Chapter One

1.1 Introduction:

A biogas plant is a set-up device that converts fermentable organic matter into a combustible gas and matured organic manure. It works by subjecting the material to microbial decomposition in the absence of air, yielding finally, methane, carbon dioxide, and water. This process is called anaerobic decomposition and was known to occur naturally. It was only during the past hundred years that this secret of nature was unraveled by scientists, who chanced upon this process, which is a source of energy and plant nutrients. Biogas obtained through this process is known by several names like the swamp gas, sewer gas, fuel gas, marsh gas or wet gas, and in India, it is more commonly referred to as goober (cattle dung) gas. Biomethanation in conventional biogas plants is a simple and low-cost process, which can be economically carried out in rural areas, where organic waste is generated aplenty, which otherwise pollutes the environment and poses health hazards. In the recent years, in view of the fuel crisis and the environmental pollution associated with the fossil fuel, biogas has attracted wide attention, and its importance as an efficient and non-polluting energy source is now well appreciated. To a large extent, biogas can be used directly for heating and lighting purposes or in an engine-driven generator to generate electricity. The effluent released from the biogas plant is an excellent fertilizer, which tends to improve the physical properties of the soil such as aeration, and moisture-holding and water infiltration capacities. Biogas plants help in improving the ecology and the environment by providing means for the safe disposal of sewage, and animal and human faeces in the urban and rural areas. It has been estimated that about 980 million Tones (by wet weight basis) of cattle dung is produced in India alone every year, which could suffice for producing over 41 000 million cubic meters of biogas per annum (sufficient to generate 196 MW of electric power). An integrated energy system based on biogas can also help in preventing soil erosion and deforestation. Besides, biogas provides some exciting possibilities and solutions to counter problems of global warming by minimizing fossil fuel consumption and curbing greenhouse gas emissions. Though biogas is a viable and environment-friendly technology, it has not been as popular as it should have been, mainly because of the weaknesses in institutional arrangements.( Navindu Gupta, R S Khoiyangbam January 2011) .

One of the main environmental problems of today’s society is the continuously increasing
production of organic wastes. In many countries, sustainable waste management as well as waste prevention and reduction have become major political priorities, representing an important share of the common efforts to reduce pollution and greenhouse gas emissions and to mitigate global climate changes. Uncontrolled waste dumping is no longer acceptable today and even controlled landfill disposal and incineration of organic wastes are not considered optimal practices, as environmental standards hereof are increasingly stricter and energy recovery and recycling of nutrients and organic matter is aimed (Teodorita Al Seadi October 2008).

1.2 Objectives Of the study:

1. To help in Environmental sanitation.

2. Biogas production considered as clean energy source

3. Biogas production cleans out the plant and animal waste
Chapter Two

2.1 Biogas: A renewable energy source:

Energy sources can be grouped into two main categories, namely, renewable and non-renewable. Renewable energy includes solar, wind, hydro, oceanic, geothermal, biomass, and other sources of energy that are derived from “solar energy”, and can thus be renewed indefinitely in nature. Renewable resources are the inexhaustible resources that have the inherent ability to reappear or replenish themselves by recycling, reproduction, regeneration or replacement. Renewable resources can be replaced within a few human generations. The phrase “few generations” is important because some resources are replaceable on very long, geological time-scales. For instance, rocks, which are recycled in nature, occur several thousands times slower than the rates at
which they are used, and therefore, for all intents, are almost non-renewable. Thus, nonrenewable energy resources are those that are replenished by extremely slow natural cycles or those that for practical purpose are not recycled at all. Solar energy, despite the sun’s definite life period, is considered renewable due to two reasons: first, solar energy is actually supplied faster than we can use, and second, numerous human generations would have evolved before the sun virtually ceased to emit energy. Biogas is a renewable energy source. Like all other renewable energy sources, the energy for biogas generation comes from the sun, through photosynthesis by plants. The plant biomass, the storehouse of solar energy, is used either directly as feedstock or after partial digestion in animal guts to run the biogas plant. Ruminants consume plant biomass in both dry and green forms. The dry fodder consumed is usually in the form of crop straw, residuals of cereals, pulses, and oilseeds that are obtained after the harvesting of the crop produced, while in mountain regions, grasses from permanent pastures and forest areas usually form the feed of these animals. Biogas is obtained from the animal waste after it is subjected to anaerobic digestion in the digesters. Inside the biogas plant, the complex organic polymers, primarily carbohydrates, lipids, and proteins, in the biomass are fermented to produce biogas, which mainly comprises methane and carbon dioxide. In the last few decades, biogas has assumed considerable importance as an alternative to conventional energy sources throughout the world, particularly in developing countries like China and India. (Navindu Gupta, R S Khoiyangbam January 2011)
2.2 Historical development of Biomethanation:

