrate is found in the terminal product methane. Since less energy is available for the growth of organism, less microbial biomass is produced and, consequently, disposal of sludge after the digestion may not be a major problem.

The various microbial groups involved in the flow of carbon from complex polymers to methane-based model described by McInerney and Bryant¹ have been discussed.

Complex polymers are broken down to soluble products by enzymes produced by fermentative bacteria (Figure 1, Group 1) which ferment the substrate to short-chain fatty acids, hydrogen and carbon dioxide. Fatty acids longer than acetate are metabolized to acetate by obligate hydrogen-producing acetogenic bacteria (Figure 1, Group 2). The major products after digestion of the substrate by these

two groups are hydrogen, carbon dioxide, and acetate. Hydrogen and carbon dioxide can be converted to acetate by hydrogen-oxidizing acetogens (Figure 1, Group 3) or methane by carbon-dioxide-reducing, hydrogen-oxidizing methanogens (Figure 1, Group 4). Acetate is also converted to methane by aceticlastic methanogens (Figure 1, Group 5). Nearly seventy per cent of methane from biogas digesters fed with cattle dung is derived from acetate^{2,3}. Representative reactions occurring in biogas digester and their free energy under standard and typical conditions are given in Table 1.

Interactions between the various microbial groups

Microbial diversity in biogas digesters is as great as that of rumen⁵ wherein seventeen fermentative bacterial species have been reported to play important role⁶ for production of biogas. Furthermore, it is the nature of the substrate that determines the type and extent of the fermentative bacteria present in the digester⁷. Ramasamy *et al.*³ reported higher presence of proteolytic organisms in cow dung-fed digesters and other animals waste-fed digesters. However, Preeti Rao *et al.*⁸ observed that while cow dung-fed digesters supported higher amylolytic microorganisms, poultry waste-fed digesters showed higher proteolytic population. Among fermentative organisms, *Bacteroides succinogens*, *Butyrivibrio fibrisolvens*

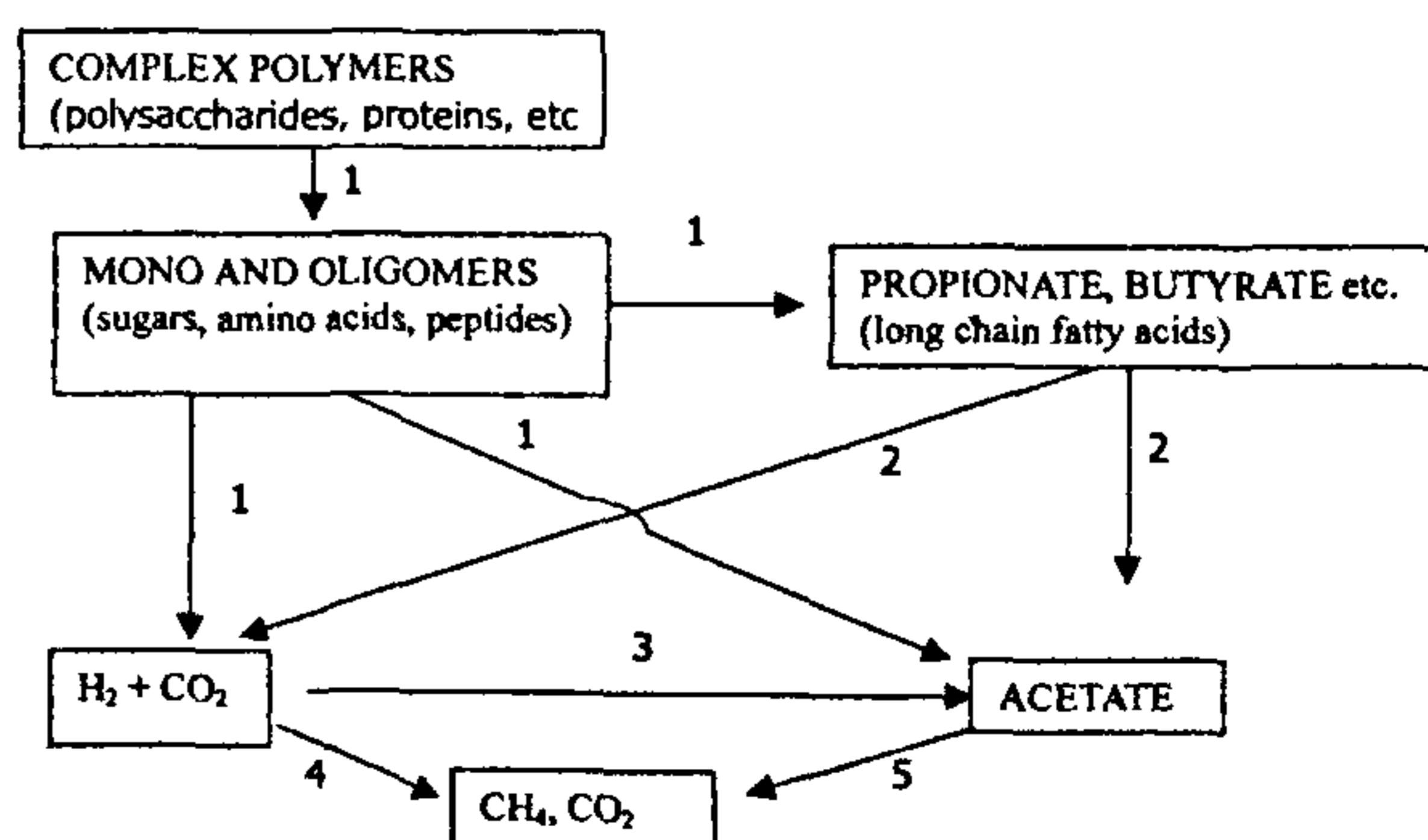


Figure 1. Microbial groups involved in biogas production. Group 1, fermentative bacteria. Group 2, obligately hydrogen-producing acetogenic bacteria. Group 3, hydrogen-consuming acetogenic bacteria. Group 4, carbon-dioxide-reducing methanogens Group 5, aceticlastic methanogens.

