



Review Paper

A Review on Trends issues and Prospects for Biogas Production in Developing Countries

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Abstract

During this century, sustainable development programs focusing on global environmental concerns such as conservation of energy, preservation of water and management of environment through appropriate alternatives and technologies. In this context, biogas production technology has been developed and proven successful presently. In recent past, research on this technology has limited on some instances like optimization of different substrate and parameters like temperature, pH, total solid concentration, retention time, etc. A critical analysis reveals that few issues associated with the system used for biogas production at present, while various prospects lifted during the review gives an opportunity to enhance biogas production using advance techniques in future. Pretreatment of lignocellulosic substrate, incorporation of biological, chemical or inorganic additives and enzymes to the substrate were found as effective for increasing biogas production. Another interesting trend in this field is the development of potential microorganism for enhancing biogas production through gene manipulation. Multi-stage processes with separation of hydrolysis, acidogenesis, acetogenesis and methanogenesis would provide biological stability for each step which absolutely enforces the reaction at temporal dimension.

Keywords: Anaerobic digestion, biogas, pretreatment, additives, biodigester.

Introduction

A major environmental issue in today is associated with the release of green house gases¹. Fossil fuels are the major contributors for green house effect and at the same time its depletion occurs rapidly in each advancing year. In order to minimize this global energy demand and environmental impacts, an alternate energy source is needed^{2,3}. The production of biogas from biomass has been identified and used as renewable high energy biofuel from past few decades for various purposes. Biogas production technology in India has been started from 1950s whereas at the moment this technology never becomes popularized. Compared to other developing countries, biogas production in India has some limitations and failure rate was reported previously. While the energy crisis in 1970, does promote many countries to investigate an alternative energy source through feasible technology. The emergent energy crisis in the late 1970s in India direct towards the development of national biogas programme⁴. Therefore by the early 1980s about 80,000 small scale biogas digesters were built in India. Even though, the exploration of this technology in India has yet to attain its extreme. This may be due to the lack of knowledge and awareness of this technology to villagers. Also comparatively simple techniques are used to arrest biogas in developing countries at that time, while the techniques could not be fully trapped the potential biogas. Furthermore, modification in the technology was desired to get rid of diverse

confines and in consequence to expand the technology in all developing countries in the meanwhile.

Anaerobic digestion is a promising method for biogas production in which organic substances are converted in to biogas through the sequential involvement of different groups of bacteria⁵. Conversely, biogas production by this pathway has various excellent opportunities to use. Well functioning biogas system can yield whole range of benefits to the users, the society and the environment in future. The benefits of biogas production technology through anaerobic digestion is it offers an alternative fuel, heat and electricity, waste management system, good fertilizer, complete recycling of wastes, green house gas reduction and environmental protection from pollutants⁶. These strategies focused on the improvement of biogas production technology all over the world. There are many factors influencing the performance of anaerobic digestion process. These are mainly correlated to the characteristics of feeding material, type of biodigester and operational conditions used for the process in real time. Characteristics of the material are considerable because they affect the process stability and biogas production during anaerobic digestion. These include rate of humidity, volatile solid content, carbon-nitrogen ratio, magnitude of particle and degradable nature of feedstock. The feasible circumstances for instance temperature, pH, loading rate and retention time could also maintain to increase biogas production^{7,8}. However all these

practice bring in to being only little increase in its production instead of predictable gas production.

From this background, the present review had framed to analyze confronts related to traditional biogas production systems and relevant shifts for biogas production in current scenario. It is the time that various technological modifications have been developed in order to enhance biogas production. Pretreatment and modification of feeding materials with various additives would possess an advantage for enhancing speed of fermentation process^{9,10}. Gene manipulation of microorganisms and development of pertinent biogas plants would be expected to provide a great opportunity for biogas production at current position^{11,12}. However, few concerns linked with present trends and it is crucial to overcome the issues associated with biogas production through above aspects. Hence in connection to the issues explored, current review also encounters the prospects raised from each phase with the aim of eliminating the entire constraint of biogas production technology in future.

Anaerobic Digestion Process

Anaerobic digestion refers to bacteria-aided degradation of organic materials with the lack of oxygen under controlled conditions. The entire process carried out in an airtight tank called digester in which organic waste has to be dumped and decomposed. Bacteria within the digester decompose the organic matter that results in the production of gases such as methane, carbon dioxide, hydrogen sulfide, carbon monoxide, ammonia and nitrogen¹³. Methane is the prominent portion of biogas, is a flammable gas that can be substitute as an alternative energy source for various purposes⁵. Those bacteria

feed upon the initial feedstock and as a result the feedstock becomes converted to simpler compounds and finally it course in the evolution of biogas through various intermediate steps. Several bacteria synchronize together to driven different stages of anaerobic digestion in sequence to obtain absolute biogas from substrate¹⁴.

