

# Chapter 12

## Current Trends and Aspects of Microbiological Biogas Production



Chayanika Putatunda, Abhishek Walia, Rashmi Sharma, and Preeti Solanki

**Abstract** The whole world today is practically on the verge of severe energy crisis. The industrialization and modernization have improved the overall living conditions of a segment of human population on one hand and also resulted in greater energy requirements as well as a huge burden on the already dwindling land resources. Again, sustaining the ever-increasing human population with the ever-decreasing arable land is becoming a mammoth task. So, there is an urgent need to try to look for possible solutions. Biogas production or biomethanation can be an answer to the twin problem of food and energy since the process leads to generation of biofuel (biogas) as well as effluent slurry, which can act as a very good manure. Another, very significant advantage of the process is that it is very helpful in solid waste management. A wide variety of waste materials like animal excrements, sewage sludge, agricultural residues, industrial wastes, etc. can be used as substrate for biogas production. Moreover, the solid-state biomethanation is also gaining popularity due to negligible water requirements. This chapter presents some of the major developments in the field of biogas production.

**Keywords** Biogas · Methane · Microbiological diversity · Microbial decomposition · Anaerobic digestion · Digestors

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## 12.1 Introduction

Energy and population explosion are two of the major issues being faced by the whole world today. The energy needs are increasing day by day due to the ever-increasing population as well as greater industrialization and modernization (Amigun et al. 2008; Li et al. 2009; Sárvári Horváth et al. 2016). However, we are still heavily dependent on fossil fuels for the energy requirements, which in turn are depleting on an alarming rate (Shafiee and Topal 2009; Nel and Cooper 2009; Abas et al. 2015). Also, these fossil fuels like coal, petroleum products, etc. apart from being nonrenewable are also major source of environmental pollution and are not only impacting the environment but also gravely affecting the human health (Smith et al. 1999; Höök and Tang 2013; Watts et al. 2015; Lelieveld et al. 2015; Perera 2017). Apart from that, the agriculture sector is also under extreme pressure due to the food requirements of the growing human population (FAO 2017). On the other hand, the developmental measures and urbanization are also taking a great toll on the available land resources, thus diminishing the already dwindling arable land (Agus and Irawan 2006; Beesley and Ramsey 2009; Azizan and Hussin 2015). Thus, the farmers are relying on more energy-intensive agricultural practices as well as huge amounts of chemical fertilizers and pesticides which in turn is having a grave impact on the environment (Tilman et al. 2002; Cold and Forbes 2004; Moss 2008; Alamdarlo 2018; Budzinski and Couderchet 2018). Not only this, the greater human activities are also leading to a very large quantity of waste materials which are again posing a serious threat to the environment (Dyson and Chang 2005; Giusti 2009; Fu et al. 2015). So, these problems are very intricately related with each other. Hence, there is an urgent need to address these issues.

A potential solution to these problems is the process of biomethanation. The process involves anaerobic digestion of organic materials, carried out by several groups of microorganisms, leading to production of methane and some other gases in the form of biogas (Dieter and Angelika, 2008). As per Martins das Neves (2009), “The energy content of 1.0 m<sup>3</sup> of purified biogas is equal to 1.1 L of gasoline, 1.7 L of bioethanol, or 0.97 m<sup>3</sup> of natural gas.” Biogas can be used as a fuel for cooking and heating purposes as well as for electricity generation (Xiaohua and Jingfei 2005; Riva et al. 2014; Chang et al. 2015). There is also the possibility of using it as a transportation fuel (Börjesson and Mattiasson 2008; Holm-Nielsen et al. 2009). Moreover, biogas production also yields an effluent slurry which is known to be rich in several plant nutrients (Leela Wati et al. 2008). The effluent can be applied as an organic manure which improves the soil quality as well as plant growth. Sogn et al. (2018) have reported that the biogas plant digestate proved to be a good source of the major plant nutrients like nitrogen, phosphorus, and potassium, and at least for the wheat crop, the digestate alone was sufficient to overcome the requirement of any chemical fertilizer. Diatta et al. (2019) reported that 1:1 mixture of phyto-ash and biogas slurry was found to be effective in improving the quality of soil contaminated with heavy metals. Similarly, Nafees

et al. (2018) reported improvement in growth as well as antioxidant properties of *Brassica napus*, grown in chromium-contaminated soil by addition of biogas slurry and *Burkholderia phytofirmans* PsJN. The anaerobic conditions occurring during biomethanation also tend to decrease the load of pathogenic microbes, so the safety concerns associated with the direct soil application of waste materials are also mitigated (Weiland 2010).

Conventionally, cattle dung has been used as the most common substrate for biogas production (Işık and Polat 2018). However, a variety of waste products (either alone or mixed with other wastes) have been reported to be effective for biogas production. Some of the substrates used for biogas production are the excretory materials of various animals like sheep (Sarabia Méndez et al. 2017), goat (Zhang et al. 2013), pigs (Wu et al. 2010), camel (Kheira et al. 2017), and poultry birds (Malik et al. 2008); human excreta (Singh et al. 1993); kitchen wastes (Iqbal et al. 2014; Srinvasa Reddy et al. 2017); agricultural wastes like bagasse (Eshore et al. 2017), wheat straw (Mancini et al. 2018) etc.; industrial wastes like whey (Antonelli et al. 2016); paper industry wastes (Priadi et al. 2014); palm oil industry wastes (Ohimain and Izah 2017); food processing industry wastes (Fang 2010), etc. So, biomethanation can be a boon for both industrial and agricultural sectors, since the industries can use biogas as a possible source of energy and at the same time, the process is helpful in dealing with their waste products. Thus, apart from solving the energy as well as food problem, biogas production is very useful in addressing the environmental issues also. García-González et al. (2019) have concluded that by harvesting the methane in the form of biogas, one can expect mitigation of the greenhouse gas emission. Thus, this can be helpful in reducing the phenomenon of global warming.

Biogas conventionally comprises approximately 60–75% methane, 25–40% carbon dioxide, and traces of other gases like water vapor, hydrogen sulfide, ammonia, etc. (McKendry 2002; Zinoviev et al. 2010). Out of these, methane is the ingredient which acts as the fuel. It is a clean burning fuel and has a high calorific value (Kaltschmitt et al. 2001). Reports have already indicated improvement in health conditions of people working in kitchens supplied with biogas as compared to kitchens where cattle dung cakes, coal, or wood is used as fuel (Dohoo et al. 2012). However, in spite of all these benefits, the process of biomethanation has not gained the desired level of popularity. Lack of awareness among the general public as well as limitation of funds may be the reason for this, but there are some areas of concern with respect to biogas production technology (Surendra et al. 2014). For example, the relatively low efficiency of biogas production especially in areas with too high or too low temperatures is an important drawback of this process. Again, another issue is related to the storage and purification of methane from the rest of the constituents of biogas. Even the availability of substrate and labor requirements has also been thought to impact the popularity of biogas production adversely (Tucho et al. 2016). Apart from these, the conventional biogas production technologies also need input of lots of water (Tucho et al. 2016). This especially becomes challenging in arid and semi-arid regions of the globe.

In order to address these types of concerns, research efforts are being undertaken. Scientists are working on improving the overall process efficiency by modifying the digester designs as well as the process parameters. Similarly, efforts are going on for developing microbial consortia which can not only improve the yield (Zhong et al. 2016; Krzysztof et al. 2016) but also can work efficiently under relatively harsh temperature ranges (Hniman et al. 2011; Kinet et al. 2015). Apart from that, several researchers have shown that by combining different wastes as substrates for biogas production, the process efficiency can be improved (Li et al. 2011a; Munda et al. 2012; Tasnim et al. 2017). Again, several workers have also developed various techniques for improved purification of biogas. Another major step in popularizing the biogas production technology is by the application of solid-state biogas production which minimizes the amount of water requirement for biomethanation (Brown et al. 2012; Brown and Li 2013). These steps are discussed in detail in the following sections.