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Table 1. Representative reactions occurring in a biogas digester and their free energy under standard and typical conditions*

Representative reactions	Products	Free energy (kJ/mol)	
		ΔG°	$\Delta G'$
Glucose + 3 H ₂ O	3 CH ₄ + 3 HCO ₃ ⁻ + 3 H ⁺	- 403.6	- 399.1
Glucose + 4 H ₂ O	2 CH ₃ COO ⁻ + 2 HCO ₃ ⁻ + 4 H ⁺ + 4 H ₂	- 206.3	- 318.5
CH ₃ COO ⁻ + H ₂ O	CH ₄ + HCO ₃ ⁻ + H ⁺	- 31.0	- 24.5
4 H ₂ + HCO ₃ ⁻ + H ⁺	CH ₄ + 3 H ₂ O	- 135.6	- 31.6
4 H ₂ + 2 HCO ₃ ⁻ + H ⁺	CH ₃ COO ⁻ + 2 H ₂ O	- 104.6	- 7.0
Butyrate + 2 H ₂ O	2 CH ₃ COO ⁻ + H ⁺ + 2 H ₂	+ 48.1	- 17.4
Propionate + 3 H ₂ O	CH ₃ COO ⁻ + HCO ₃ ⁻ + H ⁺ + 3 H ₂	+ 76.1	- 5.4
Benzoate + 7 H ₂ O	3CH ₃ COO ⁻ + HCO ₃ ⁻ + 3 H ⁺ + 3 H ₂	+ 89.7	- 15.7

*Standard conditions are solutes, 1 molar; gases, 1 atmosphere; 25°C; pH 7.0. Typical conditions for an anaerobic digester were estimated to be 37°C and pH 7.0, and the concentrations of products and reactants were as follows: Glucose and benzoate, 10 micromolar; acetate, butyrate, propionate, 1 millimolar; HCO₃⁻, 20 millimolar; CH₄, 0.6 atmosphere; and H₂, 10⁻⁴ atmosphere (Thauer *et al.*⁴).

Clostridium cellobioparum, *Ruminococcus albus* and *Clostridium* sp. were predominant⁹. Ramasamy *et al.*⁵ observed that a clear differentiation existed in the type of cellulolytic bacterial distribution in rumen and biogas digester. Whereas in rumen, *Ruminococcus* sp. alone accounted for 60 per cent of the total population, in the biogas digester the predominant species belonged to the genera *Bacteroides* and *Clostridium* rather than the genus *Ruminococcus*. Later Ramasamy¹⁰ reported that *Ruminococcus flavefaciens*, *Eubacterium cellulosolvens*, *Clostridium cellulosolvens*, *Clostridium cellulovorans*, *Clostridium thermocellum*, *Bacteroides cellulosolvens* and *Acetivibrio cellulolyticus* were some of the other predominant fermentative bacteria present in cattle dung-fed digesters.

Most of these bacteria adhere to the substrate prior to extensive hydrolysis. Ramasamy¹¹ showed that while the digester slurry contained higher cellulolytic population, the outlet of the digester recorded the least cellulolytic population. Furthermore, Ramasamy *et al.*³ reported that the particulate-bound cellulolytic bacteria were the predominant group in the slurry of the digester. It was observed that out of the total cellulolytic population of 42 × 10⁴ ml⁻¹ of slurry, the particulate-bound bacteria accounted for 34 × 10⁴ ml⁻¹ of slurry. Furthermore, the particulate-bound bacteria predominated up to 20th day of initiation of biogas digester. It is also known that the particulate-bound bacteria showed direct relation to the biogas yield from the digester¹⁰ (Figure 2). In a recent review, Salom Gnana Thanga and Ramasamy¹² elaborated the need for adherence of anaerobic bacteria in cellulose hydrolysis.

Kelkar *et al.*¹³ compared the activity of cellulolytic clostridia isolated from cattle dung-fed digesters and reported that *C. populeti* recorded higher degradation of cellulose than *C. cellobioparum*, and *Clostridium* sp. Sivakumaran *et al.*¹⁴ characterized the cellulase enzymes

present in biogas digesters and reported that *Acetivibrio* sp. showed higher cellulase activity than *Bacteroides* sp. and *Clostridium* sp. isolated from biogas digesters.

Though a variety of products are formed by the action of fermentative bacteria, volatile fatty acids are the primary products of carbohydrate fermentation in biogas digesters, as they are in rumen. The partial pressure of hydrogen can influence the products of carbohydrate metabolism¹. The partial pressure of hydrogen can be maintained either by hydrogen-oxidizing methanogens or sulphate-reducing bacteria. However in biogas digesters, the action of former organisms is preferred resulting in methane as the endproduct. Under these conditions, oxidation of NADH and the conversion of hexose to acetate, H₂ and CO₂ by fermentation occurs, yielding 4 ATP molecules per hexose molecule by glycolysis or acetyl phosphate pathway (Thauer *et al.*⁴). But under conditions of higher partial pressure of hydrogen, formation of more reduced products results in the following order: propionate, butyrate, ethanol, and lactate. Also, the fermentation of hexose either to ethanol or lactate yields only 2 ATP per hexose molecule by glycolysis¹, depriving thereby methanogens of the substrate (acetate) needed for its growth and activity¹⁵. Ramasamy *et al.*³ studied the interaction of cellulolytic bacteria, *Acetivibrio* sp., and methanogens, *Methanosarcina* sp., and *Methanobacterium* sp., using cellulose and cellobiose as substrate. They observed that using co-cultures, the growth of both *Acetivibrio* sp. and *Methanosarcina* sp. was higher, and that the methane content of biogas was enhanced by twenty per cent.

Though less in number, obligately hydrogen-producing acetogenic bacteria are one of the important groups in biogas digesters. These organisms oxidize the fatty acids that are longer than acetate to acetate and thereby release