The mysterious appearance of flickering lights and flames from below the surface of swamps was noted by Plinius (Van Brakel 1980). Van Helmont recorded the emanation of an inflammable gas from decaying organic matter in 1630. He listed among 15 different kinds of gases, an inflammable gas that evolves during putrefaction and is also a part of intestinal gases. Shirley is sometimes quoted as having discovered marsh gas (methane) in 1667. Alessandro Volta concluded in 1776 that there was a direct correlation between the amount of decaying organic matter and the amount of flammable gas produced, and that in certain proportions, the gas obtained forms an explosive mixture with air.

In 1801, Cruikshank proved beyond doubt that methane does not contain oxygen. In 1808, Sir Humphry Davy stated that methane was present in the gases produced during the anaerobic digestion of cattle manure. He collected 0.3 litre of methane and twice as much carbon dioxide from cattle manure kept in a retort under vacuum. During 1804–10 Dalton, Henry, and Davy, through their study established the chemical
composition of methane, and confirmed that coal gas was very similar to Volta’s marsh gas. Bunsen (1856) and Hoppe-Seyler (1886) made important contributions to the first microbiological knowledge base on anaerobic digestion. By the time Söhngen wrote his thesis in 1906, it was understood that organic materials were hydrolysed by what we now call enzymes and broken down into alcohols and fatty acids, whereas methane was formed from these products (Van Brakel 1980). The adoption of the anaerobic technology began in the late 19th century and early 20th century, prompted by the sanitation concerns of individuals and municipalities. Anaerobic microbes (Clostridium) were first described by Pasteur during a study on butyric fermentation. Not only did he observe the ability of microbes to grow in the absence of oxygen but also found that oxygen in quite small amounts was toxic. At a time when the experiments of Priestley and Lavoisier established oxygen as the obligatory “staff of life”, Pasteur’s discovery was not readily accepted by his contemporaries. Gayon, a student of Pasteur, fermented manure at 35°C and obtained as much as 100 litres methane per cubic metre of manure. While presenting this finding at the Academy of Science in Paris in 1884, Pasteur concluded that this fermentation process could be a source of heating and lighting. At about the same time (1875), the Dutch farmer Wouter Sluys became the first to use methane for the purpose of illumination. However, the gas was not generated by fermentation, but was the natural gas obtained from a well. From 1860 onwards, the idea of using a septic tank was introduced in sewage purification. Although it was known that methane was formed in these tanks, it was not collected for use. It was only in 1895 that Cameron in England designed a septic tank in which he collected the gas for lighting streets in Exeter, England, while gas from human waste in the Matinga Leper Asylum in Mumbai, India, was used for lighting in 1897. By the 1900s, anaerobic digesters were used in many parts of the world, mostly in anaerobic ponds. In 1904, Travis developed a two-stage process, in which the suspended material from the waste water was charged into a separate “hydrolysing” chamber. It was not until towards the end of the 19th century that methanogenesis was found to be connected to microbial activity. In 1868, Bechamp named the “organism” responsible for methane production from ethanol. In 1876, Herder reported that acetate in sewage sludge was converted stoichiometrically into equal amounts of methane and carbon dioxide (Zehnder 1978). In 1906, Söhngen was able to enrich two distinct acetate-utilizing bacteria, and he found that for mate and hydrogen, along with carbon dioxide, could act as precursors for methane. The development of microbiology as a science led to extensive research by Buswell and others in the 1930s to identify anaerobic bacteria and the conditions that promote methane production. Buswell explained the fate
of nitrogen in anaerobic digestion, the stoichiometry of reaction, the 
production of energy from farm wastes, and the use of the process for 
industrial waste (Buswell and Hatfield 1930, 1936). Barker’s studies 
contributed significantly to our knowledge of methane bacteria, and his 
enriched cultures enabled him to carry out basic biochemical studies 
(Barker 1956). Schnellen in 1947 isolated two methane bacteria, namely, 
*Methanosarcina barker* and *Methanobacterium formicicum*. Numerous 
additional studies led to a better understanding of the importance of 
seeding and pH control in the operation of anaerobic digestion systems.

During the World War II, crude oil shortages led to the rediscovery 
of biogas as an alternative fuel. However, the efforts were short-lived 
with the end of the War and availability of cheap and plentiful oil. From 
the 1940s until the 1970s, biogas technology was largely ignored in North 
America and Europe. The period 1950s to 1970s saw the proliferation of 
small-scale biogas plants in India and China. While most of the world 
wasted biogas during the period, China and India began using it for 
heating, lighting, and cooking.