## 12.2 Conventional Biogas Production

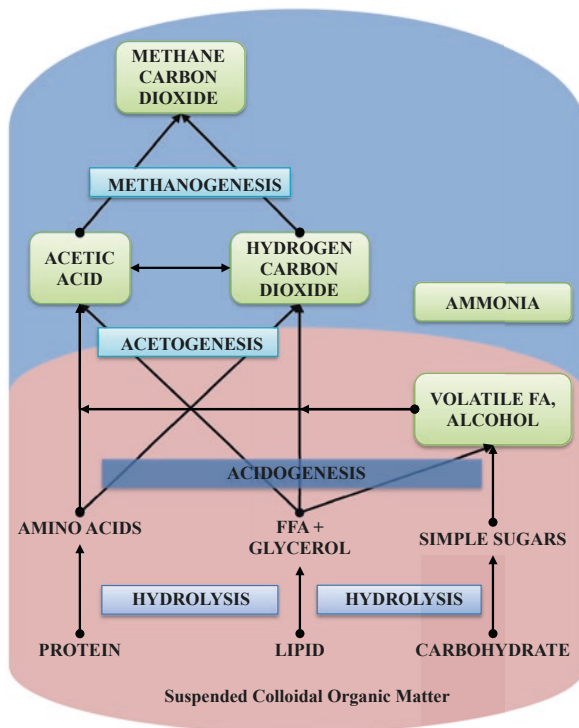
Recent studies have suggested that biogas production using anaerobic digestion (AD) processes delivers compelling edge over the rest of the sources of bioenergy as AD is not only an energy-efficient but also an environmental-friendly technique (Nishio and Nakashimada 2007; van Forest 2012). Many countries like the USA, China, and India have recently been investing in alternative processes for the production of biogas from cellulosic resources, and they are going to be the future producers of biogas (Lin and Tanaka 2006; Soetaert and Vandamme 2009). The AD technology has been enhanced from the knowledge of the production of compost which is a high-value fertilizer and has today given a boon to biogas economy (European Biogas Association 2011).

### 12.2.1 Principle of Anaerobic Digestion

AD is based on microbial decomposition of organic matter in the absence of air/oxygen, utilized for metabolism in microbes, leading to their growth and resulting in production of methane. The process can be divided into four characteristic phases depending upon the group of microorganisms involved (Fig. 12.1).

Effective regulation of the AD process is necessary for stable digestion and is maintained by coupling the various biological conversions, i.e., hydrolysis, acidogenesis, acetogenesis, and methanogenesis, that prevents the accumulation of any intermediary compounds. This also helps in the maintenance of physical conditions of digestion like pH which may be affected by the concentration of intermediary VFAs, leading to decrease in pH, which inhibits methanogenic bacteria, and further decreases the pH.

**Fig. 12.1** Principle of anaerobic digestion process: (a) hydrolysis: nonsoluble biomolecules (protein, lipids, carbohydrates) present in the colloidal organic matter of the waste are converted to simpler and soluble organic compounds. (b) Acidogenesis: soluble organic compounds → volatile fatty acids (VFA) + CO<sub>2</sub>. (c) Acetogenesis: VFA → CH<sub>3</sub>COO<sup>-</sup> + H<sub>2</sub>. (d) Methanogenesis: conversion of acetate and CO<sub>2</sub> plus H<sub>2</sub> to methane gas



### 12.2.2 Types of Biogas Systems

AD technology is applicable to both wastewater and solid waste treatments with the final product being methane and carbon dioxide. The installation required for AD is both simple and economical in terms of technology, energy, and space and can be divided into two types based on biomass retention (de Mes et al. 2003):

- (a) High-rate biogas systems: These systems are characterized by biomass retention and short hydraulic (HRT) but long sludge retention times (SRT) and maybe utilized for many types of wastewater treatments. Examples are anaerobic filter, contact process, upflow anaerobic sludge blanket (UASB) or expanded granular sludge bed (EGSB), and fluidized bed systems. The biomass retention may be possible in two ways:
  - (i) Bacterial films are fixed on solid surfaces.
  - (ii) Bacterial mass is suspended and biomass is retained by settling process internally or externally.
- (b) Low-rate biogas systems: These systems do not retain biomass and are used for solid waste treatment and digestion of slurries. They have long enough HRT to be equal to SRT. Examples are continuously stirred tank reactor (CSTR) system, plug flow system, and batch accumulation system.

### **12.2.3 Substrates for Biogas Production**

Various types of wastes can be used for the production of biogas, like the ligno-cellulose waste from agricultural and municipal sources, sewage sludge, solid municipal waste, animal manure and slurry, food wastes, etc. These wastes have been evaluated for their biogas yield ( $\text{m}^3$ ) as well as electricity produced (kW-h) per ton of fresh biomass (Stucki et al. 2011). The molecular constituents, i.e., carbohydrates, fats, proteins, cellulose, lignin, and hemicellulose, are the factors contributing to the quantity and quality of biogas yield. It is also known that fats provide a better biogas yield in comparison to carbohydrates and proteins, but the latter are however converted faster by the microbes (Zubr 1986; Braun 1982, 2007).

Lignocellulosic wastes, i.e., agricultural wastes, sewage, and wastes from energy crops, are potent sources of biofuels (Philbrook et al. 2013) and are made of primarily three components—cellulose, hemicellulose, and lignin (Kumar et al. 2009; Iqbal et al. 2011). Lignin makes up 30–60% of wood and 5–30% of agricultural wastes and grasses (Ratnaweera et al. 2015), whereas wastes from energy crops primarily contain hemicellulose (Demirbaş 2005). It has been shown that higher lignin content in the feedstock biomass decreased the degradation efficiency (Grabber 2005) and the structure and composition of lignin positively influence the process of hydrolysis, i.e., the first step in biogas production, hence increasing the biogas production efficiency (Ladisch et al. 2010).

#### **12.2.3.1 Treatment of Slurries and Solid Wastes**

The content of total solids (TS) present in the solid wastes and slurries determines the systems to be used for their digestion. Broadly, either wet fermentation systems or dry fermentation systems are employed (de Mes et al. 2003) (Table 12.1).

#### **12.2.3.2 Wastewater Treatments**

High-rate AD treatment systems like UASB, contact process, and anaerobic filter are better suited for dilute feedstock like wastewater. In these systems,  $\text{SRT} > \text{HRT}$  and the sludge is either recycled or fixed on support material. Different types of wastewater treatment systems used worldwide are contact process, upflow anaerobic sludge blanket (UASB), Biobulk system by Biothane (Biothane, 2001), fixed film fluidized bed system, hybrid systems, anaerobic filter (AF), anaerobic fixed film reactor (AFFR), and expanded granular sludge bed (EGSB) (Frankin 2001).