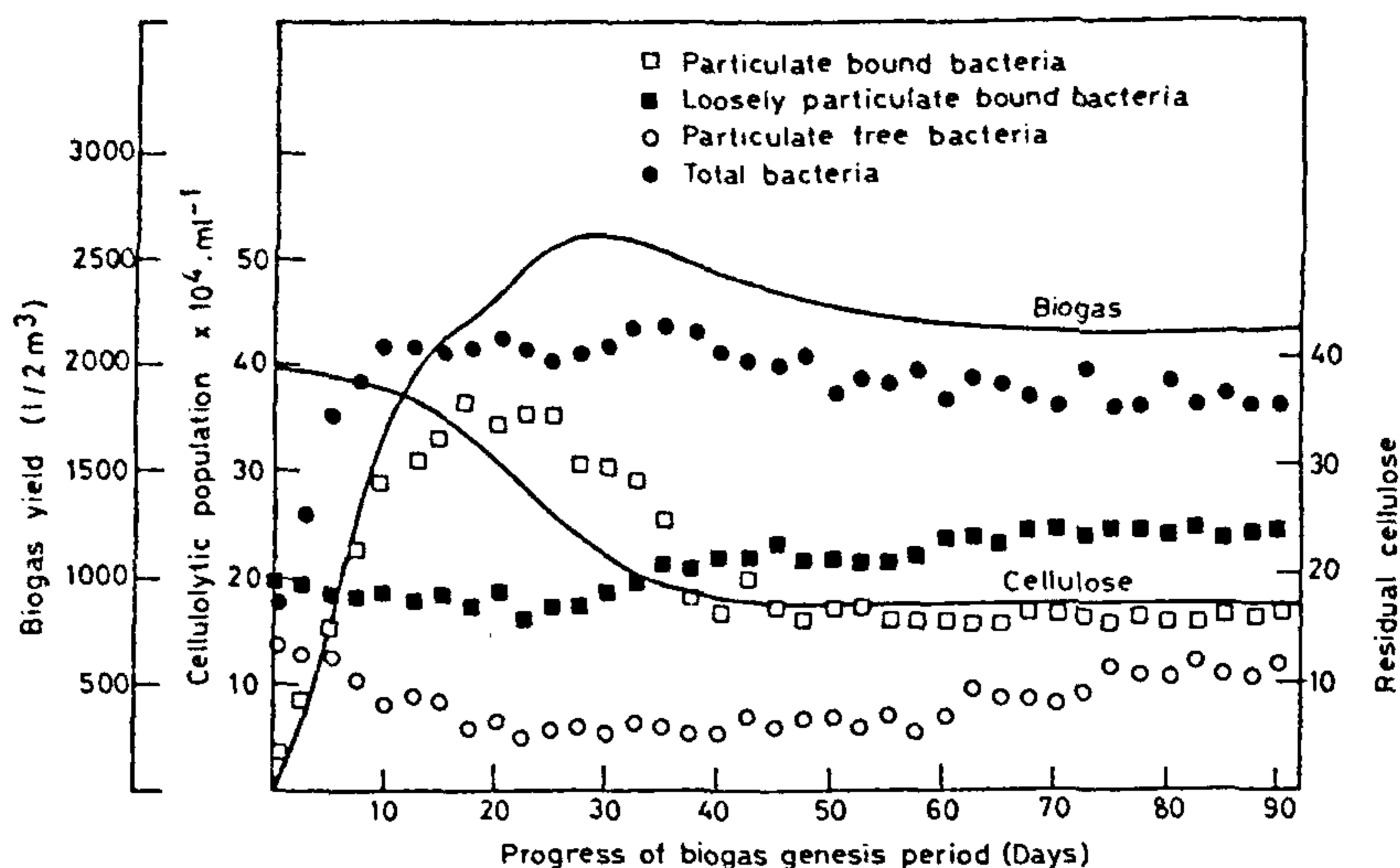


Figure 2. Particulate-bound bacteria and their relation to biogas production.

energy from the substrate in the form of methane. Boone and Bryant¹⁶ isolated *Syntrophobacter wolinii* which beta-oxidized propionate to acetate, and later McInerney *et al.*¹⁷ isolated *Syntrophomonas wolfei* which beta-oxidized C₄ to C₇ fatty acids. But these reactions are favourable only if the hydrogen partial pressure is below 10⁻³ atmosphere. However, for propionate oxidation the partial pressure has to be still lower¹⁷. Nagamani *et al.*¹⁸ reported that accumulation of propionic acid and lauric acid resulted in acidification of digester which inhibited further methane formation in castor-oil-cake-fed digesters. Furthermore, they reported¹⁹ that propionate is toxic to methanogens even at a concentration of 8 mM. Subsequently, Nagamani *et al.*²⁰ developed a consortia comprised of syntrophic co-cultures in association with hydrogen-utilizing methanogens, which stabilized methane production from castor-oil-cake-fed digester. Earlier, Meher and Ranade²¹ isolated a propionate-degrading bacterium in association with *M. formicicum* from cattle dung-fed digesters. Meher *et al.*²² also reported the presence of butyrate-degrading syntrophic co-culture in biogas digesters fed with cattle dung. Though these organisms occurred at a pH < 6.0 and below 45°C, methanogenesis was observed at pH > 6.5 and above 40°C.

Reductive halogenation of various halogenated aliphatic and aromatic compounds is the next important process by fermentative group of organisms. Thauer *et al.*⁴ observed that these reactions are thermodynamically feasible and can support growth. Anaerobic degradation of aromatic compounds also depend on hydrogen-consuming bacteria⁷. Mountford and Bryant²³ isolated a bacterium which oxidized benzoate to acetate in obligate co-culture with methanogens or sulphate-reducing bacteria. Doraisamy *et al.*²⁴ showed that phenol

at 1000 ppm concentration inhibited the gas production up to six weeks, which increased with prolonged incubation. However, a mixed culture consortia enriched with phenol utilized the phenol immediately, showing an increase in gas production without any lag phase. Earlier, Doraisamy *et al.*²⁵ studied the anaerobic degradation of aromatic compounds and observed that benzoic acid was the major intermediary product, which was further metabolized to acetate and butyrate. Kalaichelvan²⁶ screened various anaerobic bacteria for their potential for degradation of aromatic compounds. He reported that out of the 17 compounds tested, 14 were amenable for anaerobic degradation and more than 50 per cent of the concentration of these compounds disappeared in 14 days.

Hydrogen-consuming acetogenic bacteria are the minor groups involved in fermentative reactions in biogas digesters. Mackie and Bryant²⁷ reported that their activity in the formation of acetate by reduction of CO₂ in cattle dung-fed digesters accounted for less than 5 per cent of the total acetate formed. However, a substantial activity by these group of organisms should definitely increase methane formation, as acetate is the preferred substrate for *Methanosarcina barkeri*, the predominant methanogen in biogas digester in cattle waste-fed digester³. However, Ranade *et al.*²⁸ reported that *Methanobacterium formicicum* was the predominant methanogenic bacteria in cattle dung-fed digesters, followed by *Mb. ruminantium*.

Methanogens possess very limited metabolic repertoire, using only acetate or C₁ compounds (H₂ and CO₂, formate, methanol, methylamines or CO), with methane being the endproduct of the reaction. Of the methanogenic genera, *Methanosarcina* sp. and *Methanosaeta* sp. form methane by aceticlastic reaction. Whereas the apparent K_m for methane formation from acetate for *Methanosaeta* sp. was under 1 mM, for *Methanosarcina* sp. it was

3–5 mM (ref. 8). Therefore, while faster-growing *Methanosarcina* sp. are predominant in high-rate, shorter-retention digesters wherein acetate concentration is higher, *Methanosaeta* sp. are predominant in low-rate, slow-turnover digesters. Pathway of methane formation by *Methanobacterium thermoautotrophicum* is given in Figure 3.

Both carbon-dioxide-reducing and aceticlastic-methanogens play an important role in maintaining stability of the digester. The failure in a biogas digester can occur if carbon dioxide-reducing methanogens fail to keep pace with hydrogen production. Whereas apparent K_m for hydrogen consumption in methanogenic environments is near 10^{-2} atmosphere²⁹, hydrogen consumption must be below 10^{-3} atmosphere for oxidation of fatty acid to acetate, and thus the carbon dioxide-reducing methanogens in a biogas digester are greatly undersaturated for hydrogen³⁰. Despite this, hydrogen in biogas digester can build up rapidly to levels inhibitory to methanogenesis due to failure of the activity of hydrogen-scavenging organisms, shifting the fermentation products away from acetate. Moreover, failure of aceticlastic methanogens to keep up with acetic acid production results in the accumulation of fatty acids, resulting thereby in failure of the digester. Maheswari *et al.*³¹ developed a simple test kit that can be used at field level to detect the problems of microbiological process during anaerobic digestion of organic wastes to methane.

