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ANAEROBIC FERMENTATION OF MEAT PROCESSING WASTES AT 40°C AND 50°C AS A SOURCE OF ENERGY

by

© TOMAS J. SCHMIDT

A THESIS

SUBMITTED TO THE FACULTY OF GRADUATE STUDIES AND RESEARCH IN

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(Supervisor)

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Date October 12th, 1982

ABSTRACT

Meat processing wastes were subjected to anaerobic digestion at 40°C and 50°C. An effective system was developed at both temperatures resulting in final Chemical Oxygen Demand (COD) reduction of at least 90% at 4 day Solids Retention Time (SRT) for both temperatures. Further increase in SRT to 2 days resulted in 73% and 81% COD reduction respectively. However, in this instance, the effectiveness of the operation at 40°C was subject to great variation.

Only about 57 and 54% reduction in total volatile solids was achieved at 4 days SRT at 40°C and 50°C respectively. Even at 20 days SRT, the volatile solids reduction increased to only 62% indicating the presence of non-degradable portion, probably in the form of inorganic salts.

The greatest difference between the two treatment temperatures was observed in the efficiency of lipid utilization. At 4 days SRT and 40°C, only about 31% of total lipid removal was achieved, compared to 52% at 50°C. To obtain at least 50% reduction in total lipid content at 40°C, a 20 days SRT period was required; 86% of total lipid was utilized at 50°C under the same conditions.

Maximum gas production was observed at 4-8 days SRT period, for both temperatures. Total methane gas production at 50°C and 4 day SRT, 1023 ml/day, was 36% higher as compared to 750 ml/day production at 40°C and 4 day SRT. When organic load for both digesters was increased

to the equivalent of 2 day SRT, methane gas production decreased by 67% at 40°C, and by 55% at 50°C.

Methane content of the digester gas at 50°C remained relatively constant for all loading rates, ranging from 62.6 to 67.3%. However, at 40°C, a significant reduction from 66.5 at 4 day SRT to 48.6% at 2 day SRT was observed.

At both temperatures investigated, the gas production decreased at SRT greater than 8 days, indicating the substrate availability to be the rate limiting factor. At 2 days SRT, the reduction in treatment efficiency, combined with the increase in total volatile acids content indicates that the acid utilization by methanogenic organisms was the rate limiting step.

An economic analysis of the unit processes for a potential large scale application indicated that maximum benefit can be obtained at 4 days SRT and 40°C. Although the gas production at 50°C was significantly higher, it would not be sufficient to compensate for the supplemental heat required to maintain the digester at 50°C.

The cost of chemicals (484 $/day) accounted for a major portion of the total cost at 40°C followed by the operating and maintenance cost (150 $/day) and the capital cost.

The economics of a Multi-Stage Flash (MSF) evaporation principle was examined in an attempt to evaluate the feasibility of decreasing the total waste flow and thus reducing the initial capital cost required. A concentration factor of 10 was selected for this study.

Total MSF cost of the evaporation/anaerobic digestion system at 50°C was approximately 50% higher than that of the simple anaerobic treatment at 40°C (2022 $/day vs 1059 $/day) respectively. Thus, the

MSF evaporation concept does not appear to be economically feasible in the context of this study. The major contributing factor was the cost of steam required to achieve the desired concentration.

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The author would like to express his sincerest appreciation to Dr. P. Jelen for his assistance during the preparation of this thesis.

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CHAPTER 1

INTRODUCTION

1.1 Industrial Waste Problem

Food processing industry is one of the most significant sources of industrial wastes and waste waters. Processing of raw agricultural materials into food products generates wastes which are characterized by high volume and/or high organic strength. The amounts of wastes associated with production of certain food products (e.g. cheese) often exceed several-fold the amount of the finished product.

Most of the food processing plants are located at, or near, large communities and in many instances, a provision is made for their waste to be treated by the municipal sewage treatment plant. With the increase in production level, there is a corresponding increase in the volume (not necessarily the strength) of discharged liquid. In many instances this results in overloading of the working capacity of a municipal treatment plant. Sometimes, toxic materials might be present in discharged liquid, creating operational difficultie as well as a health hazard.

Because of the increased cost of waste treatment, most municipalities now require that the organic strength of discharged liquid be equivalent to that obtained from municipal secondary treatment. In order to achieve this level, generally at least 85% of BOD must be removed, in combination with sufficient reduction in sedimentable suspended solids.

Although industrial wastes can differ in their composition, their characteristics and organic strength are described in common

terms, such as biochemical oxygen demand (BOD), chemical oxygen demand (COD), weight of suspended solids, pH, etc.

Main problems associated with the treatment of the food processing wastes are their large volumes, and low organic strength. Because of their volume, relatively large treatment installations are required, resulting in high capital investment. Reduction in the size of the treatment plant could be achieved by increasing the rate of waste stabilization, e.g. by increase of the treatment temperature. Majority of the anaerobic treatment plants operate at 25-30^{o} temperature range. It has been documented that the increase in digester temperature to 35-37^{o}C resulted in significant increase in waste utilization.

There has been a limited amount of research indicating that effective anaerobic treatment can be achieved with strong organic wastes at a temperature range from 35 to 60^{o}C. The rate of methane gas formation was sufficient to maintain the digester temperature. However, there is no information available concerning the efficiency and economics of treatment of dilute food processing wastes at temperatures at or above 40^{o}C.

Dilute wastes may not be considered to be suitable for economical methane production. Because of this uncertainty and/or lack of data, no special attempts have been made to evaluate the technical aspects of methane gas production in the overall economics of the anaerobic treatment of dilute wastes. Furthermore, most of the attention was devoted to the actual rate of waste stabilization, while no attempts were made to stimulate methane gas production by increase in temperature, by addition of nutrients, etc.

1.2 Research Objectives

This study was concerned with the anaerobic digestion of dilute food processing wastes as a possible alternative to currently used waste treatment techniques. The effectiveness of the waste stabilization was studied in relation to two treatment temperatures and the rate of production of methane gas, an economically valuable product of the proposed treatment.