The energy crisis of 1973 led to an increased interest in biogas. 
India and China took the lead, undertaking massive installations of small 
digesters, with the count in millions. The energy crisis of 1979 triggered 
another round of digester development, aimed at energy production. 
China and India expanded the number of family-size biogas plants and 
started experimenting with large-sized community biogas plants catering 
to the energy needs of the village as a whole. With the passage of time, 
an aerobic digestion was increasingly recognized as an inexpensive 
technology to stabilize organic waste. Waste treatment engineers 
concentrated on developing high-rate anaerobic digestion processes. 
Systems such as up flow anaerobic sludge blanket reactor, anaerobic 
filters, and other systems that immobilized bacteria were developed. In 
recent years, municipal solid waste processing facilities have made 
significant progress towards their commercial applications. A number of 
digestion systems have been developed for high solids content waste.
2.3 What is biogas?

Biogas is a combustible mixture of gases (see table 1) It consists mainly of methane(CH4) and carbon dioxide (CO2) and is formed from the anaerobic bacterial decomposition of organic compounds, i.e. without oxygen. The gases formed are the waste products of the respiration of these decomposer microorganisms and the composition of the gases depends on the substance that is being decomposed. If the material consists of mainly carbohydrates, such as glucose, other simple sugars and high-molecular compounds (polymers) such as cellulose and hemicelluloses, the methane production is low. However, if the fat content is high, the methane production is likewise high. Methane and whatever additional hydrogen there may be makes up the combustible part of biogas. Methane is a colourless andodourless gas with a boiling point of -162°C and it burns with a blue flame. Methane is also the main constituent (77-90%) of natural gas. Chemically, methane belongs to the alkanes and is the simplest possible form of these. At normal temperature and pressure, methane has a density of approximately 0.75 kg/m3. Due to carbon dioxide being somewhat heavier, biogas has a slightly higher density of 1.15 kg/m3. Pure methane has an upper calorific value of 39.8 MJ/m3, which corresponds to 11.06 kWh/m3. If biogas is mixed with 10-20% air, you get explosive air, which as the name indicates -is explosive(Peter Jacob2009). The major component of biogas is methane which represent 50-70% followed by carbon dioxide as 30-50% while nitrogen, hydrogen, hydrogen sulfide and water vapour are other component found in small amounts (Biogas China, 2006).

Methane is a colourless and odourless gas but it is 21 times more harmful than carbon dioxide. The uncontrolled emission of methane gas tends to trap heat in the atmosphere and lead to the greenhouse effect or global warming. (Agung and Tekun, 2005).
Table (1): Composition of biogas. The actual make-up depends on what is being decomposed

<table>
<thead>
<tr>
<th>Gas</th>
<th>%</th>
</tr>
</thead>
<tbody>
<tr>
<td>Methane (CH4)</td>
<td>55 – 70</td>
</tr>
<tr>
<td>Carbon dioxide (CO2)</td>
<td>30 – 45</td>
</tr>
<tr>
<td>Hydrogen sulphide (H2S)</td>
<td></td>
</tr>
<tr>
<td>Hydrogen (H2)</td>
<td>1 – 2</td>
</tr>
<tr>
<td>Ammonia (NH3)</td>
<td></td>
</tr>
<tr>
<td>Carbon monoxide (CO)</td>
<td>trace</td>
</tr>
<tr>
<td>Nitrogen (N2)</td>
<td>trace</td>
</tr>
</tbody>
</table>

2.4 The impact of methane on the climate:

Human activities such as cattle farming, rice farming and the accumulation of waste on large waste disposal sites, etc., are thought to have caused a doubling of the atmospheric concentration of methane to the present level of 1.7 ppm. This does not, as such, have any effect on human health. However, as methane is part of the chemical processes in the atmosphere and is also a powerful greenhouse gas (about 22 times as powerful as carbon dioxide, CO2), the gas is a contributor to the greenhouse effect. Methane has thus contributed about 20% to the total increase in the greenhouse effect caused by human activities. If the greenhouse effect results in an increasing temperature rise on our planet, there is a risk that the large areas of tundra currently under permafrost will slowly thaw, which will result in the release of huge quantities of methane when organic materials are gradually decomposed. This will obviously further exacerbate the greenhouse effect. Methane in large quantities also destroys ozone. Rising methane emissions can therefore have unfortunate consequences for the ozone layer that helps protect the Earth against the harmful ultraviolet radiation from the sun (Peter Jacob 2009)
2.5 How is biogas produced?