Factors affecting biogas production

Various factors such as biogas potential of feedstock, design of digester, inoculum, nature of substrate, pH, temperature, loading rate, hydraulic retention time (HRT), C : N ratio, volatile fatty acids (VFA), etc. influence the biogas production.

Meher *et al.*³² reported that the performance of floating dome biogas plant was better than the fixed dome biogas

plant, showing an increase in biogas production by 11.3 per cent, which was statistically significant. Furthermore, the observed reduction in biogas yield was due to the loss of gas from the slurry-balancing chambers of fixed dome plant. Dhevagi *et al.*³³ used different feedstocks like cow dung, buffalo dung, dry animal waste, stray cattle dung, goat waste, and poultry droppings for their biomethanation potential and observed that poultry droppings showed higher gas production. Earlier Yeole and Ranade³⁴ compared the rates of biogas yield from pig dung-fed and cattle dung-fed digesters and reported that the biogas yield was higher in the former. They attributed this higher biogas yield to the presence of native microflora in the dung. Shivraj and Seenayya³⁵ reported that digesters fed with 8 per cent TS of poultry waste gave better biogas yield, and attributed the lower yield of biogas at higher TS levels to high ammonia content of the slurry.

Dhevagi *et al.*³³ tested the efficiency of different inoculum sources for biomethanation. Use of goat rumen fluid as inoculum at the rate of 8 per cent (v/v) was more efficient in production of biogas. Furthermore, the digesters inoculated with goat rumen fluid showed higher population of cellulolytic anaerobic bacteria than other inocula tested.

For increased gas yield, a pH between 7.0 and 7.2 is optimum, though the gas production was satisfactory between pH 6.6 and 7.6 as well. The pH of the digester is a function of the concentration of volatile fatty acids produced, bicarbonate alkalinity of the system, and the amount of carbon dioxide produced³⁶. Sahota and Ajit Singh³⁷ reported that the gas production was significantly affected when the pH of the slurry decreased to 5.0. They observed that apart from the decreased methanogenic activity due to lower pH, the population of cellulolytic bacteria, amylolytic organisms, and proteolytic organisms reduced by 4 and 2 log order, respectively. Most of the digesters in our country operate normally at ambient conditions. Northern India records a shortfall in biogas output during winters³⁶, and in some other parts of the country, especially in dry tracts, too the digester performance is affected due to higher temperatures³⁷. Nagamani and Ramasamy³⁸ observed that though there was higher production of biogas at 55°C, the process was unstable due to higher production of volatile fatty acids and that specific microbial consortia was needed for biomethanation of cattle waste at 55°C.

In the case of C : N ratio, 25–30 : 1 is optimum for biogas production³⁹. Ramasamy⁴⁰ observed that the optimum loading rate of the feedstock varied as per its nature, and likewise HRT varied as per the loading rate. Earlier Yeole and Ranade³⁴ reported that an HRT of 14 days was optimum for biogas production from cow dung. Gadre *et al.*⁴¹ investigated the optimum retention time for the production of biogas from cattle dung and reported that 15 d HRT was the best for maximum production of biogas from cow dung. They further observed that shorter

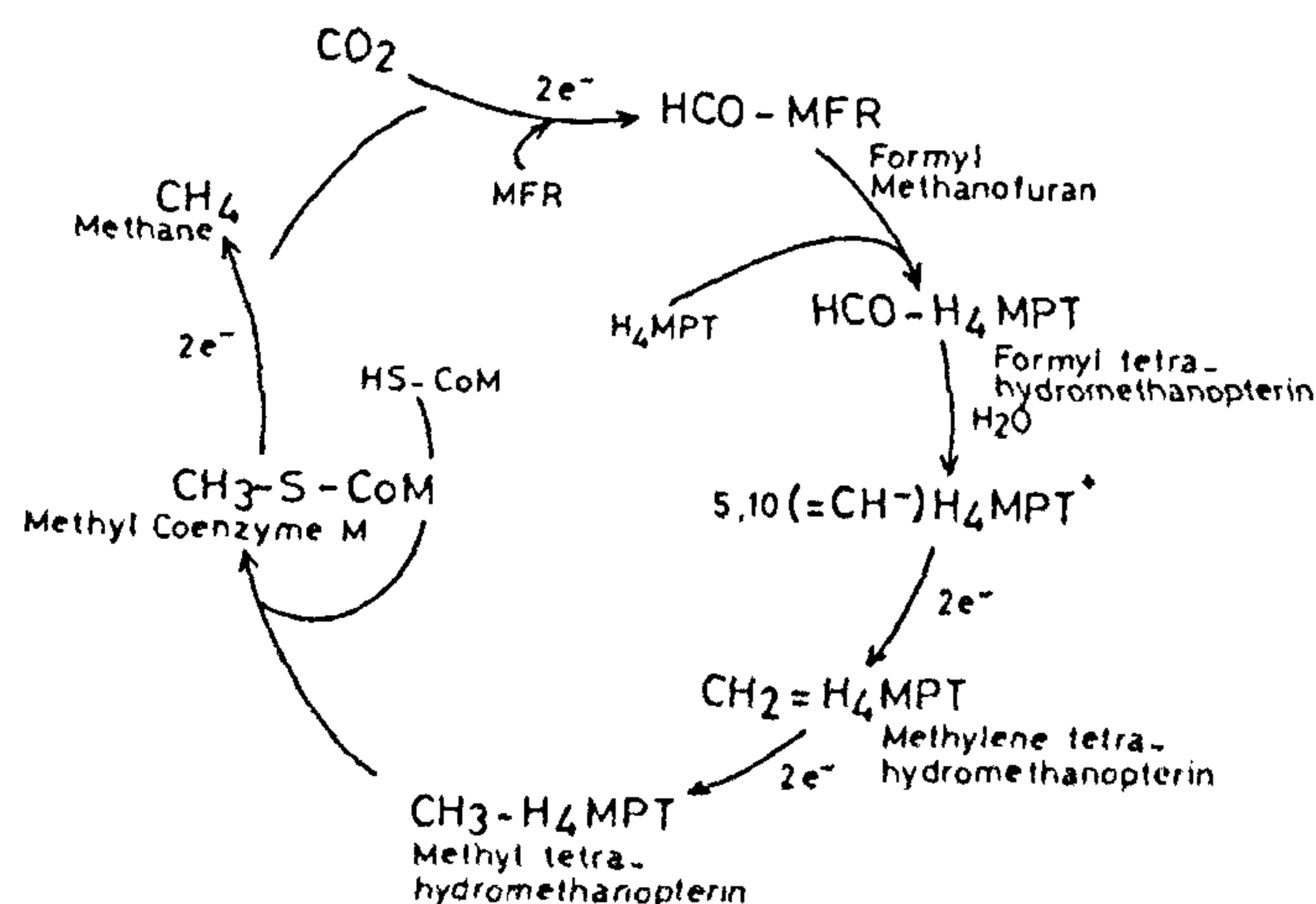


Figure 3. Pathway of methane formation by *Methanobacterium thermoautotrophicum*.