The specific objectives of this study were:

(i) To evaluate the degradation of dilute complex organic wastes (typified by liquid wastes from a meat processing operation) under anaerobic conditions as a function of temperature at 40°C and 50°C;

(ii) to evaluate the effect of hydraulic loading rates on the degree of degradation of these complex organic wastes at 40°C and 50°C;

(iii) to establish and evaluate conditions for maximum methane gas production at 40°C and 50°C;

(iv) to develop and evaluate an economical model for anaerobic treatment of complex organic waste at 40°C and 50°C.

CHAPTER 2

LITERATURE SURVEY

2.1 Characteristics of Food Processing Wastes

Depending upon the type of the food processing plant, the characteristics of its waste exhibit extreme variations. The BOD level might be as low as 50 - 100 mg/l or as high as 100 000 mg/l. Suspended solids may range from almost 0 to 110 000 mg/l. Some wastes such as found in vegetable processing, are highly acidic (pH 3.5) while others - e.g. those originating from potato processing - are highly alkaline (pH 11.0). The mineral nutrients necessary for the microorganisms are lacking in some types while excessive levels of toxicity are present in other wastes. Thus, for an optimum design of the most suitable method of food processing waste treatment, all these factors must be carefully considered.

Table 1 summarizes major food processing wastes, their origin, characteristics and a typical treatment used.

2.2 Waste Treatment Methods

Methods available for the treatment of almost any kind of waste can be broadly categorized as physical and biological. The latter can be further divided into two separate treatment groups, aerobic and anaerobic, depending respectively on the need for presence or absence of oxygen for efficient treatment. Obviously, no one treatment would completely satisfy all the needs when dealing with complex processing wastes. Almost all treatment systems are comprised of the combination of different physical and biological treatment stages. Whatever system is being used, its prime objectives remain the same, namely to decrease

Table 1: Food Processing Wastes, Origin, Character and Treatment (after Nemerow, 1971)

Industry	Origin of Waste	Major Characteristics	Typical Treatment
Fermentation Industry	Steeping and pressing of grain; residue from alcohol distillation.	High in dissolved organic solids and BOD; contain nitrogen and fermented starches or their products.	Recovery, concentration by evaporation or centrifugation; trickling filters, digestion of slops.
Dairy Industry	Dilutions of whole milk, separated milk, buttermilk, and whey.	High in dissolved organic matter, mainly protein, fat and lactose.	Biological Treatment, aeration, trickling filtration, activated sludge.
Meat and Poultry Industry	Stockyards, slaughtering of animals, rendering of bones and fats; residues in condensates; grease and wash water; picking of chickens.	High in dissolved and suspended organic matter, blood, other proteins, and fats.	Screening, settling, and/or flotation, trickling filtration, anaerobic digestion.
Canning Industry	Trimming, cutting, juicing and blanching of fruits and vegetables.	High in suspended solids, colloidal and dissolved organic matter.	Screening, lagooning, spray irrigation.
Potato Industry	Peeling cutting, blanching.	High in suspended solids and dissolved carbohydrates.	Screening, spray irrigation, aerobic biological treatment.

the initial organic strength of the waste to an acceptable level. The following overview of the available methods for waste treatment has been summarized from several textbook references (Eckenfelder and O'Connor, 1961, Nemerow, 1971, Fair et al., 1968).

2.2.1 Physical Methods of Waste Treatment

When dealing with food processing wastes, a physical treatment, such as screening or flocculation, is widely used as a pretreatment. Filtration or sedimentation may be used as a final "polish" treatment after some kind of biological stabilization before wastes discharge into a body of water. Several types of physical treatments are common.

(a) Screening is used to remove coarse particles, It is widely used in fruit, vegetable and potato processing plants.

(b) Flocculation refers to the aggregation of small particles into larger ones. Depending on the waste characteristics, chemicals might be added to induce aggregation and settling of fine suspended matter and colloidal substances. Ferrous oxide, due to its ability to flocculate fats, is widely used in meat processing waste treatment.

(c) Floatation method of removal of finely divided suspended solids and particles is characterized by the injection of fine gas bubbles into the liquid phase. As the bubbles attach themselves to the particulate matter, the aggregate increases in size and the buyoant forces acting on the combined particles and gas bubbles cause the particle to rise to the surface. This method is applied mainly for the removal of suspended matter and for concentration of biological sludges.

(d) Sedimentation is a removal of suspended particles by gravity settling. It also has an application for grit removal, particulate

matter in a primary settling basin and biological floc removal in an activated sludge basin.

(e) Filtration is mostly used for a final removal of suspended solids remaining after the biological or chemical treatment. The process consists of "sand and gravel" or fine silica sand filters. Its greatest use is for the treatment of municipal water and industrial process water. When combined with the biological treatment such as a trickling filter, it can provide two treatments in a single pass, e.g. biological stabilization and suspended solids removal.

2.2.2 Aerobic Waste Treatment

The oxygen demand in discharged industrial wastes is basically exerted by three different classes of materials (Annon., 1969):

(i) Carbonaceous organic matter that is used as a food substrate by microorganisms.

(ii) Oxidizable nitrogen compounds derived from ammonia, nitrite and organic nitrogen compounds. It serves as a nitrogen source for different types of microorganisms.

(iii) Chemical reducing compounds such as sulfite, sulfide and ferrous iron.

In the aerobic treatment, oxygen is supplied to the substrate by mechanical means such as circulating pumps and injection of compressed air to support microbial metabolism and growth, thus resulting in conversion of the various waste materials to biomass. The aerobic digestion treatment can be carried out in several typical facilities.

(a) Aerated lagoons are mixed reactors that are operating on a flow-through basis and do not employ cell recycling. They consist

of a large, shallow earthen basins mixed and aerated by surface aerators. The removal and/or stabilization of the waste is achieved through conversion of the complex waste to bacterial mass and CO_2. This type of treatment must be followed by sedimentation or other means of separating microbial mass from the discharged effluent.

(b) Activated sludge treatment employs the use of mechanical aerators and/or compressed air or oxygen to increase the efficiency of the process. In contrast to the aerated lagoon, after a specific period of time a portion of the liquid is withdrawn to a separate tank and allowed to settle. Part of the settled sludge is later returned to the reactor to maintain relatively stable microflora.