**Table 12.1** Fermentation treatment of slurries and solid wastes

TS 15–25% Low solids AD	TS > 30% High solids AD
Wet fermentation	Dry fermentation
<p><i>CSTR</i></p> <ul style="list-style-type: none"> <li>- Feed introduced in the reactor is continuously stirred for proper mixing of contents, and an equal volume of effluent exits the reactor</li> <li>- RT can be varied</li> <li>- Low operating cost</li> <li>- TS 2–10%</li> <li>- For treatment of agricultural wastes, animal manure, household waste, sewage sludge, feces, kitchen waste, urine, and/or mixtures of these substrates</li> <li>- SRT = HRT</li> <li>- Digester volume: 6–400 m<sup>3</sup>/day</li> </ul>	<p><i>Valogra system</i></p> <ul style="list-style-type: none"> <li>- Waste is screened and crushed to particle size &lt;80 mm</li> <li>- Crushed waste is mixed with some excess process water and heated via steam</li> <li>- Four high-solids mesophilic reactors are combined with incineration of non-digested matter and residues</li> <li>- Mixing in reactor occurs under pressure by reverse circulation with a small volume of the biogas</li> <li>- Methane content in biogas produced after 18–25 days is 55–60% which may be purified to 97% for commercial use</li> </ul> <hr/> <p><i>DRANCO (Dry Anaerobic Composting) system</i></p> <ul style="list-style-type: none"> <li>- Operates with high solid fraction and high temperatures (50–58 °C)</li> <li>- Feed is daily added from top and digested material removed from base after 15–30 days</li> <li>- Part of digested biomass is recycled to be used as inoculum, and the rest is dried to form organic compost</li> <li>- No mixing within the reactor except the plug flow movement of the digested waste</li> </ul>
<p><i>Plug flow digesters</i></p> <ul style="list-style-type: none"> <li>- Basic design made of an underground trough with expandable gas-tight cover, vertical mixing in pipe</li> <li>- Hydrolysis and methanogenesis occur separately through the pipe length</li> <li>- TS 10–12%</li> <li>- SRT = HRT</li> <li>- For treatment of slurries with high TS fractions</li> <li>- Low loading rates</li> </ul>	<p><i>Kompogas system</i></p> <ul style="list-style-type: none"> <li>- Reactor is a horizontal cylinder; hence, movement of material in reactor is in a horizontal type plug-flow process; an intermittent agitator is also present inside the reactor</li> <li>- Feed is introduced daily from one end and digested biomass removed from the other end after 20 days</li> <li>- Part of digested biomass is recycled to be used as inoculum, and the rest is sent to AD wastewater treatment to produce more biogas</li> </ul> <hr/> <p><i>BIOCEL process</i></p> <ul style="list-style-type: none"> <li>- Batch process with high solids and mesophilic temperatures</li> <li>- Waste is mixed with the inoculum, sealed into the bioreactor without any stirring, and kept till 21 days for biogas production, i.e., till there is no more methane produced</li> <li>- Leachate formed due to AD is heated for recirculation through waste biomass</li> </ul>

### 12.2.4 Types of Biogas Plants

The construction of a biogas plant depends upon its structural strength and fluid dynamics. The best possible solution may be an egg-shaped vessel which is generally used for treatment of sewage on large scale as it is expensive. Digesters in the shape of a cylinder having conical bottoms and covers are much simplified design to build and maybe available as prefabricated units in the market. They are however unfavorable due to surface volume ratio and must have equal height and diameter. Comparison of different types of small-scale biogas plants and their advantages and disadvantages are summarized in Tables 12.2 and 12.3. The basic designs of fixed dome and floating drum biogas plants are given in Figs. 12.2 and 12.3, respectively.

### 12.2.5 Physical Factors Affecting AD

- (a) Temperature: AD starts with 0 °C and increases with increasing temperatures reaching a maximum at 35–37 °C, i.e., ideal conditions for mesospheric microorganisms. Maximum methanogenesis occurs at 55 °C or higher, and the choice of temperature will depend upon the biogas yield and the energy demands (Lettinga and Haandel 1993). Some of the important characteristics of different types of anaerobic digestion processes (based on temperature) are presented in Table 12.4.

**Table 12.2** Comparison of different types of small-scale biogas plants (Biogas Digest Volume II: Biogas—Application and Product Development, 2010)

Factors	Fixed dome	Floating drum	Tubular design	Plastic containers
Gas storage	Gas storage internal; large drum sizes up to 20 m <sup>3</sup>	Gas storage internal; small drum sizes	Internal and eventually external plastic bags	Gas storage internal; small drum sizes
Gas pressure	60–120 mbar	~20 mbar	Low, ~ 2 mbar	Low, ~ 2 mbar
Skills of contractor	High; masonry, plumbing	High; masonry, plumbing, welding	Medium; plumbing	Low; plumbing
Availability of material	Yes	Yes	Yes	Yes
Durability	Very high >20 years	High; weak drum	Medium; depends on chosen liner	Medium
Agitation	Self-agitated by the biogas pressure	Manual steering	Not possible; plug flow type	Manual steering
Sizing	6–124 m <sup>3</sup> digester volume	~20 m <sup>3</sup>	Combination possible	~6 m <sup>3</sup> digester volume
Methane emission	High	Medium	Low	Medium



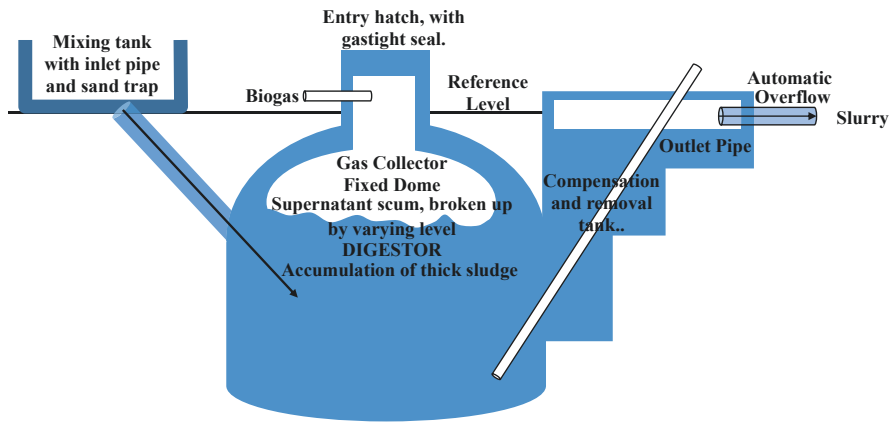
**Table 12.3** Advantages and disadvantages of different types of small-scale biogas plants (Biogas Digest Volume II: Biogas—Application and Product Development, 2010)

Biogas reactor type	Advantages	Disadvantages
(a) Fixed dome biogas plants	<ul style="list-style-type: none"> <li>• Initial costs are low</li> <li>• Life span is long and useful</li> <li>• Parts involved are neither rusting nor moving</li> <li>• Compact well-insulated underground basic design which saves space</li> <li>• Opportunity for skilled local employment</li> </ul>	<ul style="list-style-type: none"> <li>• Gas-holder masonry requires special sealants</li> <li>• High technical skills required for gas-tight construction</li> <li>• Gas leaks are frequent</li> <li>• Gas utilization is complicated due to fluctuation in gas pressures</li> <li>• Amount of biogas produced may not be immediately visible</li> <li>• Plant operation is not easily understandable</li> <li>• Exact planning of levels is required, and in bedrock areas, the excavation will be difficult</li> <li>• Environmental disadvantage—methane emission from expansion chamber</li> </ul>
(b) Floating drum biogas plants or gobar gas plant (by Jashu Bhai J Patel, 1956)	<ul style="list-style-type: none"> <li>• Mode of operation is continuous feed</li> <li>• Can be used for both animal and human feces</li> <li>• <i>Water-jacket floating-drum plants:</i> <ul style="list-style-type: none"> <li>– Easy to maintain</li> <li>– Universally applicable</li> <li>– Do not stuck in scum layer even with high solid content</li> <li>– Long life</li> <li>– Esthetic and more hygienic</li> <li>– Used in fermentation of night soil</li> </ul> </li> </ul>	<ul style="list-style-type: none"> <li>• Steel drum is maintenance intensive and expensive</li> <li>• Life span of drum is short, i.e., 5–15 years only</li> <li>• Gas holder has a tendency to be stuck if fibrous substrates are used</li> <li>• A guide is always required for the drum to be removed for repair</li> </ul>
(c) Low-cost polyethylene tube/balloon biogas digester	<ul style="list-style-type: none"> <li>• Easy design and low cost</li> <li>• Easy installation of digester bag, replaceable</li> <li>• Can be installed in areas with high water table</li> <li>• Easy monitoring of gas</li> <li>• Ideal for warmer climates</li> </ul>	<ul style="list-style-type: none"> <li>• Shorter life span than traditional biogas plants</li> <li>• Gas pressure is lower than fixed dome and floating drum biogas plants</li> <li>• Production time is highly dependent on the ambient temperature, less insulated</li> </ul>
(d) Earth pit plants	<ul style="list-style-type: none"> <li>• Easy design and low cost</li> <li>• Easy installation</li> <li>• Potential for improvements based on self-help approaches</li> </ul>	<ul style="list-style-type: none"> <li>• Short life span</li> <li>• Can be made only in certain impermeable soils</li> <li>• Can be constructed only above groundwater table</li> </ul>