HRT resulted in accumulation of VFA, whereas at HRT longer than 15 d the digester components were not fully utilized. Ranade *et al.*⁴² studied the influence of different TS content of biogas production and reported that the optimum production was observed at 8 per cent TS. However, the methane content of the gas produced did not vary significantly with varying levels of TS. Hence, they suggested that high TS content of 14% cattle dung (2 : 1 dilution) can be followed in areas during the time of water scarcity rather than discontinuing the feeding of the digester.

For a normal anaerobic fermentation process, concentration of volatile fatty acids in terms of acetic acid should not exceed 2000–3000 mg l⁻¹ (ref. 43). Hajarnis and Ranade⁴⁴ reported that the toxicity of *n*-butyrate, *n*-valerate, and *n*-caproate was more on *Methanobacterium bryantii*, *Methanobacterium formicicum*, and *Methanosarcina barkeri* than iso-forms of these acids. Ramasamy *et al.*⁴⁵ and Nagamani *et al.*⁴⁶ observed that even a concentration of 8 mM of propionate and 20 mM caproic acid completely inhibited the activity of both *Methanosarcina* sp. and *Methanobacterium* sp., thereby affecting the biogas production. Earlier Ramasamy *et al.*³ observed that caproic acid accumulation was toxic to methanogenesis from cow dung-fed digesters.

Design of digester and distribution of anaerobic microorganisms

Floating gas holder type and fixed dome type are the two tested designs, that are being widely employed. Though economics of the model and their suitability are well documented, not much work is done on the microbiological aspects of these digesters. Kalaichelvan *et al.*⁴⁷ studied the relationship between the different digesters, namely, KVIC steel drum, Ganesh model, Deenabandhu, and Gayathri model biogas plants, and the microbiological aspects related to biogas production. They observed that though there was no significant variation in biogas production between the different models tested, Deenabandhu model biogas plant harboured higher methanogenic population than other model biogas plants. In general, fixed dome digesters showed higher microbial distribution of all trophic levels than floating drum type biogas digesters.

Effect of metals on biogas production

Presence of some metals also influences the biogas production. Seenayya *et al.*⁴⁸ studied the influence of various metals on biogas production. They observed that addition of calcium (5 mM), cobalt (50 µg g⁻¹ TS), iron (50 mM), magnesium (7.5 mM), molybdenum (10–20 mM), nickel (10 µg g⁻¹ TS) individually as well as in combination enhanced the biogas production and

attributed this to the increased methanogenic population in the digesters. Geetha *et al.*⁴⁹ later observed that addition of nickel at 2.5 ppm increased the biogas production from digesters fed with water hyacinth and cattle-waste blend and attributed this to higher activity of nickel-dependent metallo-enzymes involved in biogas production. Jain *et al.*⁵⁰ also reported that Cd and Ni at 600 and 400 µg g⁻¹ of dry matter, respectively, increased the biogas production and methane content. But they further observed that iron or manganese at 1100 µg g⁻¹ of dry matter did not influence the yield of biogas. However, Preeti Rao and Seenayya⁵¹ observed that addition of iron as ferrous sulphate at 50 mM level showed faster bioconversion of both the cow dung and poultry waste. They further reported that addition of iron at 20 mM level increased the population of methanogens, and that methanogenesis was also enhanced by 40 and 42 per cent in cow dung-fed digesters as well as in a poultry waste-fed digester, respectively. In the case of cobalt, Jarvis *et al.*⁵² observed that addition of cobalt (0.2 mg l⁻¹) improved the gas yield and methane content of gram clover silage-fed digester. Furthermore they reported that addition of cobalt helped to achieve a higher organic loading rate of 7.0 g VS l⁻¹ d⁻¹ in a period of 2 days. Whereas without cobalt addition even a organic loading rate of 5.0 g VS.l⁻¹.d⁻¹ was achieved only after 70 days of operation. Raju *et al.*⁵³ observed that addition of cobalt, nickel, and iron increased the biogas production from mango peel-fed digester which was several folds higher than the control. Singh *et al.*⁵⁴ observed that addition of borax and diborane at 0.2 g/l increased the gas production from digesters fed with water hyacinth as the substrate.

Ammonia on methanogens and methanogenesis

Hajarnis and Ranade⁵⁵ reported that the methanogens required 5 mM NH₃N for its maximum specific growth rate and minimum doubling time. But, at the same time exposure to higher levels of ammonia was toxic to methanogens. They also observed that *Ms. barkeri* was sensitive to the presence of ammonia than *Mb. bryantii* and that the revival of growth and activity was better in the case of *Mb. bryantii*⁵⁶ and *Mb. formicicum*⁵⁷. They further observed that diluting the digester slurry to a level of < 450 mM ammonia helped in re-establishment of microflora in the digester than temporary discontinuation of feeding.

Alternate feedstocks

Animal wastes are generally used as feedstock in biogas plants and their potential for biogas production is given in Table 2. But, the availability of these substrates is one of the major problems hindering the successful operation

Table 2. Potential biogas production from different feedstocks⁶⁰

Feedstock	Availability (kg animal ⁻¹ d ⁻¹)	Gas yield (m ³ kg ⁻¹)
Cattle waste	10	0.36
Buffalo waste	15	0.54
Piggery waste	2.25	0.18
Chicken waste	0.18	0.011
Human excreta	0.4	0.028

of biogas digesters. Khendelwal⁵⁸ reported that the availability of cattle waste can support only 12–30 million family-size biogas plants against the requirement of 100 million plants. A significant portion of 70–88 million biogas plants can be run with fresh/dry biomass residues. Of the available 1,150 billion tons of biomass, a fifth would be sufficient to meet this demand⁵⁹.

Many workers have explored various substrates for biogas production. For biogas production, the two most important parameters in the selection of particular plant feed stocks are the economic considerations and the yield of methane for fermentation of that specific feedstock⁶¹. They assessed the methane yield from fresh-water aquatics, forage grasses, roots and tubers, and marine species and reported that highest yield was obtained from root crops followed by forage grasses, and fresh-water aquatics. Marine species yielded the lowest yield of methane⁶². Chynoweth *et al.*⁶³ postulated that methane yield and kinetics were generally higher in leaves than in stems. Results of Sharma *et al.*^{64,65} confirmed the above concept, using *Ipomoea fistulosa* as substrate. Observations on biogas production from food industry wastes⁶⁶ and from different biomass⁶⁷ have shown that pretreatment of feedstock improved the biogas yield and methane content from biomass-fed digesters. Gunaseelan⁶⁷ observed that though the agricultural residues, such as straw, need to be reduced to a particle size of 0.4 mm for better biogas production, succulent leaves such as *Mirabilis* sp., *Ipomoea fistulosa*, etc. can be fed without any shredding. Ramasamy *et al.*⁶⁸ evaluated the biogas potential of *Gliricidia* sp., *Albizia* sp., and *Parthenium* sp. in plug flow digester and observed that biogas yield from these feedstocks improved with specific inoculum. Gunaseelan⁶⁹ reported that pretreatment with NaOH increased the gas production from parthenium-fed digesters (0.46 m³ m⁻³ d⁻¹ and the methane yield was 0.11 m³ kg⁻¹ VS). Earlier Deshpande *et al.*⁷⁰ and Mallick *et al.*⁷¹ reported the use of water hyacinth as alternate substrate together with cattle waste for biogas digesters. Chanakya *et al.*⁷² developed horizontal plug flow (PF) digester and solid state stratified bed (SSB) digester for biomethanation of biomass and observed that SSB performed better in terms of stability and gas production.