(c) Aerobic digestion applies to aerobic destruction of insoluble organic wastes in a slurry reactor. It is used to remove excess activated sludge formed during the treatment of soluble wastes. In this process, the sludge is aerated for an extended period of time in an open tank using conventional air diffusers or surface aerators. The advantage of this process is that it could be either batch or continuous operation, but in the latter case, it must be followed by a separate settling tank.

(d) Trickling filters consist of a bed of coarse permeable media such as rock or plastic balls. While the waste effluent flows through the filter, it comes into contact with microflora attached to the media. The organic material is absorbed and utilized by these organisms. As the thickness of slime containing the microbial flora increases during extended filter operation, the oxygen is more rapidly utilized, creating anaerobic conditions near the bottom of the filter. This type

of waste treatment provides a large surface area for the microbial growth and is very stable to organic and hydraulic shock loading. At times, it is necessary to backwash the filter in order to remove some of the excessive microbial mass build up.

(e) A rotating disc contactor is based on the same principle as the trickling filter. A massive microbial buildup is developed and attached to a matrix which comes into contact with the effluent. The matrix consists of parallel circular discs attached perpendicularly to a horizontal shaft which passes through their center. The discs are usually constructed from high density polyethylene. During the operation, these discs are approximately half immersed and rotate at a pre-determined speed. The rotation acts as a mixing device, and it provides constant shear force causing continual sloughing of the stabilized organic matter. These discs also function as aerators. As they rise from the solution, a thin liquid layer remains on the disc surface absorbing the oxygen as it passes through the air. Reimmersion returns this highly aerobic liquid into the reactor thus increasing the oxygen content.

2.2.3 Anaerobic Waste Treatment

The anaerobic treatment is described as a microbial stabilization of a complex organic waste in the absence of oxygen. This process can be divided into two sequential stages, each being carried out by a different group of organisms specific to the particular stage. The general anaerobic waste degradation flow diagram is presented in Figure 1. In the first stage of the process, the complex organic matter is hydrolyzed by the non-methanogenic organisms into a variety of organic

acids utilized by methane producing organisms in the methanogenic stage, with the end product being CH_4 and CO_2.

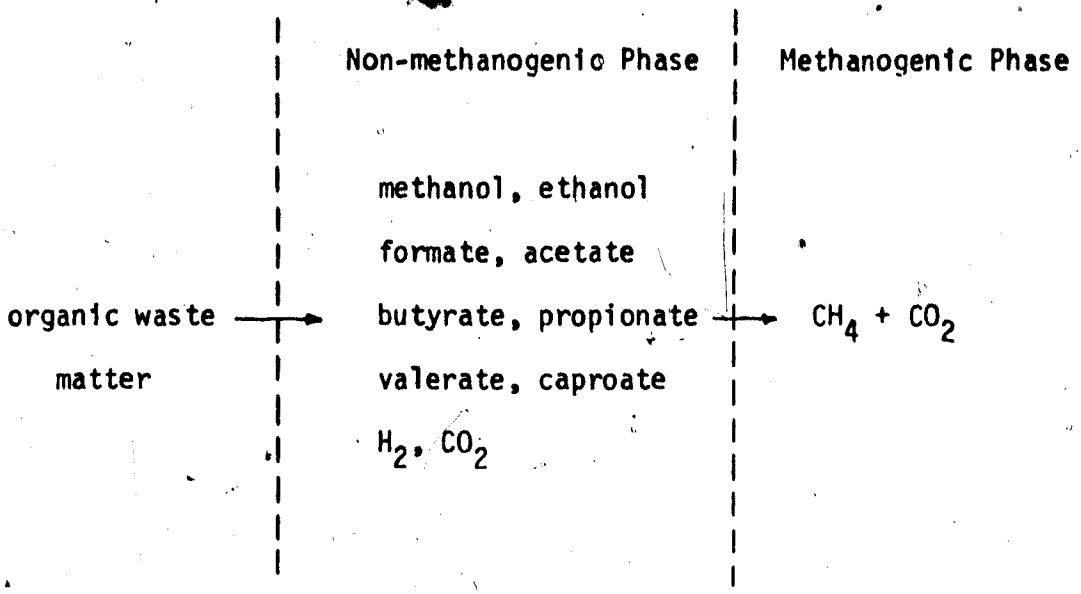


Figure 1. Schematic of anaerobic digestion.

The anaerobic waste treatment methods can be classified into three broad categories - anaerobic lagoons, septic tanks and anaerobic digesters.

(a) An anaerobic lagoon consists of a large earthen pond open to the atmosphere. Since the anaerobic process requires the absence of oxygen, it takes place beneath the surface, while aerobic treatment takes place in the top layer. The operating

temperatures are generally low, resulting in a long retention time required to achieve sufficient breakdown.

(b) A septic tank is an enclosed anaerobic system where settling solids are held sufficiently long to undergo partial or complete digestion. The gases produced are released to the atmosphere, while the stabilized waste is periodically discharged. Similarly, as for the anaerobic lagoons, the temperatures of the treatment depend on ambient conditions resulting in long retention periods.

(c) Anaerobic digestion refers to the process operated above ambient temperatures. The gas formed in the process of microbial waste stabilization can be collected and used for heating purposes. The increase in operating temperatures increases the rate of biological stabilization resulting in decreased retention time. The treatment unit can be considerably smaller in size as compared to lagoons.

Newer developments, such as an anaerobic contact process, greatly increased the feasibility for treatment of dilute industrial wastes. In principle, this process is analogous to the activated sludge treatment in that biological solids present in the final effluent are concentrated by sedimentation and portion is returned to the reactor. By maintaining a high concentration of microorganisms in the reactor, rapid and efficient treatment can be achieved.

The anaerobic filter designed by Young and McCarty (1967) is similar in appearance to an aerobic trickling filter. The incoming waste is distributed across the bottom and the flow is in an upward direction. This ensures that the filter is completely submerged without void spaces. Anaerobic organisms cling to rock surfaces

and the waste comes in contact with an active biological mass as it passes through the filter.