The digestion process encompasses four stages such as hydrolysis, acidogenesis, acetogenesis, methanogenesis and the organism involved in each stage were categorized as hydrolyser, acidogen, acetogen and methanogen, respectively. An outline of major pathways occur during anaerobic digestion process is shown in figure-1. The initial stage is hydrolysis in which depolymerization of the organic matter occurs. Accordingly, polymeric compounds present in the substrate like carbohydrates, proteins and lipids are being converted to monomeric compounds like sugars, amino acids, peptides and long chain fatty acids by means of enzymes formed by hydrolytic bacteria. Cellulase, protease, lipase, amylase, etc were recognized as the major hydrolytic enzymes produced by hydrolysers. In the second stage, acidogens convert the products obtained from hydrolysis in to organic acids like long-chain (propionic, butyric, valeric acids) and short chain (acetic acid) fatty acids along with alcohols, carbon dioxide and hydrogen. Acetogenesis, the third stage result in the formation of formic acid, carbon dioxide, acetic acid and hydrogen with the contribution of acetogens from long chain fatty acids obtained during acidogenesis. During the final stage methanogenesis, obligate anaerobic methanogens fulfill the conversion of hydrogen and carbon dioxide in to biogas under severe anaerobic conditions⁶.

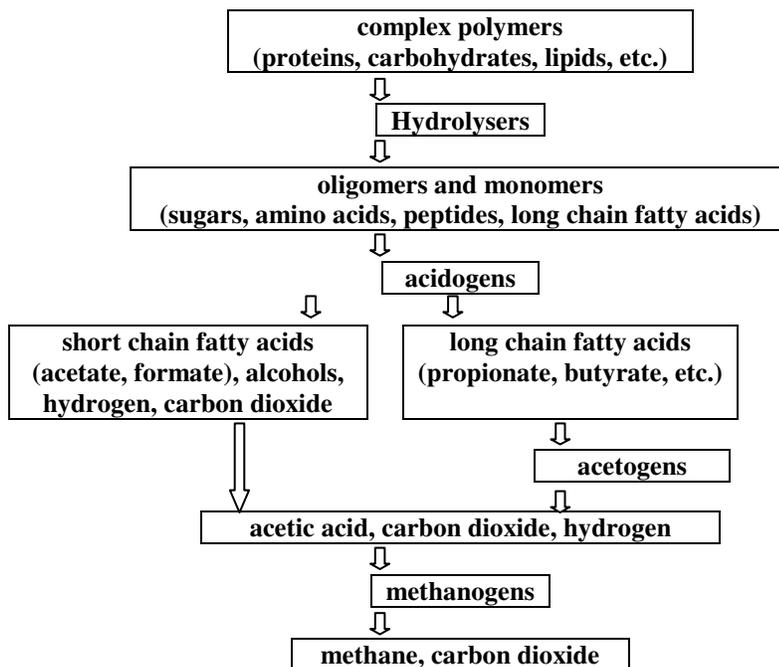


Figure-1
 Outline of anaerobic digestion process for biogas production

Successful anaerobic digestion ensues only if the digester contains four groups of bacteria for appropriate stages. Previous studies reported that *Acetivibrio sp.*, *Clostridium sp.*, *Syntrophomonas sp.*, *Methanosarcina sp.*, were the prominent bacteria for hydrolysis, acidogenesis, acetogenesis and methanogenesis, respectively^{15,16,17}. For obtaining better biogas production, all the parameters such as feedstock characteristics, hydrogen ion concentration, temperature, rate of substrate loading, hydraulic retention time, total solid and volatile solid contents, C: N ratio, digester model, etc should have to be maintain during anaerobic digestion⁴. A slight change in parameters could adversely affect the whole system. Thus the exploration of this technology by standardization of all the parameters requires coordination of scientific and engineering principles in meanwhile.

The main advantage of anaerobic digestion is it generates biogas which is the perfect energy source. Biogas production from wastes and agricultural byproducts by anaerobic digestion has waste reduction potential. The complete degradation of manure for biogas production provides a great extent to reduce pathogen content and odour of the manure. Biogas burn more cleanly than coal and emits less carbon dioxide. Carbon dioxide is the second most causative agent of global warming while biogas production technology driven capture of it along with the major green house gas methane instead of its release to the environment and thereby reduce environmental pollution. The other major advantage of this technology is it augments the development of rural wealth. The expansion of this technology in certain developing countries has resulted industrialization of the area and which provided more jobs to the people¹⁸. According to the review, industrialization of this technology ensures manifold benefits to the consumer along with development of the nation. Hence biogas production from organic materials would provide admirable option in outlook.