Radhika *et al.*⁷³ reported that a mix of coir pith and cattle waste at 3 : 2 ratio gave a better gas output and that the methane content was in the range of 80–85 per cent. Later, Jagadeesh⁷⁴ reported that calcium-hydroxide-

treated coir pith mixed with cattle waste slurry to adjust to a total solids content of 9.2 per cent gave a biogas yield of 0.152 m³ kg⁻¹ of dry matter d⁻¹. However, Deivanai and Kasturi Bai⁷⁵ observed that biomethanation of coir pith was slow, attributing this to higher lignin content of the waste.

Dar and Tandon⁷⁶ reported a 62 per cent higher yield in biogas fed with NaOH-pretreated *Lantana camera* along with cattle waste. Deoiled castor-oil cake as an alternate feedstock at a loading rate of 8 kg TS m⁻³ d⁻¹ was reported by Gollakota and Meher⁷⁷. Nagamani *et al.*¹⁸ also studied the potential of biogas production from different deoiled cakes like castor, neem, groundnut, and coconut and reported that accumulation of long-chain fatty acids, and propionate after four weeks of digestion inhibited the methanogenic process.

Sharma *et al.*⁶⁴ observed that size-reduced (particle size 0.4 mm) banana peel gave better gas production. Mallick *et al.*⁷¹ reported the use of *Cannabis* for biogas production; but observed that use of fresh *Cannabis* at 31% completely stopped the biogas production due to the higher presence of alkaloids. Jagadeesh *et al.*⁷⁸ observed that slacked lime pretreatment and partial aerobic decomposition for 6 days removed the methanogenic inhibitors, improving the digestibility of *Eupatorium odoratum*. Kalia and Kanwar⁷⁹ reported that the partially decomposed (5 days aerobic decomposition) *Ageratum* can be used as substrate together with cattle waste for biogas production. Farmers of Jodhpur, Rajasthan, successfully used tea leaf waste as a feedstock, which yielded 12 cu.feet gas kg⁻¹ of waste. Conventional biogas digesters require 10 kg of cattle waste to produce an equivalent quantity of biogas⁸⁰. Sahota and Rajinder⁸¹ reported that addition of rice husk soaked in water at 20 per cent level to cattle dung digester increased the gas production.

Abbasi *et al.*⁸² studied the biogas potential of eight aquatic weeds and reported that *Salvinia* and *Ceratopteris* yielded a biogas output as high as 0.2 m³ kg⁻¹ VS. Later Abbasi and Nipanay⁸³ reported that addition of inoculum sustained biogas production from *Pistia* for 10 days. Balasubramanian and Kasturi Bai⁸⁴ suggested that *Wolffia* and *Lemna* grown in slurry of cow dung-fed digesters can be effectively used for biogas production together with cow dung.

In the case of fruit wastes, yields of 0.62, 0.56, 0.77, 0.72 and 0.63 m³ of biogas/kg of VS were obtained from tomato, mango, pineapple, lemon, and orange processing waste, respectively⁸⁵. Mango peel supplemented with urea to adjust the C : N ratio to 20–30 : 1 resulted in the stability of the digester⁶⁶. He further reported that addition of nitrogen in the form of silkworm waste and oilseed extracts, such as neem and castor, increased the methane content. Viswanath *et al.*⁸⁶ investigated the influence of successive addition of fruit and vegetable solid wastes on the performance of biogas digester and

reported that the digester was stable at a loading rate of $3.8 \text{ kg VS m}^{-3} \text{ d}^{-1}$. They further observed that minor manipulation in nutritional and operational parameters helped in the functioning of the digester fed with different fruits and vegetable wastes (mango, pineapple, tomato, jack fruit, banana, and orange) for a considerably long time without any noticeable changes in the rates and yields of biogas. Pilot plant (1.5 m^3 KVIC digester) studies with mango peel showed that supplementation with essential nutrients improved the digestibility of feedstock, yielding as high as $0.6 \text{ m}^3/\text{kg VS}$ with a biogas content of 52% at a loading rate of 8–10%. Also, addition of sugarcane filter mud at a rate of 200 g/4 kg of mango peel in 1.5 m^3 digester increased biogas yield substantially with a methane content of 60%. Krishnanand⁶⁶ also reported that addition of additives such as extract of nirmali seeds, hybrid beans, black gram, and guar gum seeds at 2–3% level increased the biogas production significantly, and attributed this increase to the galactomannan constituent of the leguminous seeds which increased the floc formation, thereby retaining the organisms in the digester.

Sarada and Joseph⁸⁷ studied the microbiology of digesters fed with tomato-processing waste. They observed that in batch digestion, the population of methanogens was less due to the drop in pH of slurry.

However in semi-continuous digestion, the population of cellulolysers, xylanolysers, pectinolysers, proteolysers, lipolysers, and methanogens increased with increase in hydraulic retention time (HRT) and reported a biogas yield of $0.42 \text{ m}^3 \text{ kg}^{-1} \text{ VS}$. Earlier Mahadevaswamy and Venkataraman⁸⁸ investigated the feasibility of mango processing waste for biogas production and reported a biogas output of $0.21 \text{ m}^3 \text{ kg}^{-1} \text{ TS}$. A summary of selected alternate feedstocks and their potential for biogas production is given in Table 3.