Biogas is produced naturally in swamps, bogs, rice paddies, etc., and in the sediment at the bottom of lakes and oceans where anaerobic conditions prevail at a certain depth. Methane is also created in the rumen of ruminant animals (cows, sheep, deer, camels, lamas, etc.) (Peter Jacob 2009)

Biogas in developing countries

A number of developing countries use biogas extensively. In India and China alone, there are more than one million small, simple plants, each treating waste (sewage, animal manure, crop residues, etc.) from a single household. The plants are dug into the ground and are unheated. The biogas is used in the housekeeping for cooking and the digested biomass is used as fertilizer (Peter Jacob 2009)

2.6 The biogas process:

The complete biological decomposition of organic matter to methane (CH4) and carbon dioxide (CO2) under oxygen-depleted conditions – i.e. anaerobic – is complicated and is an interaction between a number of different bacteria that are each responsible for their part of the task. What may be a waste product from some bacteria could be a substrate (or food) for others, and in this way the bacteria are interdependent. Compared with the aerobic (oxygen-rich) decomposition of organic matter, the energy yield of the anaerobic process is far smaller. The decomposition of, for example, glucose will under aerobic conditions give a net yield of 38 ATP molecules, while anaerobic decomposition will yield only 2ATP molecules. This means that the growth rate of anaerobic bacteria is considerably lower than that of aerobic bacteria and that the production of biomass (in the form of living bacteria) is less per gram decomposed organic matter. Where aerobic decomposition of 1 g substance results in the production of 0.5 g biomass, the yield under anaerobic conditions is only 0.1 g biomass. The biogas process is often divided into three steps: Hydrolysis, a cidogenesis and methanogenesis, where different groups of bacteria are each responsible for a step (Peter Jacob 2009)
2.6.1 Hydrolysis

During hydrolysis long-chain molecules, such as protein, carbohydrate and fat polymers, are broken down to monomers (small molecules). Different specialized bacteria produce a number of specific enzymes that catalyse the decomposition, and the process is extracellular – i.e., it takes place outside the bacterial cell in the surrounding liquid. Proteins, simple sugars and starch hydrolyze easily under anaerobic conditions. Other polymeric carbon compounds somewhat more slowly, while lignin, which is an important plant component, cannot be decomposed under anaerobic conditions at all. Cellulose – a polymer composed of a number of glucose molecules – and hemicelluloses – composed of a number of other sugars – are complex polysaccharides that, are easily hydrolyzed by specialized bacteria. In plant tissue both cellulose and hemicelluloses are tightly packed in lignin and are therefore difficult for bacteria to get at. This is why only approx. 40% of the cellulose and hemicelluloses in pig slurry is decomposed in the biogas process. Normally the decomposition of organic matter to methane and carbon dioxide is not absolute and is frequently only about 30-60% for animal manure and other substrates that have a high concentration of complex molecules. (Peter Jacob 2009)

Hydrolysis is theoretically the first step of anaerobic digestion, during which the complex organic matter (polymers) is decomposed into smaller units (mono- and oligomers). During hydrolysis, polymers like carbohydrates, lipids, nucleic acids and proteins are converted into glucose, glycerol, prunes and pyridines. Hydrolytic microorganisms excrete hydrolytic enzymes, converting biopolymers into simpler and soluble compounds as it is shown below:

\[ \text{Lipids} \xrightarrow{\text{lipase}} \text{fatty acids, glycerol} \]

\[ \text{Polysaccharide} \xrightarrow{\text{Cellulase, cellobiase, xylanase, amylase}} \text{monosaccharide} \]

\[ \text{Proteins} \xrightarrow{\text{protease}} \text{amino acids} \]

A variety of microorganisms is involved in hydrolysis, which is carried out by exoenzymes, produced by those microorganisms which decompose the undissolved particulate material. The products resulted from hydrolysis are further decomposed by the microorganisms involved and used for their own metabolic processes (Teodorita Al Seadi October 2008)
2.6.2 Fermentation – acidogenesis

In a balanced bacterial process approximately 50% of the monomers (glucose, xylose, aminoacids) and long-chain fatty acids (LCFA) are broken down to acetic acid (CH3COOH). Twenty percent is converted to carbon dioxide (CO2) and hydrogen (H2), while the remaining 30% is broken down into short-chain volatile fatty acids (VFA). Fatty acids are monocarboxylic acids that are found in fats. Most naturally occurring fatty acids contain an even number of carbon atoms. VFAs have fewer than six carbon atoms. LCFAs have more than six carbon atoms. If there is an imbalance, the relative level of VFAs will increase with the risk of accumulation and the process “turning sour” because the VFA degrading bacteria have a slow growth rate and cannot keep up. A steady degradation of VFAs is therefore crucial and often a limiting factor for the biogas process. Hydrolysis of simple fats results in 1 mol glycerol and 3 mol LCFA. Larger amounts of fats in the substrate will thus result in large amounts of long-chain fatty acids, while large amounts of protein – that contain nitrogen in amino groups (-NH2) – will produce large amounts of ammonium/ammonia (NH4+/NH3). In both cases this can lead to inhibition of the subsequent decomposition phase, particularly if the composition of the biomass feedstock varies. (Peter Jacob 2009)