The anaerobic filter is of simple design and requires no sludge or effluent recycle to maintain a high treatment efficiency. The production of solids is low and with protein or fatty acid substrates the filter can be operated for well over a year with no need for solids wasting (Young and McCarty, 1967). When high carbohydrate wastes are being treated, sludge wasting or backwash must be done periodically due to excessive biological solids accumulation. Another advantage of this type of treatment is its rapid response to change in incoming waste characteristics, thus having application for food processing plants operated on a periodic or seasonal peak basis.

Schematic diagrams of the anaerobic contact process and the anaerobic filter are illustrated in Figure 2.

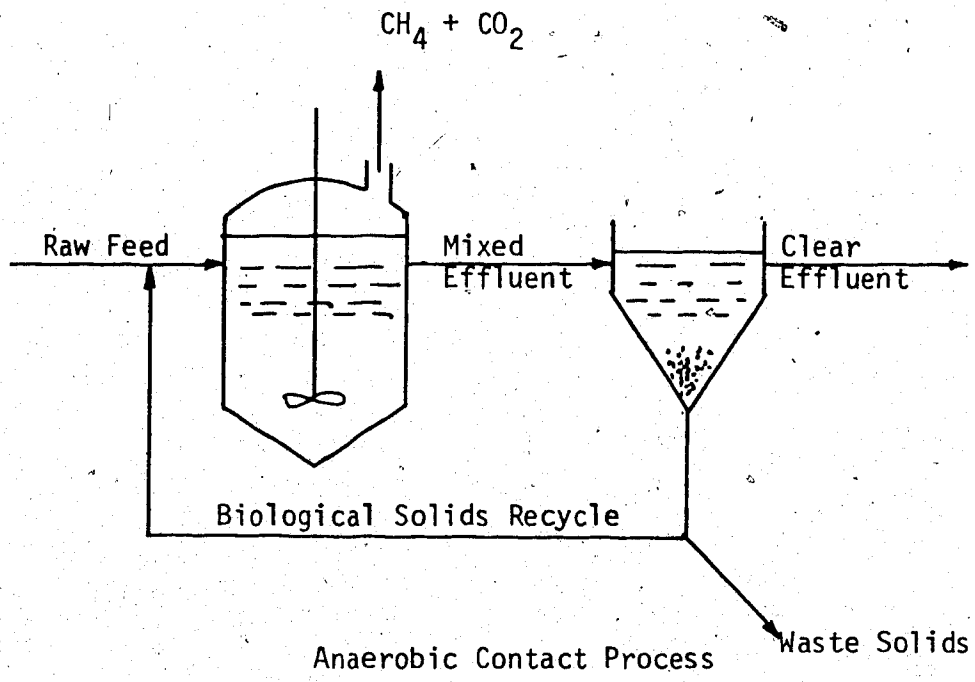
2.3 Anaerobic Treatment of Food Processing Wastes

Initially, the anaerobic digestion in form of septic tanks has been used to treat domestic wastes. As a result of new, more efficient treatment methods, it has been gaining importance for industrial applications. The following survey of waste treatment methods in various branches of the food industry indicates that the anaerobic digestion has been studied, recommended and/or applied to a variety of liquid waste materials.

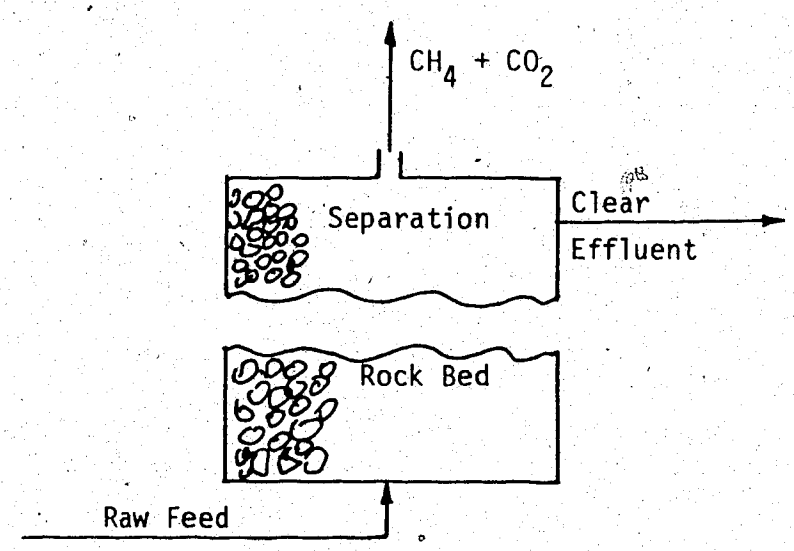
2.3.1 Fermentation Industry

Pearson et al. (1955) applied the anaerobic treatment to winery wastes. With operating temperatures of 35.5 - 37.7^{o}C and loading rates of 1.6 - 3.2 kg of volatile solids (VS)/m^{3}/day, 87 - 88% reduction of VS

Figure 2: Schematic Diagram of the Anaerobic Contact Process and the Anaerobic Filter System



Anaerobic Contact Process



Anaerobic Filter System

and BOD was achieved. The gas produced contained on an average 62% methane. Stander (1967) described the anaerobic treatment of wine distillery wastes. He achieved the final COD reduction of 97% using the maximum organic load of 3.2 kg COD/m^3/day at 30°C, and retention time of 7.2days. The amount of gas produced was sufficient to maintain the desired operating temperature up to 45°C. The estimated treatment cost amounted to approximately to 12.3 ¢/m^3 of raw sewage.

Newton et al. (1961) studied the anaerobic treatment of brewery wastes. Their results showed an average 96% BOD reduction at a loading rate of 1.58 kg BOD/m^3/day. According to these workers, loading up to 2.88 kg BOD/m^3/day could be handled with a removal efficiency of 92 to 97%. No data were obtained on quantities of gas produced but the assumption was that considerable supplementary fuel would be required for digester heating. Probably the major problem in the anaerobic treatment of brewery wastes is their extreme variation in composition. O'Rourke and Tomlinson (1962) demonstrated this point while studying the waste composition of a large brewery. The concentration of biological degradable organic matter varied from 24 to 4820 mg/l in hourly composite samples, while COD varied from 128 to 8420 mg/l. The pH range was between 3.64 and 7.1 during a 24-hour period. Generally, extreme peaks in BOD and COD corresponded closely to brew time. Despite all these variations, the biological system studied was 99% efficient in the utilization of the soluble biologically degradable organic materials in the wastes.