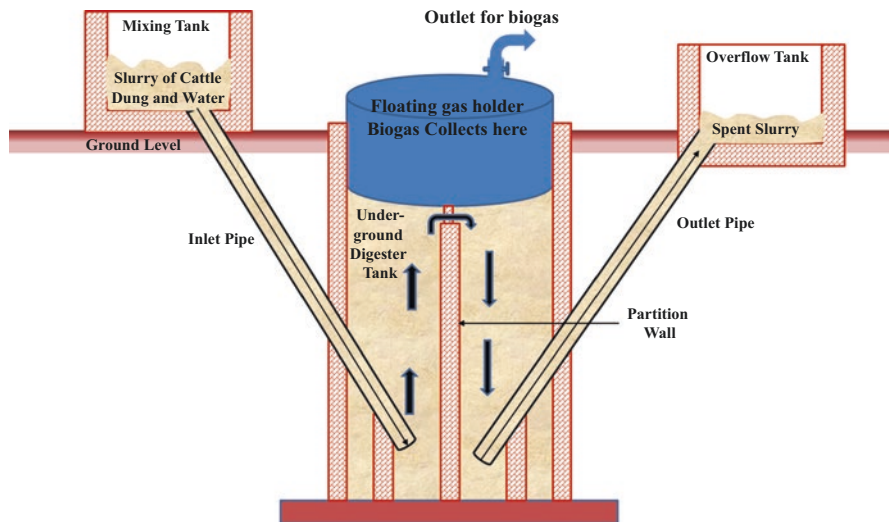
(continued)

**Table 12.3** (continued)

Biogas reactor type	Advantages	Disadvantages
(e) Ferro-cement plants	<ul style="list-style-type: none"> <li>• Smaller volumes (&lt;6 m<sup>3</sup>)</li> <li>• Can be prefabricated</li> <li>• Can be applied as an earth pit lining or as self-supporting shell</li> <li>• Reliability is proven if made with cemented-on aluminum foil</li> </ul>	<ul style="list-style-type: none"> <li>• Requires substantial amount of cement</li> <li>• Skillful workmanship is required</li> <li>• Uses expensive wire mesh</li> <li>• Adequately not yet time tested</li> <li>• Requires special sealing measures for gas holder</li> </ul>



**Fig. 12.2** Basic components of a fixed dome plant (Nicarao design)



**Fig. 12.3** Floating drum biogas plants or gobar gas plant

**Table 12.4** Classification of digestion process based on temperature

Psychrophilic	10–20 °C	Bacterial growth and degradation of the substrate are slower	Requires long retention times and large reactor volumes
Mesophilic	20–40 °C	Moderate bacterial growth and substrate degradation	Lesser retention times and reactor volumes
Thermophilic	50–60 °C	Applicable when wastewater is discharged at high temperature or when pathogen removal is essential	Can be used with high loading rates

- (b) pH: AD start-up processes may occur at a variable pH, but methanogenesis requires neutral pH and is the rate limiting step (Lettinga and Haandel 1993). Hydrogen carbonate ions are required in sufficient amount to maintain the optimal pH for methanogenesis to occur.
- (c) Alkalinity and toxicity: Accumulation of intermediaries like VFA and ammonia, or cations like those of sodium, potassium, and calcium, heavy metals, sulfides, and other xenobiotics also adversely affect methanogenesis.

### 12.3 Microbiology of Biogas Production

The process of biogas production from organic matter is a complex and dynamic process and involves intricate interactions between the members of microbial community. The microbial communities of seven different anaerobic sewage sludge digesters were analyzed by using 16S rDNA sequencing (Riviere et al. 2009). They classified the microflora into three categories, viz., the phylotypes common in most of the digesters, the phylotypes found in few digesters, and the phylotypes observed only under certain specific conditions.

The overall anaerobic digestion process has been divided into four major steps, viz., hydrolysis, acidogenesis, acetogenesis, and methanogenesis. During the first step of hydrolysis, the macromolecules like cellulose, starch, hemicellulose, proteins, lipids, etc. present in the organic materials are converted to simple, soluble monomeric compounds like formate, acetate, propionate, butyrate, ethanol, carbon dioxide, and hydrogen (Kelleher et al. 2000). Depending on the type of starting materials, different microbes may become dominant during this phase. According to Rao (1993), cattle dung-fed digesters show higher amylolytic population while the digesters containing poultry waste possess higher proteolytic population. Some of the commonly reported hydrolytic microbes include *Bacteroides succinogenes*, *Butyrivibrio fibrisolvens*, *Clostridium cellobioparum*, *Ruminococcus albus*, *Clostridium* sp., etc. (Khan 1980; Godbole et al. 1981). The small molecules produced by the fermentative and hydrolytic microbes are acted upon by acidogenic microbes which result in the production of volatile fatty acids (VFAs), alcohols, hydrogen, and carbon dioxide along with by-products like ammonia (NH<sub>3</sub>),

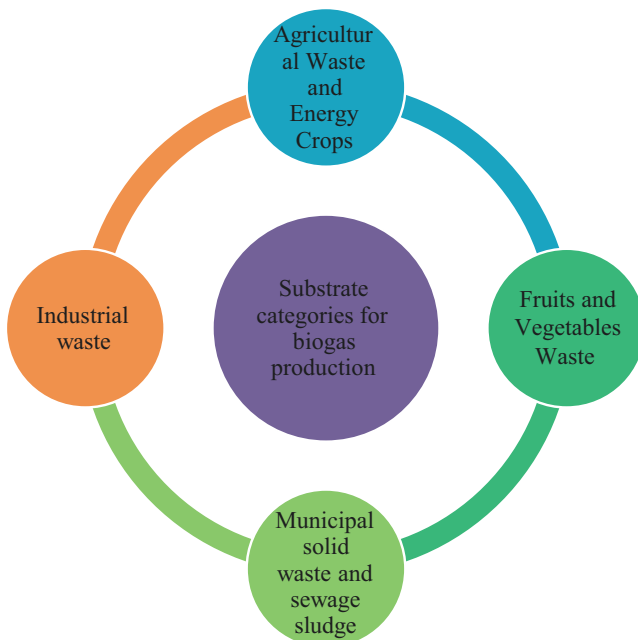
hydrogen sulfide (H<sub>2</sub>S), etc. (Li et al. 2011a, b; De la Torre and Goma 1981; Dinopoulou et al. 1988). Acidogenesis is followed by acetogenesis where organic acids and alcohols are converted into acetate, carbon dioxide, and hydrogen. The obligate hydrogen-producing acetogenic bacteria are one of the most important groups in the biogas digesters. The classical examples of this group of microbes are *Syntrophobacter wolinii* (Boone and Bryant 1980) and *Syntrophomonas wolfei* (McInerney et al. 1981). The most common homoacetogenic bacteria are *Acetobacterium woodii* and *Clostridium aceticum*. These consume hydrogen and carbon dioxide and produce acetate (Balch et al. 1977; Ohwaki and Hungate 1977; Bharathiraja et al. 2018). The last step of anaerobic digestion is carried out by methanogens. There are two broad groups of methanogenic bacteria: aceticlastic bacteria (which convert acetate into methane and carbon dioxide) and hydrogenotrophic methanogens (which convert hydrogen and carbon dioxide into methane). Most of the methanogens belong to the latter group, while the former group is represented by a few like *Methanosarcina barkeri*, *Methanococcus mazei*, and *Methanotherix soehngenii* (Schink 1997). The syntrophic interaction between acetogens and hydrogenotrophic methanogens is very crucial for the performance of an anaerobic digester.