Other wastes

Yeole *et al.*⁹¹ reported the feasibility of using industrial canteen waste as a feedstock in biogas digesters. They suggested a reduction in particle size of the waste below 2 cm, and feeding at the rate of 8–10 TS, for successful operation. Biomethanation potential of market waste was studied by Ranade *et al.*⁹² and they reported that the digestion process was stable at 20 days HRT with 48% reduction in VS and with biogas production of $35 \text{ litre kg}^{-1} \text{ d}^{-1}$. They also studied the biogas production from solid waste that originated from biscuit and chocolate industry. They reported that 40 d HRT was optimum for biogas production of 466 l kg^{-1} of waste d^{-1} with 57% methane and 65% VS distribution. They reported that with lower

Table 3. Selected alternate feedstocks and their potential for biogas production

Feedstock	Hydraulic retention time	Organic loading rate ($\text{kg VS m}^{-3} \text{ d}^{-1}$)	Methane yield ($\text{m}^3 \text{ kg}^{-1} \text{ VS}$)	Reference
Fruit wastes	8	3.8	0.030	Viswanath <i>et al.</i> ⁸⁶
	16	3.8	0.250	
	20	3.8	0.320	
	24	3.8	0.190	
	16	5.7	0.110	
	16	7.6	0.040	
	16	9.5	0.420	
Tomato processing waste	24	4.3	0.420	Sarada and Joseph ⁸⁷
Banana peeling				Sharma <i>et al.</i> ⁶⁴
0.088 mm size	NA	NA	0.408	
0.40 mm size	NA	NA	0.409	
1.0 mm size	NA	NA	0.396	
6.0 mm size	NA	NA	0.374	
30 × 10 mm size	NA	NA	0.271	
Cauliflower waste				Sharma <i>et al.</i> ⁶⁴
0.088 mm size	NA	NA	0.423	
0.40 mm size	NA	NA	0.423	
1.0 mm size	NA	NA	0.423	
6.0 mm size	NA	NA	0.407	
30 × 10 mm size	NA	NA	0.358	
Mirabilis leaves				Sharma <i>et al.</i> ⁶⁴
0.088 mm size	NA	NA	0.339	
0.40 mm size	NA	NA	0.341	
1.0 mm size	NA	NA	0.329	
6.0 mm size	NA	NA	0.327	
30 × 10 mm size	NA	NA	0.290	

Table 3. (Contd.)

Feedstock	Hydraulic retention time	Organic loading rate (kg VS m ⁻³ d ⁻¹)	Methane yield (m ³ kg ⁻¹ VS)	Reference
<i>Ipomoea</i> sp.				
0.088 mm size	NA	NA	0.429	Sharma <i>et al.</i> ⁶⁴
0.40 mm size	NA	NA	0.427	
1.0 mm size	NA	NA	0.421	
6.0 mm size	NA	NA	0.413	
30 × 10 mm size	NA	NA	0.387	
Bermuda grass				
0.088 mm size	NA	NA	0.226	Sharma <i>et al.</i> ⁶⁴
0.40 mm size	NA	NA	0.228	
1.0 mm size	NA	NA	0.214	
6.0 mm size	NA	NA	0.205	
30 × 10 mm size	NA	NA	0.137	
Wheat straw				
0.088 mm size	NA	NA	0.249	Sharma <i>et al.</i> ⁶⁴
0.40 mm size	NA	NA	0.248	
1.0 mm size	NA	NA	0.241	
6.0 mm size	NA	NA	0.227	
30 × 10 mm size	NA	NA	0.162	
Paddy straw				
0.088 mm size	NA	NA	0.365	Sharma <i>et al.</i> ⁶⁴
0.40 mm size	NA	NA	0.367	
1.0 mm size	NA	NA	0.358	
6.0 mm size	NA	NA	0.347	
30 × 10 mm size	NA	NA	0.241	
<i>Ipomoea fistulosa</i> stem				
0.4 mm size	NA	NA	0.361	Sharma <i>et al.</i> ⁶⁵
0.4 mm size	NA	NA	0.182	
6.0 mm size	NA	NA	0.181	
<i>Gliricidia maculata</i> leaves	5	4.95	0.034	Gunaseelan ⁸⁹
<i>Parthenium hysterophorus</i>	10	2.48	0.117	Gunaseelan ³⁹
	20	1.24	0.115	
	40	0.62	0.101	
<i>Ageratum</i>, partially decomposed	NA	NA	0.241*	Kalia and Kanwar ⁷⁹
<i>Pistia stratiotes</i>	NA	NA	0.361	Abbasi and Nipaney ⁸³
<i>Salvinia</i> sp.	NA	NA	0.242	Abbasi <i>et al.</i> ⁸²
<i>Azolla pinnata</i>	NA	NA	0.132	Abbasi <i>et al.</i> ⁸²
	NA	NA	0.117*	
<i>Lemna minor</i>	NA	NA	0.106*	Abbasi <i>et al.</i> ⁸²
<i>Lantana camera</i>	NA	NA	0.236*	Dar and Tandon ⁷⁶
Night soil (NS)				
100% NS	NA	NA	0.500	Mohan Rao and Satyanarayanan ⁹⁰
50% NS + 50% cow dung	NA	NA	0.350	
100% Cow dung	NA	NA	0.130	

NA – Not available; *Values calculated from the reported data.

HRT, viz. 20 and 30 days, the digester showed accumulation of VFA, and the methanogenesis was inhibited⁹³. Yeole *et al.*⁹⁴ reported that higher loading rate corresponding to a shorter HRT turned the fermenting slurry of waste from liver and beef extract industry waste as acidic, and it adversely affected the biogas production. They reported, at 30 d HRT removal of 91, 89, and 74% BOD, COD and VS was attained, yielding $0.897 \text{ m}^3 \text{ kg}^{-1} \text{ TS d}^{-1}$ of biogas. Later, Yeole *et al.*⁹⁵ reported that a loading rate of $0.7 \text{ kg COD/m}^3/\text{d}$ was optimum for biogas production of $13 \text{ m}^3/\text{m}^3$ of waste/day from the liquid waste emanating from liver and beef extract industry. Yeole and Ranade⁹⁶ also studied the biogas production from mycelial waste and reported that the process was stable at 50 days HRT, yielding $0.652 \text{ m}^3 \text{ biogas m}^{-3}$ of digester. Shorter HRT of 30 and 40 days resulted in acidification of digester due to accumulation of VFA.

Chitra and Ramasamy⁹⁷ studied the potential of night soil-fed digesters for biogas production, and also assessed the survival of enteric pathogens in these digesters. Chatterjee *et al.*⁹⁸ evaluated the performance of night soil-based biogas digesters made of high density polyethylene and observed that though the waste stabilization was less, the generation of methane (0.6 m^3 of slurry d^{-1}) was higher than the conventional digester ($0.15 \text{ m}^3 \text{ m}^{-3}$ of slurry d^{-1}).

Fate of pathogens in biogas digester

Gadre *et al.*⁹⁹ reported that the pathogenic *Salmonella* sp. introduced to the cattle dung-fed digester at the start of the digestion process was eliminated in nine days of digestion. Later, Kunte *et al.*¹⁰⁰ reported that *Salmonella typhi* added to cattle dung-fed digesters as single dose at the start of the digestion was completely eliminated after 12 days of digestion with shorter retention period of 15 days, whereas it needed 26 days for complete elimination with 30 day HRT. Similarly with daily dose of the pathogen too, four-fold log reduction was observed in 15 day HRT, whereas it was only two fold with 30 day HRT. This was attributed to higher production of volatile fatty acids in digesters with shorter retention period. Earlier Chitra and Ramasamy⁹⁷ observed that enteric pathogens were eliminated after two weeks of digestion.