Lowan and Foree (1971) examined application of the anaerobic filter treatment for brewery press liquor waste. The greatest efficiency of COD removal (90%) was accomplished in the bottom 6

inches of the filter at a loading rate of 1.6 kg COD/m^3/day at 35^oC. The efficiency of COD removal did not significantly increase in the next 4 feet of the filter. This type of filter could be operated for long periods of time without sludge wasting, however, periodical backwash had to be applied in order to avoid serious plugging problems.

Stander (1967) operated an anaerobic treatment plant for wine distillery wastes at a maximum organic load of 3.2 kg COD/m^3/day at 30^oC, representing 7.2 days retention period. During the steady state operation period, gas production reached about 14 - 15 litres/liter of feed. It was observed that gas production declined rapidly after a short interruption in the feed supply and virtually ceased after two to four hours interruption. In addition, when the effect of the reactor temperature was examined, it was found that the increase in temperature by 10^oC in the range 15 - 30^oC approximately doubled the biological activity and thus doubled the permissive load rate. Increase to 45^oC resulted in a sharp decline of biological activity and extended acclimatization was required. The maximum loading rate was lower than that at 35^oC indicating inefficiency of utilization.

Sen and Bhaskaran (1962) studied the anaerobic digestion of liquid wastes of molasses distillation process. They reported 90% BOD reduction with loading rate of 3.0 kg BOD/m^3/day operating at 37^oC. The increase of the loading rate to 3.7 kg BOD/m^3/day resulted in incomplete digestion followed by the digester failure. The recirculation of CO_2 resulted in further reduction of BOD and in an increased percentage of CH_4 in the gas phase.

A comparative study by Basu and LeClerc (1973) on the treatment

of wastes from a beet molasses distillery indicated that there was no distinct advantage or justification in increasing the digester temperature from 35 to 55°C. BOD removal was reported to be only slightly higher at the thermophilic temperature range at organic loading of 2.0 kg BOD/m^3/day. At both temperatures, rapid decrease in treatment efficiencies occured when the loading rate was increased to 3.52 kg BOD/m^3/day. The thermophilic digester exhibited more variation in day to day BOD reduction and susceptibility to variation in pH.

The anaerobic filter system was applied for the treatment of pharmaceutical wastes (Sachs et al. 1978). The system was operated at 35°C and 36 hours retention time. With influent concentration of 2.0 kg COD/m^3/day, COD removal ranged from 70 to 80%, and BOD removal was about 94%. Gas production averaged about 0.5 l/hour. When compared to an existing aerobic treatment plant, the anaerobic filter was found to be almost 33% more efficient in COD utilization removing 1.34 kg COD/m^3/day, as compared to 0.90 kg COD/m^3/day while shortening the retention time by a factor of five (8 days versus 36 hours for anaerobic filter). The increase in loading rate to 6.0 kg COD/m^3/day resulted in incomplete COD reduction.

In contrast, Jennett and Dennis (1975) reported using loading rates ranging from 0.22 to 3.52 kg COD/m^3/day for the anaerobic filter treatment of pharmaceutical waste at 35°C. At retention times ranging from 12 to 48 hours, in all cases at least 93.7% COD reduction was achieved. The reactor was able to operate over a six month period without the need for solids disposal. Shock loads in organic loading

were reported to have no significant influence on treatment efficiency.

Effluent from production of bakers yeast was subjected to anaerobic treatment at 30°C (Hansford and Richter, 1975). With effluent strength of 16.0 kg COD/m^3/day, 60 to 70% COD reduction was achieved at hydraulic retention times varying from 5 to 10 days. Reduction in the volatile content of dissolved solids followed closely the COD reduction, both averaging about 62%. Relatively low reduction, 40 - 50%, was reported for total dissolved solids as the waste contained a high portion of soluble organic salts. Gas production was found to be susceptible to changes in loading rates. Increase in organic loading rate to 21.0 kg COD/m^3/day resulted in fermenter failure.

2.3.2 Slaughterhouse and Meat Processing Wastes

The anaerobic digestion is especially suited to meat packing wastes, as they contain appreciable amounts of fats, proteins, and high concentration of other nutrients, all of which are essential for good biological treatment.

Fullen (1953) described the operation of an anaerobic digester at the Wilson & Co. packing plant at Albert Lee, Minnesota, built after pilot plant experimental studies. With retention time of 24 to 30 hours, more than 90% BOD was removed at loading rates ranging from 0.98 to 1.30 kg BOD/m^3/day. The temperature of the digester averaged 35°C throughout the study. To achieve better separation of solids, degassification was employed.

The treatment at Geo. A. Hormel & Co. at Austin, Minnesota, (Schroepfer et al., 1955, Steffen and Bedker, 1961) averaged 96% BOD

removal. Although the loading rates were significantly higher - 1.6 to 3.2 kg BOD/m^3/day, the retention period was only 3 - 4 hours on total flow and 12 - 15 hours on the raw waste flow. The digester gas produced contained approximately 85% CH_4. Recycling of solids from the settling tank increased the efficiency of treatment to a point where the full load from the plant, without bypassing the most dilute portions, was successfully treated by anaerobic digestion.

The waste treatment operation at the Wilson & Co. plant at Cherokee, Iowa, employed both anaerobic and aerobic treatment (Hester and McClurg, 1970). Over the period of four years, almost 80% BOD removal was averaged in an anaerobic cell at 7-8 days retention period. The aerobic treatment was used as a "polishing" step to remove further BOD. The overall treatment was reported to be 92-95% efficient.