Previous constraints of biogas production

Anaerobic digestion is known as effective method for waste treatment from past few decades. Since, it was relatively slow process in view of the fact that simple methods were used to detain the technology. Few studies revealed that long hydraulic retention time of around 30-50 days are required for conventional biogas plants¹⁹. It is also reported that instability of the process, lesser feedstock loading rate and more time taken for system recovery subsequent to breakdown are the major limitations with most of the biogas production systems in earlier⁸. Another problem related to gas production is that there is a drop in the production during winter season due to the fall in temperature²⁰.

During past 35 years, the anaerobic digestion process has been known and put to work in a number of different applications²¹. High throughput screening on former studies showed that biogas production from organic wastes restricted in some aspects like pH, operating temperature, loading rate, retention time,

composition of organic wastes and special microbial consortium^{22,4}. Improper handling of all these parameters causes unsteadiness of the process. Maintenance of C: N ratio of the feedstock, total solids, volatile solids, etc could provide considerable increase in gas production^{23,7}. Design of biodigester is another important criterion. Traditionally used fixed dome biogas plant reported decreased gas production when compared to floating dome biogas plant²⁴. Co-digestion of different substrate and the addition of urea to the substrate were also provided good results²⁵.

The optimum circumstances at which the microorganism grow is of paramount importance in the anaerobic digestion process. Growth requirements such as pH, temperature, carbon source, nitrogen source etc have to be maintained to achieve maximum growth and activity of microbes²⁶. Thus process optimization could satisfy growth requirements of microorganisms and hence it could also boost the anaerobic digestion efficiency of the system. Feed to inoculums ratio is one more factor affecting anaerobic digestion. Highest gas production was reported in the system with lower feed to inoculums ratio²⁷. Further relevant techniques are needed to run the system with higher feed to inoculums ratio in order to enhance the quantity of biogas. However, only a negligible increase of biogas production was achieved from these studies. Hence this could be possible to fulfill the constraints throughout the process by introducing novel technology.

Pretreatment of lignocelluloses

Biomass has been known as the cheap source of renewable energy from past few decades. Lignocelluloses comprise the most part of biomass²⁸. Lignocellulosic materials include agricultural residues and forest products represent the most abundant renewable energy resource available on earth. They are composed of three types of components including cellulose, hemicelluloses and lignin, which are interlinked together to form a crystallized structure. Generally the degradation of lignocelluloses by bacteria is very difficult due to its structural complexity and high molecular weight²⁹. Hence appropriate pretreatment is essential, that improves the accessibility of hydrolytic enzymes by substrate which results solubilization of complex lignocelluloses and thereby increasing the efficiency of biogas production³⁰.

There are several pretreatment methods have been proposed in recent years. These are physical (mechanical and non-mechanical), chemical (acid or alkaline hydrolysis, oxidative delignification and solvent extraction), physico-chemical (ammonia fiber explosion and CO₂ and steam explosion), and biological (microbial treatment) pretreatments^{31,32}. However, the physical pretreatment is far too expensive and physico-chemical treatment requires high energy and suitable system that increases the cost of the total process. Certain inhibitors formed during physico-chemical treatment may severely affect the growth of microorganisms³³. Pretreatment of petrochemical

wastewater with hydrogen peroxide showed an increase of its biodegradability and decrease of hydraulic retention time during anaerobic digestion³⁴. Even as in the case of chemical pretreatment, acidic or alkaline water generated may be unfavorable to methanogen. Hence appropriate disposal of toxic water after pretreatment is essential to grant environmental safety⁹. Any technology could arise in the current economical system should compete with available technology by the way that treating society. Moreover the technology should be eco-friendly and cost effective. While the above described pretreatment methods possess some inconvenience to the society.