Table 2 Examples of fatty acids. **Short-chain (volatile) fatty acids (VFA) have less than six carbon atoms in the molecule chain. Long-chain fatty acids (LCFA) often have well in excess of six**

<table>
<thead>
<tr>
<th>Monocarboxyl acids</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Volatile fatty acids (VFA)</strong></td>
<td></td>
</tr>
<tr>
<td>Formic acid</td>
<td>HCOOH</td>
</tr>
<tr>
<td>Acetic acid</td>
<td>CH3COOH</td>
</tr>
<tr>
<td><strong>Butyric acid</strong></td>
<td>C3H7COOH</td>
</tr>
<tr>
<td>long-chain fatty acids (LCFA)</td>
<td></td>
</tr>
<tr>
<td>Lauric acid</td>
<td>C11H23COOH</td>
</tr>
<tr>
<td>Palmitic acid</td>
<td>C15H31COOH</td>
</tr>
<tr>
<td>Stearic acid</td>
<td>C17H35COOH</td>
</tr>
<tr>
<td>Oleic acid</td>
<td>C17H33COOH (one double bond)</td>
</tr>
<tr>
<td>Linoleic acid</td>
<td>C17H31COOH (two double bonds)</td>
</tr>
<tr>
<td>Linolenic acid</td>
<td>C17H29COOH (three double bonds)</td>
</tr>
</tbody>
</table>
During acidogenesis, the products of hydrolysis are converted by acidogenic (fermentative) bacteria into methanogenic substrates. Simple sugars, amino acids and fatty acids are degraded into acetate, carbon dioxide and hydrogen (70%) as well as into volatile fatty acids Volatile fatty acids (VFA) and alcohols (30%). (Teodorita Al Seadi October 2008)

2.6.3 Methanogenesis

The last step in the production of methane is undertaken by the so-called methanogenic bacteria or methanogens. The methanogens belong to a kingdom called Archaea, part of a taxonomic system that also comprises eukaryotes and bacteria at this level. A kingdom is the highest taxonomic level and Archaea are therefore at the same level as the other kingdoms – plants, animals, bacteria (Eubacteria), protozoa and fungi. Methanogens are believed to have been some of the first living organisms on Earth. Two different groups of bacteria are responsible for the methane production. One group degrades acetic acid to methane and the other produces methane from carbon dioxide and hydrogen. Under stable conditions, around 70% of the methane production comes from the degradation of acetic acid, while the remaining 30% comes from carbon dioxide and hydrogen. The two processes are finely balanced and inhibition of one will also lead to inhibition of the other. The methanogens have the slowest growth rate of the bacteria involved in the process, they also become the limiting factor for how quickly the process can proceed and how much material can be digested. The growth rate of the methanogens is only around one fifth of the acid-forming bacteria. As previously mentioned, the methanogens do not release much energy in the process. But due to the anoxic conditions, the competition from other bacteria is limited, which is why they manage to survive. (Peter Jacob 2009)
Table (3): Energy yield of methanogens from decomposition of different sources

<table>
<thead>
<tr>
<th>Source</th>
<th>Process</th>
<th>Energy yield kJ/mol methane</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hydrogen</td>
<td>$4H_2 + CO_2 \rightarrow CH_4 + 2H_2O$</td>
<td>131</td>
</tr>
<tr>
<td>Formic acid</td>
<td>$4HCOOH \rightarrow CH_4 + 3CO_2 + 2H_2O$</td>
<td>145</td>
</tr>
<tr>
<td>Methanol</td>
<td>$4CH_3OH \rightarrow 3CH_4 + CO_2 + H_2O$</td>
<td>105</td>
</tr>
<tr>
<td>Acetic acid</td>
<td>$CH_3COOH \rightarrow CH_4 + CO_2$</td>
<td>36</td>
</tr>
</tbody>
</table>

The production of methane and carbon dioxide from intermediate products is carried out by methane organic bacteria. 70% of the formed methane originates from acetate, while the remaining 30% is produced from conversion of hydrogen (H) and carbon dioxide (CO2), according to the following equations:

Acetic acid $\xrightarrow{\text{methanogenic bacteria}}$ methane + carbon dioxide