Baker and White (1971) described combined anaerobic lagoons and trickling filters to treat the waste from a meat packing plant at Denison, Iowa. The raw waste was first pre-treated by air floatation to remove grease before entering anaerobic lagoons. Over the period of two years, average BOD removal was 82% at 0.47 kg BOD/m^3/day loading rate and a retention time of 5 days. The operating temperature within the lagoon averaged 20.5°C. Further BOD removal was achieved by treating the discharged effluent in trickling filters. The overall efficiency of the treatment exceeded 95% BOD removal.

Anaerobic lagoons were also used to treat waste water in Union City, Tennessee (Saucier, 1969). With a load of 0.98 kg BOD/m^3/day, the BOD removal averaged 86%. The cost of the treatment was estimated at 15.4$/kg BOD removed.

The treatment at Parity Packing Co. in Knox City, Tennessee, was reported to remove in excess of 99% of BOD (Saucier, 1969). However, the cost of the treatment was estimated to be as high as 280$/kg of BOD removed.

Sollo (1960) described the operation of an anaerobic pond for treatment of meat wastes. On the average, BOD reduction in excess of 85% in 24°C was reported. In another study (Coerver, 1964), the same type of treatment was 92-94% efficient in BOD removal. The packinghouse wastes, which included blood and pouch manure, were successfully treated in low cost anaerobic ponds without a significant nuisance or health hazard.

Cowie (1960) described the operation of an anaerobic digester for treatment of meat processing wastes in New Zealand. Over the period of two years, BOD removal averaged more than 90% at a retention time of 14 days, and a temperature of 17.7°C. The effluent was further treated by sedimentation.

Schroepfer and Ziemke (1959) examined the effect of temperature, loading rate, solids concentration and mixing on the treatment of different wastes on a small laboratory scale. Using meat packing wastes as one of the substrates, loading rates up to 4.0 kg BOD/m^3day resulted in better than 90% BOD removal when operated at 35°C. When the operating temperature was decreased to 25°C, the maximum loading rate had to be lowered to 1.44 kg BOD/m^3/day in order to achieve 90% or more BOD reduction. A marked reduction in the degree of BOD removal was reported when mixing was not provided. The gas produced contained on the average 90% CH_4.

2.3. 3 Dairy Industry

Most dairy plant wastes respond quite well to biological treatments. The composition of these wastes is somewhat similar to that of domestic sewage, but in effect, much more concentrated. For this reason, at least two successive treatment stages are required in order for discharged effluent to meet municipal and governmental standards.

Dairy wastes are usually treated by biological oxidation methods. Most common are activated sludge, trickling filter, aerated lagoons and a combination of these. The efficiency of these treatments is generally in excess of 90%.

The use of anaerobic process to stabilize the dairy processing wastes appears to be limited, even though it has been practised for many years in small dairy operations by means of septic tanks. On a large scale, anaerobic process has not been successful as a complete treatment.The final effluent is of poor quality and does not meet stream discharge standards. However, when combined with other treatments, anaerobic digestion offers an efficient, low cost pre-treatment process.

Parker (1971) examined the possibility of methane production from whey. On a pilot scale, BOD reduction of 99% was achieved with the retention time of 6-7 days and a temperature of 35^{o}C. The gas production averaged 31 m^3/m^3 of waste.

A two stage whey treatment process was designed by Holder et al. (1978). The whey was readily stabilized by anaerobic digestion followed by an aerobic stage. An initial load of 60 kg COD/m^3/day was reduced in the anaerobic stage to about 1.0 kg COD/m^3/day and further reduced to 0.1 kg COD/m^3/day by the aerobic treatment. The gas production was

30 m^3/m^3 of raw waste with a CH_4 average content of 45%. COD reduction of 98.5% was achieved at loading rates up to 13.0 kg COD/m^3/day. A further increase to 16.5 kg COD/m^3/day resulted in digester failure.

Cleaning liquids and wastes from milk trucks are highly alkaline and contain detergent formulations and small quantities of milk. When subjected to anaerobic digestion at 31°C (Aulenbach and Hallock, 1972) 71% of COD was removed at pH 7.1 and a 7 day retention time. No gas production was observed under these conditions. Interestingly, when the pH of the digester was adjusted to 8.1, an increase in COD reduction to 77% was observed coupled with the gas production of 0.38 m^3/kg COD removed.

2.3.4 Canning Industry

The efficiency of the anaerobic process for treatment of wastes from the canning industry exhibits a high degree of variation among the reported studies.

Canham (1949) described the operation of a pilot plant at Ladoga, Indiana. The average BOD reduction was 39.8% at a loading rate of 0.62 kg BOD/m^3/day and a temperature of 15 to 24°C. The pH of the digester was maintained at 6.5 during the investigation. Similar BOD reduction (average 40%) was obtained for the treatment of tomato and lima bean canning wastes by anaerobic lagoons with the digester loading rate averaging 0.54 kg BOD/m^3/day (Canham, 1951).

Oliver and Dunstan (1955) reported more than 90% BOD reduction for anaerobic treatment of pea-blancher waste. The loading rate of 2.08 kg BOD/m^3/day and retention period of 10 days were used. The increase of loading rate up to 3.36 kg BOD/m^3/day had no adverse effect on the digester efficiency. It was reported that the use of

NAOH for pH control coincided with the inhibition of methane production.

Norgaard et al. (1960) studied the problem of seasonal discharge of waste from fruit canning plants in the San Jose area. During the canning season, increases were noted for the average weekly flow (from 7×10^3 to $1.2 \times 10^4 m^3$), suspended solids loading (from 59 to 100 tons) and BOD loading (from 45 to 190 tons). The pilot plant available for the study was operated only for a few canning seasons. The temperature was maintained between 26 and 29°C; however, the anaerobic treatment failed to remove sufficient BOD to justify further consideration of its use.

Parker (1966) described the treatment of food canning wastes by lagoons and ditches in Shepparton, Australia. The treatment consisted of the distinct but closely integrated installations including aerobic and anaerobic lagoons and an oxidation ditch. An effective treatment (on the average 75-80% BOD reduction) was achieved in both anaerobic and aerobic treatments when canning wastes were mixed with sewage treatment plant effluents to assure adequate supply of nitrogen and phosphate. The maximum loading rate varied according to the type of fruit or vegetables from 0.73 kg BOD/m^3/day during the citrus operation peak to 1.5 kg BOD/m^3/day for tomato and up to 2.2 kg BOD/m^3/day for the fruit processing peak.