## 12.4 Current Trends in Biogas Production

With the advent of modernization and improvement in the living standards, the energy requirements are skyrocketing. On the other hand, the conventional energy sources are plummeting. So, the world is now trying to look for alternative and renewable energy sources (Nasir et al. 2012). Biomethanation is not a new phenomenon, but nowadays, several improvements or modifications are being done in order to enhance its applicability as a reliable energy source. Some of the approaches are being discussed here.

### 12.4.1 Utilization of Wider Substrate Range

The substrate feedstocks serve as the nutritional base for the microflora involved in biogas production. The types of nutrients within substrate feedstock determine microbial growth and hence affect degradation process and biogas yield (Cheng 2009; Cooke 2014; De Clercq et al. 2016). Nutrients should be in abundance for an efficient digestion process. Therefore, the substrate should be chosen in a way that meets the nutritional demands of microflora in the digester, produce high biogas and methane yield, and produce a high-quality digestate. Different categories of substrates used for biogas production are depicted in Fig. 12.4, and these categories include various feedstocks such as animal manure, slurries, organic wastes generated from agriculture, dairies, food industries and wastewater sludge, organic inputs from municipal solid wastes, households, as well as energy crops (Cheng 2009).



**Fig. 12.4** Substrate categories utilized during anaerobic digestion for biogas production

Cattle dung or cattle manure is the chief organic waste generated within agriculture sector. It has been commonly used as the substrate for biogas production. It is nutrient rich and used by farmers as soil conditioner for increased crop productivity (Cooperband 2002; Yorgey et al. 2014). Cattle manure can also be used as ideal substrate for biogas production, and later the digestate may be used as a soil conditioner. It has been reported that manure digestion increases the availability of nutrients for plants, particularly nitrogen (LeaMaster et al. 1998). Besides beneficial effects, animal manure can pose harm to the environment if not handled properly (Phetyim et al. 2015). High concentration of nitrogen and phosphorus may cause nutrient imbalance; it also contains some fractions of toxic substances such as heavy metals, antibiotics, and some growth regulators. Moreover, the presence of many pathogenic microorganisms in the manure can lead to the outbreak of serious plant and human diseases (LeaMaster et al. 1998). Thus, dumping of the animal manure in an environment-safe way is a matter of prime concern. So, biomethanation offers an attractive avenue of dealing with cattle dung.

#### 12.4.1.1 Agricultural Waste as Substrate Feedstock

Manure could be used as solid phase or slurry; however, more biogas yield has been reported with slurry phase (Sebola 2015). Manure has its own properties and can be mixed with other agricultural substrates during anaerobic digestion for optimum

biogas yield. In fact, several studies indicate that combining different wastes with cattle dung can be effective in enhancing the biomethanation process. A 16–65% increase in methane yield was observed when cattle dung was mixed with grass silage and sugar beet litter at the organic loading rate of 2 kg VS/m<sup>3</sup> d at 35 °C (Lehtomäki et al. 2007). Anaerobic digestion of a mixture of water hyacinth, cattle manure, algae, and rice husk gave more methane yield and a more nutrient-rich digestate as compared to cattle manure alone (Ghosh and Das 1982). A higher methane yield was reported from anaerobic digestion of cattle dung mixed with lantana slurry, apple-peach leaf litter, and wheat straw (Dar and Tandon 1987). Different studies have reported a high biogas yield with amendments of various organic agricultural wastes with cow dung; a mixture of water hyacinth, cattle dung, and poultry waste increased biogas yield by 100% (Madamwar et al. 1990). Similarly, a high biogas yield has been reported from anaerobic digestion of sugarcane bagasse, cattle dung, and poultry waste (Mallik et al. 1990).

#### 12.4.1.2 Energy Crops

Crop residues and various energy crops are used as feedstocks for digestion during biogas production. Grass is the most preferred energy crop due to easy digestibility, and also its availability is not hindered by seasonal change. A high methane yield (70–80%) has been reported from grass silage digestion, and moreover, methane produced from grass has been found to be suitable for automobile fuel (Gerin et al. 2008; Abu-Dahrich et al. 2011). A significant biogas yield has also been reported from anaerobic digestion of various plant materials such as *Ageratum*, *Calotropis procera*, *Beta vulgaris*, water hyacinth, and even marine macroalgae (Kalia and Kanwar 1990; Traore 1992; Hassan 2003; Singhal and Rain 2003; Hughes et al. 2012).

#### 12.4.1.3 Fruit and Vegetable Waste

Markets and food industries generate tons of fruit and vegetable waste every year. Fruits and vegetables are high moisture-rich (70–90%) substrates, and anaerobic digestion is an ideal way of recycling waste containing 50% or more moisture content. But, the use of fruit and vegetable waste as a sole substrate for biomethanation is an exigent process due to poor substrate composition (Asquer et al. 2013; Bouallagui et al. 2003; Sanjaya et al. 2016). Therefore, different pretreatments are given to increase biogas yield from fruit-vegetable waste (Asquer et al. 2013). The major setback of using fruits during digestion is that they rapidly acidify digestion mixture due to the low pH of waste and accretion of volatile fatty acids, which reduce the activity of methanogens in the digester. Therefore, preliminary treatment is required for efficient digestion of fruit and vegetable wastes (Hall 1995; Appels et al. 2008; Arthur et al. 2011).

#### 12.4.1.4 Municipal Solid Waste and Sewage Waste

Waste generated and collected from residential areas, institutions, and mercantile activities is called municipal solid waste (MSW). MSW is usually collected as mixed stream and then dumped through landfilling sites. Landfilling is not considered an ideal way to get rid of MSW as it is a waste of energy and nutrients. MSW contains a significant biodegradable fraction (65–70%); therefore, it can be subjected to anaerobic digestion for biomethane production. Literature has well documented the production of methane from anaerobic digestion of MSW (Davidson et al. 2008; De Clercq et al. 2016).

Sewage sludge is generated from municipal and industrial wastewater treatment plants, and its production is increasing swiftly with increase in industrialization (Appels et al. 2008). The sludge is a storehouse of nutrients like nitrogen and phosphorus and organic matter; therefore, it is used as manure in crop fields. However, sewage sludge also contains some toxic substances like heavy metals and pathogenic microorganisms; thus, its direct use as manure is environmental sensitive and should be reconsidered (Appels et al. 2008). Sludge treatment through anaerobic digestion is the method of choice for biomethane production, and the digestate produced is nutrient rich and safe and can be used as manure.