Hydrogen + carbon dioxide $\xrightarrow{\text{methanogenic bacteria}}$ methane + water

Methanogenesis is a critical step in the entire anaerobic digestion process, as it is the slowest biochemical reaction of the process. Methanogenesis is severely influenced by operation conditions. Composition of feedstock, feeding rate, temperature, and pH are examples of factors influencing the methanogenesis process. Digester overloading, temperature changes or large entry of oxygen can result in termination of methane production. (Teodorita Al Seadi October 2008)

2.7 Process parameters for a biogas plant

In order for a biogas process to be effective and productive, there are a number of parameters that have to be optimized (Peter Jacob 2009)

2.7.1 Anaerobic environment

As mentioned earlier, the methanogens need an oxygen-free environment they are obligately anaerobic. A biogas reactor therefore has to be airtight. The small amount of oxygen dissolved in the liquid/biomass fed to the plant is quickly used up by, for example, aerobic
bacteria that must have oxygen, or by facultative anaerobic bacteria that can use oxygen for their respiration, if it is present (Peter Jacob 2009)

2.7.2 Temperature
The rate of biochemical processes generally increases with temperature. As a rule of thumb, the rate is doubled for every 10-degree rise in temperature within certain limits ($Q_{10} = 2$). This is also the case with the biogas process. In this situation there are, however, several types or strains of bacteria involved that have adapted to the different temperatures:
- psychrophiles 0 – 20°C
- mesophiles 15 – 45°C
- thermophiles 40 – 65°C
Common to the bacteria is that they are very sensitive to changes in temperature. This sensitivity increases with temperature. In practice, biogas plants are run at either a mesophilic level of around 37°C, where fluctuations of approx. ± 2°C are tolerated, or at a thermophilic level of around 52°C, where fluctuations of only approx. ± 0.5°C are tolerated (Peter Jacob 2009)

2.7.3 Acidity (pH)
Despite the methanogens using organic acids for some of their food intake, they cannot cope in an acidic environment. The optimum environment is a pH of between 6.5 and 8, and the preferred level is 7.2. When the process is in balance, the acidity in the reactor will be within this range and as the buffer capacity in the reactor is very large, it takes a lot to alter it. The system is, in other words, very robust and stable. Slurry-based plants often have a somewhat higher pH (8-8.3) due to a higher ammonium content (Peter Jacob 2009)

2.7.4 Substrate (feedstock)
All organic matter can be decomposed nearly aerobically, but degree of decomposition can be increased in various ways. Lignin is, however, indigestible. (Peter Jacob 2009)

2.7.5 Comminution
The finer the material, the larger the relative surface and the easier it is for the bacteria to attack the material. (Peter Jacob 2009)

2.7.6 Dry matter content
For bacteria to be able to degrade the material, the dry matter content must not be higher than around 50%. In a biogas plant, however, it should
only be around 8-10%, if it is to remain liquid enough to be pumped. A slightly higher level can be tolerated in special reactor types with a direct feed line.

2.7.7 Carbon/nitrogen (C/N) ratio

Just like any other organism, methanogens need a number of macro- and micronutrients in order to grow. The most important macronutrients are nitrogen (N), phosphorus (P) and potassium (K). Nitrogen is used by bacteria to produce proteins. The nitrogen content is often quoted in relation to carbon, as this gives an indication of whether there is sufficient nitrogen available for bacteria. Normally the C/N ratio should be less than 30/1, as nitrogen otherwise becomes the limiting factor for bacterial growth. On the other hand, the nitrogen level should not be too high as this can then also inhibit the process.

<table>
<thead>
<tr>
<th>Essential micronutrients</th>
<th>Optimum Concentration g/m³</th>
</tr>
</thead>
<tbody>
<tr>
<td>Barium (Ba)</td>
<td>0.05</td>
</tr>
<tr>
<td>Iron (Fe)</td>
<td>0.2</td>
</tr>
<tr>
<td>Calcium (Ca)</td>
<td>0.03</td>
</tr>
<tr>
<td>Cobalt (Co)</td>
<td>0.005</td>
</tr>
<tr>
<td>Magnesium (Mg)</td>
<td>0.02</td>
</tr>
<tr>
<td>Molybdenum (Mo)</td>
<td>0.005</td>
</tr>
<tr>
<td>Nickel (Ni)</td>
<td>0.01</td>
</tr>
</tbody>
</table>

2.7.8 Organic load

The rate at which biomass is added to the reactor has to be adjusted to the growth rate of the methanogens and organic acids have to be removed at the rate at which they are produced. The normal load for a CSTR reactor is 1-6 kg COD/m³ reactor volume/day. If more biomass is added than the bacteria are able to degrade, the process will become acidic. The biomass also has to be fed to the reactor at an even rate and volume,
preferably as a continuous feed. If the substrate has to be changed, this must be done gradually, so that bacteria can adapt to the new conditions.