According to work conducted by the California State Water Resources Control Board (1968), the most effective digestion of fruit cannery waste was achieved at a hydraulic retention time 10-15 days. The maximum loading rates reached 0.16 kg VS/m^3/day and 8.0 kg VS/m^3/day for raw and settled canning waste systems respectively. The average BOD removal efficiencies were less than 25%. The gas production averaged about

0.31 m^3/kg of volatile solids destroyed with methane content of 44-45%.

Parker and Skerry (1971) reported an average BOD reduction of 75-80% over the period of two years for canning waste treatment at Shepparton, Australia. Although the maximum loading rate used was 1.5 kg BOD/m^3/day at 30°C, they suggested that provided the BOD:nitrogen ratio is held below 50:1, organic loading up to 2.2 kg BOD/m^3/day with 80% BOD removal can be achieved.

Van den Berg and Lentz (1971, 1972) examined the application of anaerobic digestion for treatment of pear wastes. COD removal efficiency up to 95% was reported, depending on liquid retention time, but independent of the volatile solids loading rate. The treatment was satisfactory over the range of volatile solids loading rates from 1.6 to 7.3 kg VS/m^3/day and retention times of 0.5 to 30 days.

The supplementation with ammonia, phosphate salts and yeast extract were required for successful digestion. Minimum requirement for yeast extract addition was established at 1.5 g/l at all loading rates tested. This is somewhat unusual as the nutrients are generally required in proportion to the microbial mass rather than at fixed concentration.

2.3.5 Potato and Starch Industry

Stander (1957) experimented with the anaerobic digestion of waste from a corn starch processing plant. The retention period ranged from 3.1 to 4.7 days depending upon whether the H_2S produced during the fermentation was removed or not. The final effluent had to be mixed with domestic sewage and further treated by trickling filter.

Ling (1961) subjected the wastes from a starch-gluten plant to anaerobic digestion. The average weekly reduction of total volatile

solids was 80% at a loading rate of 1.6 kg VS/m^3/day, retention time of 3.8 days and a temperature of 35^oC. By recycling the settled biological sludge the retention period was shortened to 14 hours at 2.1 kg VS/m^3/ day with 72% volatile solids being removed.

Hindin and Dunstan (1963) demonstrated that the mixture of up to 50% of potato chip wastes in raw sludge can be satisfactorily treated by anaerobic digestion. The increase of the potato chip portion over 75% resulted in poor digestion due to mainly nutritional deficiencies. With the potato chip content greater than 25% of total, the treatment efficiency was greatly affected by a change in temperature, or waste composition unless ammonium and phosphate salts were added. Gas production increased from 4.4 to 4.8 m^3/kg VS added as the amount of potato chip portion in the waste increased from 1 to 75% respectively. At the same time, relatively stable CH_4 content of the gas was reported while the H_2S concentration decreased with an increase in proportion of potato chip in the feed. The reverse was true for CO_2 gas content.

Fossum et al. (1964) demonstrated that potato processing wastes can be readily digested when combined with domestic sewage, even when the organic load from the processing plant is approximately 15 times that of the domestic sewage.

2.4 Biochemical and Microbiological Background of the Anaerobic Waste Treatment

In the anaerobic digestion, the stabilization is brought about by a bacterial action. The end result is the conversion of complex organic molecules into stable end products, i.e. decomposition of organic carbon containing compounds into fully oxidized and reduced carbon forms, CO_2 and CH_4 respectively. A portion of the organic matter is utilized for

cellular material of the organisms present in the system; some inert or non-degradable fractions of organic material accumulate in reactor in the form of solids.

The actual production of methane involves two distinct, sequential phases, each of which is dependent on a different group of bacteria specific to that particular stage (Toerien and Hattingh, 1969).

The first, non-methanogenic, stage consists mainly of the hydrolysis and reduction of various complex organic compounds to organic acids, aldehydes and alcohols. Since this is brought about by the metabolic action of common aerobic and facultative organisms it proceeds at rapid rate.

In the second, methanogenic phase, the end products formen in the first stage are metabolized by a strictly anaerobic group of organisms, reffered to as "methane formers". This results in the formation of CO_2 and CH_4, The rate of reaction, as compared to the first stage, is relatively slow with very little heat being generated.

In any mixed culture, such as that in the anaerobic digester, many organisms are dependent on others for their growth. This could be due to the enhancement of the growth or its inhibition by the accumulation of certain metabolic intermediates or end products.

Most of the attention of researchers to date was devoted to isolation and characterization of methane forming organisms and their biochemical behaviour. As a result, the microbiology of the non-methanogenic phase is not well understood, although the presence of different physiological groups of bacteria has been demonstrated. The complexity and extreme variations in substrate composition make

the complete understanding of its microbiology almost impossible.

2.4.1 Microbiology of the Non-Methanogenic Phase

In one of the earlier studies of the digester sludge microflora Hotchkiss (1924) reported the presence of denitrifying, albumen digesting and H_2S producing bacteria.

Hungate (1950) isolated several strains of cellulolytic bacteria from anaerobic digester. Their total numbers ranged from 8×10^2 to 2×10^3 per ml, thus being somewhat lower than results published by Maki (1954) who reported 1.6×10^4 to 9.7×10^5 cellulolytic bacteria per ml of sludge.

Synergistic function of Streptococcus diploidus with other bacteria in the liquefaction and gassification of volatile solids present in sludge was reported by Keefer et al. (1953).

The presence of aerobic and facultative anaerobic bacteria of the families Pseudomonaceae, Achromobacteriaceae and Enterobacteriaceae was established by McKinney et al. (1958).