Sludge contains significant moisture and other micro- and macronutrients but low in C:N ratio. Therefore, biomethane production is optimized by supplementing with organic fraction of MSW; the process is called co-digestion (Pastor et al. 2013). Organic fractions of MSW are high in C:N ratio, and a significant increase in methane yield is reported with co-digestion of MSW and sewage sludge (Tchobanoglous et al. 1993; Raposo et al. 2012).

#### 12.4.1.5 Industrial Waste

Industrial waste is the waste generated from various manufacturing industrial units such as paper and pulp industries, dairy industries, food industries, and agro-product-based industries. Anaerobic digestion is the method of choice for treatment of industrial waste in an environmental safe way. It has been reported that waste from paper-pulp industries and butcher houses can be utilized for biomethane production (Jia et al. 2013). Waste from paper-pulp industries is rich in organic carbon and can be easily biodegraded during anaerobic digestion; therefore, it is ideally suited as a substrate in biogas plants (Hagelqvist 2013). Anaerobic digestion of waste from paper and pulp industries is also economically beneficial to industries as it includes low transportation cost and operation and maintenance can be easily coordinated with existing organizations (Hagelqvist 2013).

Besides pulp-paper industries, wastes from textile industry, biscuit and chocolate industry, cheese industry, and distilleries have been successfully employed in biogas production (Balasubramanya et al. 1986; Mehla 1986; Ranade et al. 1989; Lebrato et al. 1990). A biogas yield of 2.2 liter per day has been observed when whey, cow dung, and poultry waste (3:1:2) were supplemented at a rate of 6 grams of total

**Table 12.5** Production of biogas from various industrial wastes

Industrial wastes used for biogas	Reference
Dairy effluents	Rani (2001)
Spent tea leaves	Goel et al. (2001)
Wastewater from food processing unit	Wei et al. (2011)
Glycerol as by-product of biodiesel production	Viana et al. (2012)
Brewery waste	Tewelde et al. (2012)
Palm mill effluent	Thong et al. (2012)
Paper mill wastes, brown grease, and corn ethanol	Zhang (2017)
Cassava industrial waste	Budiyono et al. (2018)

solids/day at 40 °C (Desai and Madamwar 1994a). Later, an increase in fuel yield was obtained after adding silica gel (Desai and Madamwar 1994b) and Tween 80 (Desai and Madamwar 1994c). Table 12.5 shows production of biogas from various industrial wastes.

#### 12.4.2 Alterations of Microflora

Biogas production involves many different groups of microorganisms working together to anaerobically degrade the organic matter and to produce methane and other gaseous components of biogas (Amani et al. 2010). Due to the inherent problems associated with dealing with the anaerobic microorganisms as well as involvement of a variety of microbes growing in close unison in syntrophic manner, it has been somewhat difficult to isolate and identify individual microorganism involved in biomethanation process by the conventional techniques. Still, a lot of work has been done in this direction, and the major groups of microbes involved in anaerobic digestion of organic matter are known.

The molecular biology techniques have been very useful in analyzing the microflora involved in biogas production. Some of the commonly used methods involved 16S rRNA analysis using 454 next-generation sequencing (NGS) technique (Zakrzewski et al. 2012), terminal restriction fragment length polymorphism (Wang et al. 2010), and quantitative real-time polymerase chain reaction (qPCR) (VanGuilder et al. 2008). While the conventional methods target primarily the dominant microflora, the metagenomic and metatranscriptomic approaches allow analysis of even lesser abundant microflora and go a long way in comprehending the microbial community dynamics of biogas production as well as provide insights into the genetic and metabolic capabilities of the microflora (Sárvári Horváth et al. 2016; Bremges et al. 2015). Greater understanding about the microbial community involved in biogas production and their dynamics will help in potentially enhancing the production of biogas.

Some workers have tried using addition of microbial inoculants for enhancing the biogas production. Bagi et al. (2007) reported that the addition of a pure culture



of *Caldicellulosiruptor saccharolyticus* (a cellulolytic, H<sub>2</sub>-producing bacterium) to various substrates like sewage sludge, plant wastes, and animal wastes leads to significant increase in biogas production. Similar results were also obtained when a mixture of these substrates were inoculated with *C. saccharolyticus* culture. However, the use of pure cultures is relatively less common, and most of the workers have focused on utilizing microbial consortia. A thermophilic consortium, obtained from various composting materials like sugarcane dregs, dried straw, and fecal material of chicken, pigs, and cattle, was efficiently degrading ligno-cellulosic substrates like rice straw, corn stalk, cassava residues, etc. (Haruta et al. 2002; Guo et al. 2011). Wei et al. (2010) reported enhanced level of biogas production by utilization of a hemicellulose-degrading microbial consortium comprising *Bacteroides* sp., *Dechlorosoma* sp., and a diverse range of Clostridiales immobilized on zeolite. Dhadse et al. (2012) obtained maximum biogas production (with 76% methane) from a consortium containing four different methanogenic bacteria, while the other two bacterial consortia containing facultative anaerobes result in lower yields of biogas. Gopinath et al. (2014) analyzed the effect of four different microbial consortia obtained from cow dung on biogas production with poultry droppings as a substrate and reported that the consortium number four resulted in maximum methane yield of 79.4%. They reported this consortium to have high concentration of methanogenic archaea. Poszytek et al. (2016) developed a microbial consortium with high cellulolytic activity, comprising 16 strains belonging to the genera *Bacillus*, *Providencia*, and *Ochrobactrum*, which lead to higher efficiency of maize silage degradation as well as higher biogas production under two-phase sequencing reactor. Suksong et al. (2019) compared two thermotolerant microbial consortia, one of which was rich in Lachnospiraceae and the other was rich in Clostridiaceae members for biogas production from palm oil empty fruit bunches. While the former was observed to be more suitable for pre-hydrolysis, the latter was found to be more suitable for direct bioaugmentation during solid-state anaerobic digestion. Tantayotai et al. (2019) have reported 6.5 times enhancement in biogas production from activated wastewater sludge and rice straw residues with the help of an ionic-liquid-tolerant and salt-tolerant consortium primarily comprising Bacteroidetes, Actinobacteria, and Methanosarcinales.

### 12.4.3 Modifications of Biogas Production Process

The main constituents of the feedstock are lignocellulosic wastes composed of cellulose, hemicellulose, and lignin as discussed before. Kumar and Sharma (2017) have summarized the percentage of these three constituents in various plant sources like sugarcane, wood, newspaper, etc. The yield from a biogas plant is dependent upon:

- (a) The type of the biogas plant
- (b) The concentration of the biomass fed into the biogas plant
- (c) The conditions of the AD process

### 12.4.3.1 Pretreatment Technology for AD

The present knowledge on pretreatment technology has only recently been investigated and needs to be optimized before use in terms of not only application and efficiency but also in regard to the economic burden. It must be more integrated into the biogas production process rather than being considered as an enhancement or a separate process.