2.7.9 Stirrin

There are a number of different plant types, but for the most common type – CSTR (Continuously Stirred Tank Reactors) – the biomass has to be vigorously agitated to avoid the formation of an impenetrable surface crust.

2.8 Biogas production:

Many studies argued about biogas production from cattle dung and different type of organic waste and plants: Elijah T. et al (2009). Used the cow dung with rice husk for biogas production at laboratory scale was the subject of his investigation and reported the average cumulative biogas production as (161.5 ML) at the temperature 26-29°C, pH values 7.29. Yusuf et al (2011), and reported Ambient temperature of biogas production from co-digestion of horse and cow dung, the average cumulative biogas production as (254.5 ML) at the temperature 28-33°C. Similarly Okeh (2014) studied the laboratory scale biogas production from rice husks generated from different rice mills and using cow rumen fluid as a source of inoculums. Feedstock to water dilution ratio of 1:6 w/v and pH values 7.00 gave the maximum biogas yield of 382 and 357 ML/day respectively.

2.9 Inhibition of the biogas process:

Inhibition means that a substance has a negative effect on bacteria without directly killing them. The process can be inhibited in many ways and the ways are often divided into endogenous and exogenous causes. Endogenous inhibition is due to conditions or material created during the process itself that under certain circumstances may inhibit the process, and exogenous inhibition is due to external conditions (Peter Jacob 2009).

2.9.1 Nitrogen inhibition

One of the most significant endogenous inhibitors is ammonia (NH3). Ammonia is created during the bacterial degradation of nitrogen-containing substances such as proteins. Nitrogen is essential for bacterial growth and ammonia is an important source of nitrogen. But ammonia at high concentrations is highly toxic to the bacteria. In an aqueous solution ammonia is always found in an equilibrium with ammonium (NH4+). This equilibrium is determined by the acidity, pH and temperature of the environment and, as ammonium is not as toxic as ammonia, this equilibrium is important (Peter Jacob 2009):

\[ \text{NH}_4^+ \rightleftharpoons \text{NH}_3 + \text{H}^+ \]
2.9.2 Antibiotics, etc.

Among the exogenous causes, antibiotics and disinfection agents are obvious inhibitors of the process, because both – by definition – are toxic to and are used to kill microorganisms. Both substances are used in livestock production to treat sick animals and to keep animal houses and milking parlous clean and can therefore also be found in the slurry, but apparently only at concentrations so low that they do not have a negative impact on the biogas plant. A slow adaptation to these substances can also take place if the supply is fed in continuously. Other substances such as heavy metals, salts and micronutrients can also inhibit the process at high concentrations \((5)\). But as previously mentioned, some of them are essential for the process at low concentrations, in the same way that vitamins are for humans\((\text{Peter Jacob 2009})\)

Table(5): Selected inhibitors with values at which they are inhibiting and toxic.

<table>
<thead>
<tr>
<th>Chemical/formula</th>
<th>Inhibition level</th>
<th>Toxicity level</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ammonia, free, NH3</td>
<td>50-100 mg N/l</td>
<td>100-200 mg N/l</td>
</tr>
<tr>
<td>Ammonia, total, NH4++NH3</td>
<td>1,000-6,000 mg N/l</td>
<td>10,000 mg N/l (pH&lt;7.5)</td>
</tr>
<tr>
<td>Chloride, Cl-</td>
<td>&lt; 8,000 mg/l</td>
<td>10,000 mg/l</td>
</tr>
<tr>
<td>Cyanide, CN-</td>
<td>2-20 mg/l</td>
<td>30 mg/l</td>
</tr>
<tr>
<td>Formaldehyde, H2CO</td>
<td>100-400 mg/l</td>
<td>500-1,000 mg/l</td>
</tr>
<tr>
<td>Phenol, C5H5OH</td>
<td>100-200 mg/l</td>
<td></td>
</tr>
<tr>
<td>Chloroform, CHCl3</td>
<td>&gt;1 mg/l (single dose)</td>
<td>&gt;50 mg/l (continuous feed)</td>
</tr>
<tr>
<td>Hydrogen,H2</td>
<td>p(H2) ca 10-4 atm.</td>
<td></td>
</tr>
<tr>
<td>Copper, Cu+++</td>
<td>10-250 mg/l</td>
<td></td>
</tr>
<tr>
<td>Chrome, Cr+++</td>
<td>50-100 mg/l</td>
<td>200-400 mg/l</td>
</tr>
<tr>
<td>Nickel, Ni++</td>
<td>100-200 mg/l</td>
<td>300-1,000 mg/l</td>
</tr>
<tr>
<td>Sodium, Na+</td>
<td>3,000-10,000 mg/l</td>
<td></td>
</tr>
<tr>
<td>Calcium, Ca++</td>
<td>8,000 mg/l</td>
<td></td>
</tr>
<tr>
<td>Magnesium, Mg++</td>
<td>3,000 mg/l</td>
<td></td>
</tr>
<tr>
<td>Zink, Zn+</td>
<td>350-1,000 mg/l</td>
<td></td>
</tr>
<tr>
<td>Sulphate, SO4 - -</td>
<td>500-4,000 mg/l</td>
<td></td>
</tr>
<tr>
<td>Sulphide, (as sulphur)</td>
<td>200 mg/l</td>
<td></td>
</tr>
<tr>
<td>Hydrogen sulphide, H2S</td>
<td>250-1,000 mg/l</td>
<td></td>
</tr>
</tbody>
</table>
2.9.3 Acidification – organic acids