Kinetics of anaerobic fermentation

Several kinetic models have been developed to describe the anaerobic fermentation process. Monod¹⁰¹ showed a hyperbolic relationship between the exponential microbial growth rate and substrate concentration. In this model, the two kinetic parameters, namely, microorganisms growth rate and half velocity constant are deterministic in nature, and these predict the conditions of timing of maximum biological activity and its cessation. This model can be

used to determine the rate of substrate utilization (r_s) by the equation:

$$r_s = q_{\max} \times S_x / K + S,$$

where S is limiting substrate concentration, K is half constant, x is concentration of bacterial cells, and q_{\max} is maximum substrate utilization rate.

The above equation is applicable for low substrate concentration.

However for high substrate concentration, the equation is re-written as:

$$r_s = q_{\max} \cdot x.$$

The Monod model suffers from the drawback that one set of kinetic parameters are not sufficient to describe biological process both for short- and long-retention times, and that kinetic parameters cannot be obtained for some complex substrates. To alleviate limitations of the Monod model while retaining its advantages, Hashimoto¹⁰² developed an alternative equation, which attempts to describe kinetics of methane fermentation in terms of several parameters. According to this equation, given below, for a given loading rate S_0/θ daily volume of methane per volume of digester depended on the biodegradability of the material (B_0) and kinetic parameters μ_m and K .

$$r_v = (B_0 \times S_0/\theta) \cdot \{1 - (K/\theta \mu_m - 1 + K)\}$$

where,

r_v is volumetric methane production rate, $1 \text{ CH}_4 \text{ l}^{-1}$ digester d^{-1}

S_0 is influent total volatile solids (VS) concentration, g l^{-1}

B_0 is ultimate methane yield, $1 \text{ CH}_4 \text{ g}^{-1}$ VS added as $\theta \rightarrow \infty$

θ is hydraulic retention time d^{-1}

μ_m is maximum specific growth of microorganism d^{-1}

K is kinetic parameter, dimensionless.

Use of spent slurry

The C and N of slurry on application to soil were mineralized with increasing incubation period. A significant positive correlation was observed between the mineralization and slurry levels. Also the application of slurry improved the physical, chemical, and biological characters of the soil¹⁰³. Balasubramanian and Kasturi Bai⁸⁴ evaluated nutrient status of slurry added to the digester and observed that the total Kjeldahl nitrogen and total K were recovered after biogas production, but total P

showed a 30% decrease and ammonia N showed a 70% increase compared to the influent. A significant correlation was observed between solids and N, P, and K after anaerobic digestion.

Balasubramanian and Kasturi Bai¹⁰⁴ evaluated the growth of *Lemna* and *Wolffia* sp. in different concentration of slurry levels and reported that whereas application of slurry at 1 per cent level increased the biomass production by 150% over the initial weight, there was only 20 per cent increase in control treatment. Furthermore, they showed that biogas digester slurry can be effectively utilized for growth of aquatic plant *Wolffia*. The slurry-grown plants were rich in crude protein, K, and P, which can be recovered as animal feed¹⁰⁵.

Gnanamani and Kasturi Bai¹⁰⁶ observed that the addition of 40 t/ha slurry together with recommended dose of NPK, increased the grain yield by 80.5 per cent over control, and the cultivation of blackgram in the same plot also showed an increase in the yield of gram. They suggested that blackgram can be an ideal crop for harvesting the residual nutrient after paddy cultivation. Gnanamani and Kasturi Bai¹⁰⁷ applied the slurry @ 20 and 40 t/ha along with inorganic fertilizer supplements for rice–blackgram–rice cultivation. They reported that with 40 t/ha slurry together with recommended doses of NPK, the yield of second and third crops was comparable to conventional cultivation without requiring addition of fertilisers. Kalaichelvan and Swaminathan¹⁰⁸ reported that the spent slurry can be used as a carrier for *Rhizobium* and the shelf life was for a period of three months.

Balasubramanian and Kasturi Bai¹⁰⁵ reported that a concentration of 0.1 to 0.3% biogas slurry was optimum for maintenance of algae and *Daphnia similis*. Further, they reported¹⁰⁹ that biogas slurry applied at 0.15% level at 3 days interval increased the growth rate of *Labeo rohita*, *Cirrhinus mrigala*, and *Cyprinus carpio*, and was 3.6-fold higher than conventional cultivation systems where there was no application of slurry.

Conclusion

Although biogas production technology has established itself as a technology with great potential which could exercise major influence in the energy scene in rural areas, it has not made any real impact on the total energy scenario despite the presence of about 1.8 million biogas digesters. One of its serious limitations is the availability of feedstock followed by defects in construction, and microbiological failure. But on reviewing the literature, one finds a long list of alternate feedstocks and their potential for biogas production. It is time that substrate-specific biocatalysts are made available to reduce the lag period of biomethanation during the start-up. Regular supply of inoculum and quality control on the marketable inoculum will result in regulating the plant failures.

Furthermore, designs to suit the microbial catalysts have been discussed for long, but have yet to be realized. This indicates that the technology transfer is not complete, and that it requires coordinated efforts of scientists, and engineers to overcome these limitations in order to translate this 'high potential' technology to 'high performing' technology.

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MEETINGS/SYMPOSIA/SEMINARS

XV Asia Pacific Cancer Conference

Date: 12–15 December 1999

Place: Chennai, India

Contact: Congress Secretariat
 XV Asia Pacific Cancer Conference
 Cancer Institute (W.I.A.)
 18 Sardar Patel Road
 Chennai 600 036, India
 Phone: 91-44-2350131, 91-44-2350241
 Fax: 91-44-4912085
 Conference website: <http://apcc.uicc.org>
 e-mail: caninst@md2.vsnl.net.in

Asian Seminar on Indigenous System of Medicine

Date: 5–7 February 2000

Place: Rajgir, India

Topics include: 1. Fundamental principles of treatment in indigenous system of medicine; 2. Enrichment of bio-resources by advanced agro-technology and biotechnology; 3. Meeting challenges of congenital deformities including physical, mental and sexual infirmity and also steps to be taken in the field of reproductive biology; 4. Management of diseases caused by tobacco, drug-addiction, food-adulteration, malnutrition and population explosion; 5. Contributions of Lord Buddha, Lord Mahabir and their followers in the management of diseases and health-care and importance of keeping away from non-veg foods; 6. Discussion about the mode of life as practised in the Vedic Era and effect of glorious sunshine on life. Papers on other related important topics will also be considered.

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Short Course on Paleoseismology

Date: 11–16 October 1999

Place: Imphal

The course, sponsored by the DST, Govt of India will present the major trends and techniques in paleoseismology. It is open to post-graduate students, researchers and teachers mainly from the northeastern region with background in geology/geophysics. Applications will be reviewed by a selection committee and will be accepted no later than 30 August 1999. Travel grant and living expenses will be provided to selected candidates. Applications containing full CV with recommendation from the supervisor must be sent to one of the following addresses:

Dr C. P. Rajendran
 Course Director
 Centre for Earth Science Studies
 Akkulam, Trivandrum 695 031
 Fax: 0471-442280; Phone: 0471-4442451 Extn 326
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