Hence an alternative and environmental friendly approach widely accepted for degradation of lignocellulose is biological pretreatment. In recent years, processes that use microorganisms capable of degrading lignocelluloses could lead to a new and environmental friendly pretreatment method. Advantages of biological pretreatment includes it is inexpensive, low energy necessities, environmental safety and mild environmental conditions needed³¹. It has been found that white-rot fungi could be used as the prominent organism for efficient pretreatment of lignocelluloses, that can easily break down lignin into carbon dioxide and water which results in alteration of lignocellulose structures³⁵. White-rot fungi like *Phanerochaete chrysosporium*, *Ceriporiopsis subvermispora*, *Aspergillus sp*, *Trichoderma reesei*, *Coriolus vesicolor*, etc were capable of degrading lignin efficiently^{36,37,38,39}. Even though, a little issue explored on biological pretreatment is that appropriate separation and disposal of fungi after pretreatment is necessary to make further process feasible and also to ensure environmental safety. According to the review, identification of potential fungi and optimization of its utmost enzyme production would prospect towards high pretreatment efficiency. Controlling various parameters and use of pure lignocellulolytic enzymes during pretreatment would also direct towards expected pretreatment efficiency.

Substitution of Additives

Additives refer to the substances which are used to enhance biogas production. Progress could be achieved from some attempts that have been made in the past with various additives such as biological additives (crop residues, microbial cultures etc.), chemical additives (inorganic salts) and enzymes. An enhanced biogas production of 10-90% attained from the digestion of crop residues such as rice straw and wheat straw with partially digested cattle dung⁴⁰. Similarly, biogas production from cattle dung with 1% onion storage waste showed an increment of 40-80% gas production rate⁴¹. Integration of microorganisms was found to enhance gas production significantly. Biologically treated sugarcane bagasse with cattle dung showed higher gas production⁴². Another study used cattle manure with rumen fluid as additive to enhance production, which results that 50% addition of rumen fluid to cattle manure gave better result⁴³.

Addition of some metals such as iron, nickel and cadmium increases methanogenic population during anaerobic digestion and thereby biogas production^{44,45}. Incorporation of iron salts such as ferrous sulphate and ferric chloride in various concentrations along with substrate could also enhance gas production⁴⁶. While the degree of increment with metals was based on the type of feeding material and concentration of metal used. Few studies showed that ammonia is necessitate for maximum growth of some methanogens and thereby biogas production⁴⁷. Addition of urea and diammonium phosphate was found to enhance biogas production rate up to eleven percentage⁴⁸. Absorbents like silica gel, charcoal, etc were relevant for obtaining high methane content during biogas production^{49,50}. The incorporation of sodium hydroxide on feedstock increases its fluidity and also biogas production by this means⁵¹. However, various chemical additives showed substantial increase in biogas production, even as it is significant to standardize the finest concentration of each additive for different substrate for encouraging the whole process.

Generally enzymes are the key factor that facilitates all metabolic reactions. During anaerobic digestion, microorganisms produce different types of enzymes which act on various substrates and finally release potential energy source biogas. Now a day research is going on accelerating biogas production from various substrates using addition of enzymes. From this review, it is observed that addition of cellulase enzyme mixture with a concentration of 450mg yielded more biogas within less time⁵². Considerable differences in feedstock fluidity were also observed by the application of various hydrolytic enzymes such as cellulase, hemicellulase, xylanase, pectinase, lipase, amylase, glucosidase and protease¹⁰. Whilst the activity of enzymes depend on characteristics of the substrate, reaction time, quantity of enzymes and reaction temperature. Furthermore research is needed in the area by locating efficient additives and also by optimizing the conditions for its highest activity in prospect.

Microbial manipulation

Today, development of genetically engineered organism through gene manipulation to convert organic materials into useful products is not far away. Genetic manipulation offers gene transfer from one organism to another organism in order to accomplish desired characteristics to the recipient. There are many traditional approaches that have been used for the genetic improvement of different organisms in earlier of which mutagenesis and conjugation were mostly applicable in the case of bacteria. Mutagenesis is an effective method for gene manipulation through which genetically manipulated mutant strains can be produced from wild strains by exposing them to ultraviolet rays or mutagenic chemicals. Despite the fact that mutagenesis may result in the generation of dangerous organism instead of desired organism, it is identified as the key trouble allied with mutational gene manipulation. Hence appropriate

method for testing of mutants is needed in turn to select desired mutants and destruction of undesired mutants. The process of transfer of genetic material with desired gene among microbial species through corporal contact is referred to as conjugational gene transfer. Although the drawback behind conjugational gene transfer is it has been restricted in closely related species^{53,54}. Conversely the confines of traditional technology for gene manipulation of microbes point towards advanced technology for the meantime.