Other important endogenous process inhibitors are the organic acids formed during the process. If these are not removed as soon as they are formed – which can happen during an overload – this can lead to an acidification of the process. (Peter Jacob  2009)
Chapter Three
Materials and method:

3.1 Design method:

The study was conducted at plant laboratory at(22-1-2017) in college of Animal production science and technology to determine the amount of biogas production by fermentation of a mixture consisting of Cow dung, meat by- product, and molasses

3.2 Sample collection:

The cow dung was obtained from the Animal Production Research Center, about 6 kg of manure, the meat by- product was purchased from the slaughterhouse and was about 250 gm added with 250ML of Molasses.

3.3 Materials/instruments:

The instrument used was composed of 25ltr Capacity Iron Drum – Biogas digestion Tank, 1000ml cylinder To measure the amount of biogas Collection Unit, Control Valve, ½” CPVC Pipe_ Water output pipe , 1”CPVC Pipe Gas Collection, Container _To collect water flowing from the cylinder.

3.4 Apparatus set-up:

The apparatus was properly washed with soap solution and allowed to dry by standing over night in the laboratory. A Drum containing an open hole in the top to insert the mixture, the opening by a control valve connector Bonbon connects the gas from the Drum to the cylinder to measure the amount of gas by displacing the water in the cylinder, which is tightly sealed from the top and there are two openings one to insert the tube coming from the digestion that pumps gas and open it Another connector with a tube to remove the water joking by biogas

3.5 Parameters of biogas production and their selected operating Conditions

The research was carried out under room temperature that varied between 20 and 25°C, pHvalues7.00
3.6 Apparatus and the digester used for biogas production
Chapter Four

Results:

At the beginning of the experiment no biogas was produced. The production started at the 81th day and continued until the 90th day.

4.1 Table(6): Daily and cumulative biogas production with time

<table>
<thead>
<tr>
<th>Days</th>
<th>Daily production(ml)</th>
<th>Cumulative production(ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1----80</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>81</td>
<td>40</td>
<td>40</td>
</tr>
<tr>
<td>82----85</td>
<td>0</td>
<td>40</td>
</tr>
<tr>
<td>86</td>
<td>90</td>
<td>130</td>
</tr>
<tr>
<td>87</td>
<td>40</td>
<td>170</td>
</tr>
<tr>
<td>88----90</td>
<td>30</td>
<td>200</td>
</tr>
</tbody>
</table>

4.2 Fig(1): Biogas production with time presentation curves
CHAPTER FIVE
DISCUSSION

The results of this study will be discussed in this chapter:

5.1 Temperature:

The temperature in this study was between 20 and 25°C, this result disagreed with that reported by Elijah T. et al. (2009) as (26-29°C) and Yusuf et al. (2011) as (28-33°C). The disagreement of the result of this study with other researches may be due to lower temperature during the experiment.

5.2 pH values:

The pH values in this study was between (7.00-6.4) this result agreed with that reported by Okeh (2014) as (7.00) and (Elijah T. et al. 2009) as (7.29).

5.3 Amount of biogas production:

The amount of biogas production in this study was 200 ML which disagreed with that reported by Elijah T. et al. (2009) as (161.5) ML and Okeh (2014) as (382 ML). The differences in biogas production in this study may be due to lower temperature during the experiment.
CONCLUSION AND RECOMMENDATION

CONCLUSION:

The study was concluded that biogas can be produced from fermentation of plant and animal waste

RECOMMENDATION:

We recommend for the continuation of studies for biogas production using different animals and plant by-products
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