- i. *The primary aims of pretreatment of biogas feedstock are the following:*
  - a. Avoid the failure of the production process.
  - b. Enhance the production efficiency of biogas production—AD is faster.
  - c. Lower the methane emission into the surroundings, making the process more environmental friendly.
  - d. Increase the yield of biogas from the given feedstock.
  - e. Make the feedstock substrates more accessible to the microorganisms for degradation by converting the substrates partially or completely into fermentable sugars.
  - f. Overcome the recalcitrance of cellulose-lignocellulosic substrate that is a complex structure to break down by the microorganisms, making its degradation difficult and expensive to the biogas refineries. Recent trends are exploring the potential of genetic engineering approaches to solve recalcitrance problems (Abramson et al. 2013).
- ii. *The selection criteria for the pretreatment process are based upon the following factors (Wyman 1999):*
  - a. Avoid a method that leads to the reduction in particle size of biomass.
  - b. The chosen method must aim to preserve the hemicellulose fraction.
  - c. There should be minimal formation of the degradation products.
  - d. The energy demands of the chosen method must be minimal.
  - e. The method must select a cost-effective catalyst for pretreatment and should involve a low-cost pretreatment catalyst and/or inexpensive catalyst recycle and regeneration of high-value lignin co-product.
- iii. Types of pretreatment methods for overcoming recalcitrance in biomass can be broadly divided into biochemical and thermochemical methods and more precisely into physical, mechanical, physico-mechanical, and biological methods as summarized in Table 12.6 (ATV-DVWK 2003; Mshandete et al. 2006; Laser et al. 2009; Kumar and Sharma 2017).

### 12.4.3.2 Multiple Stage AD

Modern-day technology for improving the stability of the biogas production and efficiency of the bioreactor systems has been explored to segregate the processes of AD into hydrolysis–acidogenesis in one chamber and acetogenesis–methanation in

**Table 12.6** Broad classification of types of biomass pretreatment methods

Biochemical methods		Thermochemical methods	
<i>Advantage:</i> a. High specificity in deconstruction of biomass b. Desired product formation <i>Disadvantage:</i> a. Should be coupled with thermochemical pretreatment of low severity		<i>Advantage:</i> c. Fast process d. Low residence time e. Uses a wide variety of feedstocks in a continuous manner <i>Disadvantage:</i> b. Nonspecific deconstruction of biomass	
<i>Types of pretreatment methods based on the catalyst used:</i>			
Physical/mechanical methods	Chemical methods	Physicochemical and thermal methods	Biological and enzymatic methods
a. Mechanical extrusion b. Milling c. Microwave d. Ultrasound e. Pyrolysis f. Pulsed electric field	a. Dilute acid b. Mild alkali c. Ozonolysis d. Organosolv e. Ionic liquids f. Deep eutectic solvents g. Natural deep eutectic solvents	a. Steam explosion b. Liquid hot water c. Ammonia based d. CO <sub>2</sub> explosion e. Oxidative pretreatment f. Wet oxidation g. SPORL	a. Fungi (brown, white, soft rot) b. Bacterial c. Archaeal d. Enzymes—cellulase, hemicellulase, cellobiase, pectinase, proteases, etc. (Hosseini Koupaie et al. 2019)

another chamber of a bioreactor. Such a multiple stage bioreactor system offers the advantage to micro-manipulate the environments and feedstock loading rate for the two chambers of the bioreactor but has the drawback of being more complex and costlier, making it less feasible for commercial use (Vandevivere et al. 2002; US EPA 2006; California EPA 2008; Yu et al. 2013). Recent investigations have reported that multiple stage AD bioreactor shows higher efficiency of COD removal and production of biogas (Colussi et al. 2013), accelerated hydrolysis and faster degradation of biomass (Marín Pérez and Weber 2013), ammonia inhibition (Yabu et al. 2011), higher methane yield (Park et al. 2008), etc. A four-stage AD bioreactor coupled with activated sludge has also been used (Kim et al. 2011) that demonstrated higher AD efficiency than a one-step system. Similarly, higher growth rates of methanogenic bacteria lead to increased biogas production (Blonskaja et al. 2003) and bio-hydrogen production (Nasr et al. 2012).

### 12.4.3.3 High-Pressure AD

This technique works on the principle of increasing the working pressure within the bioreactor up to 100 bar (1 bar = 100 kPa) resulting in higher methane content (>95%) and less than 5% CO<sub>2</sub> in the biogas produced. Hence, the biogas production is integrated with the in situ high-pressure purification as a sole process that produces clean biogas with 99% methane which may be utilized for domestic and commercial applications (Bartlett 2002; Lindeboom et al. 2011; Merkle et al. 2014,

2017). This technology has shown promising results in quality biogas production but requires further research to understand the pressure effects on the growth of the microbiome.

#### 12.4.3.4 Modulating Methanogenesis

Among the four steps of AD, methanogenesis is the most critical and rate-limiting step due to slowest growth rate of the methanogens and their sensitivity to environmental factors like pH, temperature, ionic strength, and various inhibitors (Chen et al. 2008). NGS in combination with genetic engineering approaches to modify the efficiency of the metabolic pathways in the microbes may be considered to decrease the economic burden of the biogas reactors, making their use more commercially applicable. These approaches may also bear potential to improve the quality and energy output of the biofuels (Xu and Koffas 2010). Metabolic redirection has been applied in production of bioethanol in which the production of undesirable metabolic products is limited and the metabolism of the bacterial cell is directed toward the formation of targeted products (Weng et al. 2008).

#### 12.4.3.5 Start-Up Period for the AD

It is critical for every biogas reactor to have a start-up time period in order to produce an efficient, continuous, and stable supply of biogas (Escudié et al. 2011; Kim et al. 2013; Goberna et al. 2015). It is the time required by the microbes to grow and multiply while feeding upon a specific waste, till their population becomes redundant and stable. The time period hence depends upon the microbial population and the type of biomass it feeds on. If the start-up period is neglected, the biomass degradation may be incomplete, leading to accumulated intermediaries like VFA, inhibition of methanogenesis and thus inefficient productivity of biogas, and finally operation failures of the biogas reactor systems (Griffin et al. 1998; Liu et al. 2002; Escudié et al. 2011). Hence, one must keep in mind the calculation of the start-up time period considering the type of organic waste, rate of loading the feedstock, the ratio of inoculum to substrate, temperature conditions of the bioreactor, type of the reactor, etc. Next-generation sequencing (NGS) may be used as an efficient tool to screen the dynamics of complicated microbial community. NGS also aids in monitoring the successful establishment of AD start-up in terms of microbial communities inside the bioreactors, besides elucidating the degradation pathways in the process of biogas production constituted in the microbiome (Appels et al. 2011). Therefore, such tools must be utilized besides the physicochemical monitoring AD start-up.

### ***12.4.4 Solid-State Anaerobic Digestion***

Anaerobic digestion in a biogas digester is operated in two modes: digestion in slurry phase and solid-state anaerobic digestion. The classification is based upon the available solid content in substrate feedstock (Li et al. 2011b). Slurry phase operates at a total solid content of less than 15%, and solid-state digestion is carried at a total solid content of more than 15% (Brown et al. 2012). The advantages of solid-state anaerobic digestion include requirement of small reactor volume, less moving assembly, low cost-energy requirement, less water wastage, and easy handling (Brown et al. 2012; Guendouz et al. 2008; Cheng et al. 2010). Solid-state fermentation is required with substrates feedstocks rich in lignocellulosic content those contain less moisture (Brown et al. 2012; Singhanian et al. 2009). However, due to low moisture availability and complex structure, these substrates are hard to hydrolyze and require pretreatment for easy digestion (Taherzadeh and Karimi 2008; Teghammar et al. 2012).

The crystalline structure of cellulose and the presence of resistant fraction lignin are the important factors determining degradation of lignocellulosic waste (Puri 1984; Chang and Holtzapple 2000; Laureano-Perez et al. 2005). The lignin forms a cross-linked structure in between the carbohydrates, and the presence of this matrix restricts the lignocellulosic substrate digestion. This meshwork is resistant to degradation by enzymes and microbes (Pooornejad et al. 2014; Kumar 2014). Therefore, lignin degradation is required for biomethane production from lignocellulosic waste. Ethanol is employed for lignin removal as a pretreatment during biodegradation of lignocellulosic waste (Zhao et al. 2009; Binod et al. 2010). Lignin is itself a valuable by-product; therefore, the use of organic solvent as a pretreatment method helps to separate lignin in unaltered form, increase the biogas yield, and ultimately improve economy of the process (Zhao et al. 2009; Obama et al. 2012).