Comprehension of the genetics of the bacteria involved in biogas production will be of paramount significance because it may perhaps numerous interesting aspects in this field. Advanced techniques like genetic engineering, cloning, recombinant DNA technique etc could provide a platform for genetic manipulation of microorganisms at molecular level; however bacteriophages and plasmids are desirable as vectors for gene transfer⁵³. Nowadays recombinant DNA technique accepted widely due to the fact that the technique allows altering or inactivating specific genes, to obstruct the expression of unwanted genes and endorse the expression of desirable genes in a variety of organisms⁵⁴. But least developments were achieved in genetic engineering of anaerobic bacteria by the reason of insufficient genetic analysis.

In recent past, gene manipulation in fermentative bacteria has been done for improving hydrogen yield by arresting NADH dehydrogenase from *Enterobacter aerogenes* and thiolase from *Clostridium butyricum*¹¹. Similarly a mutant strain of *Methanococcus voltae* obtained after irradiation with UV or gamma rays showed that the strain uptake some promising properties⁵⁵. Transformation of such type mutated genes with desired characteristics in to wild type organisms would also helps to improve biogas production in future. An assessment on genetic manipulation in rumen bacteria could demonstrated that the development of suitable vectors, techniques for introducing chromosomal DNA in to rumen bacteria through conjugation, transformation or transduction and the maintenance and expressions of recombinant plasmids are the major barriers which hinder gene manipulation in rumen microorganisms⁵⁶. However, this review suggests that recombinant DNA technology would be successfully applied for the enhancement of microorganisms in future by resolving problems coupled with it.

Multi-stage digestion systems

Diverse bacterial groups are implicated in the anaerobic digestion of various organic supplies for biogas production. They vary in their growth parameters and nutritional requirements. Since a number of one stage systems with all the four phases put together showed process instability due to fluctuations in pH and accumulation of volatile fatty acids, which may leads to metabolic shock of methanogens. One more disadvantage of single phase digester is it can operate only with low solid content otherwise it would cause process failure¹². An

experimental model batch reactor used for the digestion of vegetable wastes has resulted inconvenient biogas production and also the study point towards the need of continuous reactors in future⁵⁷.

Hence multi-stage bioreactors are needed instead of single stage systems which could prove profitably in recent years. High loading rate and 90-93.5% suspended solid removal was reported in the case of anaerobic sequencing batch reactors. Biodegradation efficiency of 91.5% from sugar beet pulp was achieved from a two-stage pilot scale plant with a differentiation of acidogenesis and methanogenesis. Moreover, this study point out that the proposed model resulted a constraint reduction in the hydraulic retention time⁵⁸. A two-phase anaerobic sequencing batch reactor was also developed with segregation of hydrolysis-acidogenesis and methanogenesis for effective degradation of fruit and vegetable wastes resulted high process stability and considerable biogas production⁵⁹.

A major problem related to biogas production systems is the process instability by means of various factors. These include drop in pH, increase in the concentration volatile fatty acids, over loading, insufficient temperature control and unwanted entry of inhibitors to the system. Appropriate mathematical models are needed in order to diminish the rate limiting step associated with former plants¹². Hill and Barth proposed a model which shows that accumulation of unionized volatile fatty acids formed due to temperature fluctuations in digester during hydrolysis, acidogenesis and methanogenesis of insoluble organics are the rate limiting factors, of which hydrolysis affects the most⁶⁰. Some other models showed that hydrogen partial pressure and pH are the rate limiting factors during acidogenesis^{61,62}.

Furthermore, a model point out that heavy organic loading rate and volatile fatty acid accumulation are the predicted causes of digesters failure during methanogenesis of various substrates⁶³. The above proceedings showed that a slight alteration in the adequate circumstance for each step could drastically affect the whole process. According to this review, the segregation of each of the four steps of anaerobic digestion in to separate digester by providing suitable microorganisms and optimum conditions for each step would facilitate sufficient biogas production with in less time. Hence development of suitable mathematical models that remove rate limiting factors during each step of anaerobic digestion would also possess a significant effect in biogas production within a short period in future.

Conclusion

Anaerobic decomposition of organic matter and evolution of biogas with methane as an economically valued product is today's attractive feature in science and technology research. The present critical review discussed trends and issues associated with biogas production in current scenario whilst few aspects on biogas production point towards strapping prospect

to enhance biogas production. Biological retreatment of lignocellulosic wastes prior to anaerobic digestion found to be promising for increased biogas production. Understanding the dynamics of various additives along with other biochemical parameters would augment the four dominant processes to enable to enhance biogas production efficiency. Discrete experimental approaches also yielded good results with enzymes, microbial substitutions and genetically modified microbes. Several models have been proposed based on various factors shows improved gas production. Though various models and approaches found to be promising, multiphase approach incorporating the above all prospects would be effective for the time being.

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