Biogas production was examined over a range of total solid concentration, and most efficient digestion was reported at a total solid content of 13.5% (Singh et al. 1984). Similarly, Pathak et al. (1985) observed biogas yield per gram of solid consumed with solid content of manure slurry at 7.7, 10.2, and 14.8% respectively. Various studies have also highlighted a high biogas yield at total solid contents ranging between 5 and 20% (Itodo and Awulu 1999; Itodo et al. 2001; Malik et al. 2008; Leela Wati et al. 2008).

### ***12.4.5 Improvement in Biogas Purification***

The anaerobic digestion of organic materials leads to production of biogas which is basically a mixture of gases, methane being the most dominant as well as the desired component. Apart from methane, other gases include carbon dioxide (CO<sub>2</sub>), hydrogen sulfide (H<sub>2</sub>S), ammonia (NH<sub>3</sub>), and water vapors. In order to enhance the efficiency of energy yield from biogas, it is imperative that the unwanted components of

biogas be removed and methane be enriched because, ultimately, it is methane which is going to act as the energy source (Lohani et al. 2010). Moreover, the corrosiveness of hydrogen sulfide will interfere with the storage and transfer of biogas through metallic components. Similarly, water may also interfere with the efficient utilization of biogas and additionally can cause rusting of metallic components. The presence of CO<sub>2</sub> lowers the energy content of the biogas, and its compression leads to higher energy inputs (Bari 1996; Appels et al. 2008). Biogas upgrading is especially needed for utilization in vehicles and fuel cells (Kapadi et al. 2005).

Some of the methods which have been reported to be used for CO<sub>2</sub> removal include water scrubbing, pressure swing adsorption (PSA) with activated carbon or molecular sieves, physical absorption, chemical absorption, adsorption on a solid surface, membrane separation, cryogenic separation, etc. (Ryckebosch et al. 2011; Morero et al. 2017).

Water scrubbing is regarded as one of the relatively simple, economic, and practical methods for CO<sub>2</sub> removal from biogas especially in the rural areas. This method has the capability of removing CO<sub>2</sub> and H<sub>2</sub>S as well (Wellinger and Lindeberg 1999). Organic solvents like methanol and dimethyl ethers of polyethylene glycol (DMPEG) have also been employed. These also have the capacity of removing CO<sub>2</sub>, H<sub>2</sub>S, and H<sub>2</sub>O (Tock et al. 2010). Methane concentrations as high as 96–98.5% have been reported in these techniques (Bauer et al. 2013a; Sun et al. 2015).

Chemical absorption generally uses either aqueous solution of amines or aqueous solution of alkaline salts (Al-Baghli et al. 2001; Razi et al. 2013). Some of the amines which have been used include monoethanolamine, diglycolamine, diethanolamine, triethanolamine, methyldiethanolamine, and piperazine. Another category of chemicals includes caustic solvents (sodium hydroxide, potassium hydroxide, and calcium hydroxide). In this process, CO<sub>2</sub> content of biogas is reduced from about 40% to 0.5–1.0%. However, the processes are still to be standardized for large-scale operation in biogas purification, and there are many technical problems like high energy requirements as well as solvent recovery issues (Abdeen et al. 2016). The PSA process makes use of vertical columns packed with adsorbents under adsorption, depressurization, desorption, and pressurization sequences (Yeh et al. 2001). The most commonly used adsorbents are zeolite, activated carbon, activated charcoal, silica gel, and synthetic resins. This can be used to separate CO<sub>2</sub>, N<sub>2</sub>, O<sub>2</sub>, and H<sub>2</sub>S (Ryckebosch et al. 2011). Linking of several columns has been reported to reduce energy need for operation (Bauer et al. 2013b). Membrane separation process makes use of a thin membrane, which is more permeable to some biogas components than the others. These may be operated at higher pressure ranges of greater than 20–40 bar or at lower pressure range of 8–10 bar and are capable of removing CO<sub>2</sub>, H<sub>2</sub>S, H<sub>2</sub>O, and O<sub>2</sub> (Bauer et al. 2013a). Some workers have reported that efforts to obtain higher purity lead to methane losses (Persson et al. 2007; Sun et al. 2015); that is why multi-phase membrane separation systems have been suggested by Scholz et al. (2013). The cryogenic separation is a relatively newer technology and relies on the difference in the boiling points of various gaseous components of biogas. In this process, biogas is cooled with chillers nearly to –45 °C at elevated pressure. The process is especially useful for obtaining liquid

biomethane with high purity and relatively with less than 1% losses (Hosseini and Wahid 2014). The product may be directly used for vehicles or injected to grid as gas. However, the energy consumption of the process is relatively high (Johnston 2014; Sun et al. 2015).

For removal of  $H_2S$ , some of the suggested methods include iron oxide adsorption, liquid phase oxidation process, lime scrubbing, air injection, iron chloride addition, dry oxidation process, and liquid phase oxidation (Shah et al. 2016). A variety of chemicals like NaOH,  $FeCl_2$ ,  $Fe^{3+}/MgO$ ,  $Fe(OH)_3$ ,  $Fe^{3+}/CuSO_4$  and  $Fe^{3+}/EDTA$  (ethylenediaminetetraacetate), activated carbon, and zeolite have been used in different techniques (Cosoli et al. 2008; Ryckebosch et al. 2011; Sun et al. 2015). However, a major obstacle in case of  $H_2S$  is its toxic and corrosive nature. So, more research efforts are needed to tackle this problem. One possible approach is to develop a system that can combine multiple technologies for removal of  $H_2S$ ,  $CO_2$ , and other contaminants. Another important development in the direction of making the overall process more efficient is the employment of lithotrophic sulfur oxidizing microbial agents for desulfurization and biofiltration of  $H_2S$ . Some of the possible microbial candidates include *Thiobacillus*, *Paracoccus*, *Acidithiobacillus*, and *Halothiobacillus* (Montebello 2013; Mora et al. 2014).

For removal of water vapors, both physical drying and chemical drying have been used. The physical drying methods by condensation are demisters, cyclone separators, moisture traps, and water traps (Persson et al. 2007; Bauer et al. 2013b). The chemical drying can be carried out with the help of glycol, silica gel, magnesium oxide, activated carbon, and alumina (Bailón Allegue and Hinge 2012; Awe et al. 2017).

## 12.5 Conclusions and Future Perspectives

The anaerobic digestion processes for the production of biogas are poorly understood due to their complexity and are often linked to the risk of failure in terms of large-scale investments. But the growing need for the development of biofuels has shifted the R&D toward the exploration of transportation fossil fuels' replacements like biomethane. The present-day research on biofuel technology faces the challenges in terms of technical understanding, economical burden, and ecological impacts. The microbial strains and their inoculum/substrate ratio, the types of catalyst chosen, the kind of AD bioreactor, the substrate composition, the start-up period, and the pretreatment process all have to be optimized in terms of efficiency, process stability, and cost-effectiveness. So, it can be concluded that although biomethanation offers a wide range of advantages, a lot of research efforts are required in this direction to enhance the applicability and appeal of this process. It is high time that we put in maximum efforts in developing and modernizing the biogas production for the benefit of not only the mankind but the entire globe.



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