McCarty et al. (1962) studied the kinetics of anaerobic digestion using a synthetic substrate. The bacterial count of digester that received fatty acids and proteins ranged from 2×10^6 to 2×10^7 cells per ml, while those receiving carbohydrates ranged from 1.5×10^7 to 3.5×10^8 cells per ml. Initially, the predominant organism found was Escherichia coli, 9×10^6 cells per ml, however, it disappeared after several months of operation. It was suggested that these organisms may play an important role only in the initial stages of reactor start-up. This was confirmed by Toerien (1967) who was not able to isolate coliforms directly from digesting sludge after ten months of operation unless an enrichment procedure was used.

McKinney (1962) suggested that the non-methanogenic microflora of the anaerobic digester consists mainly of facultative bacteria with few strict anaerobes. This concept was questioned by Toerien et al. (1967) who reported 3.9×10^8 to 1.5×10^{10} obligate anaerobic and 8×10^5 to 1×10^8 aerobic and facultative non-methanogenic bacteria per ml of digested sludge. Furthermore, these authors found a highly significant correlation between obligate anaerobic non-methanogenic bacterial numbers and DNA contents of the digesters. Kotze et al. (1968) using this concept indicated that a much larger number of bacterial cells must have been present in the digester than were accounted for by the the aerobic counts.

In spite of all the research, uncertainty still exists about the actual roles of facultative and obligate anaerobes in the digestion process.

2.4.2 Microbiology of the Methanogenic Phase

In general, the term methane or methanogenic bacteria refers to the group of strictly anaerobic microorganisms producing methane as their metabolic by-product. Although there is a great morphological diversity among these organisms, they do have a number of similar attributes(Smith, 1965, Bryant et al., 1971, Golueke, 1958):

(i) They are all obligate anaerobes with great sensitivity to oxygen.

(ii) Their substrate and nutritional requirements are simple and narrow.

(iii) They grow only near neutral pH.

(iv) They can grow over a wide range of temperatures.

With the exception of Methanobacterium ruminantium, isolated from the rumen of fistulated cattle and sheep (Smith and Hungate, 1958), all the other methanogenic bacteria were isolated from river or lake muds and sewage sludges (Stadtman and Barker, 1951; Mylroie and Hungate, 1954).

The various species of methane bacteria that have been isolated, their substrate characteristics and classification as presented by Barker (1956), is given in Table 2.

Extreme sensitivity of these organisms to oxygen and an incomplete understanding of their nutritional requirements makes any isolation in a pure culture a difficult task. Smith (1965) studied the effect of oxygen on cell viability using pure cultures of M. ruminantium, M. formicicum, and M. barkeri and concluded that these organisms die at a rate of approximately one log unit per 4 minute exposure to oxygen.

Earlier toxicological studies indicated that the methanogenic bacteria grow satisfactorily in media containing the usual nitrite salts, CO_2, a reducing agent, an oxidizable compound, ammonia, and sulfide (Barker, 1956). Speece and McCarty (1964) used synthetic substrates to study the nutritional requirements of a mixed methane bacteria culture. They reported that the amount of inorganic salts had to be tripled when carbohydrates were used as a substrate, as compared to fats and protein substrates, in order to maintain methane production. An equation was proposed to calculate the nitrogen requirement of organisms using cellular organic nitrogen concentration, sludge retention time, loading rate and process efficiency. The phosphorus requirement for same substrates was approximately

Table 2: Classification of Methane Bacteria

(after Barker, 1956)

Family: Methanobacteriacea

1. Rod-Shaped Cells

   (a) Non-Sporulating: Methanobacterium

   M. formicicum: formate, CO_2, hydrogen

   M. propionicum: propionate

   M. sohngenii: acetate, n-butyrate

   M. suboxydans: butyrate, valerate, caproate

   (b) Sporulating: Methanobacillus

   M. omelianskii: primary and secondary alcohols, H_2, CO_2

2. Spherical Cells

   (a) Cells not in sarcina arrangement: Methanococcus

   M. mazei: acetate, butyrate

   M. vannielii: formate, hydrogen

   (b) Cells in sarcina arrangement: Methanosarcina

   M. barkeri: methanol, acetate, CO_2, H_2

   M. methanica: acetate, butyrate

1/5 to 1/7 of the nitrogen.

Smith and Hungate (1958) reported that M. ruminantium required unknown growth factors present in rumen fluid but absent in many other commonly used nutrients such as yeast extract or peptones. This was confirmed by Bryant (1965) who was able to separate this growth factor into two fractions by acidification and ether extraction. Two volatile fatty acids, acetate and 2-methyl butyric acid, present in rumen fluid were essential for growth of M. ruminantium strain M-1. Optimum concentration of acetate was 16-20 mM, suggesting that it acts as a major carbon source for the strain M-1. The 2-methyl butyric acid was required in concentrations of about 0.05 mM. More than 90% of it was incorporated into protein, and all of this was present in isoleucine (Robinson and Allison, 1969). This indicates that M. ruminantium strain M-1 lacks the ability to assimilate efficiently isoleucine from the substrate. Instead, it can biosynthesize isoleucine via the reductive carboxylation reaction.

The ether extractable growth factor, mentioned by Bryant (1965), has not been identified. It is relatively stable organic compound found in rumen fluid and produced by M. ruminantium strain PS, strain of M. barkeri, and by M. omelianskii strain MOH.

Bryant et al. (1971) studied the nutritional requirements of M. omelianskii strain MOH. The basic requirements were found to be similar to M. ruminantium with the exception that acetate was not required. Instead, CO_2 was identified as the major carbon source. The growth of strain MOH was directly proportional to the amount of CO_2 supplied at low concentrations. Similarly, as with M. ruminantium, ammonia served as the main nitrogen source. Neither peptides

nor amino acids could be substituted. One or more vitamin B complexes were either stimulatory or essential for the growth. The deletion of B_{12} or folic acid significantly depressed growth.

Ammonia serves as a main nitrogen source for methanogenic bacteria. However, severe toxicity was reported (McCarty and McKinney, 1961) when free ammonia concentration exceeded 150 mg/l. Similarly, sulfides exhibited a toxic effect when their concentration exceeded 200 mg/l.

Of great concern is the toxicity of the substrates due to the presence of heavy metals. Lawrence and McCarty (1965) examined the use of sulfates to precipitate heavy metals to control their toxic effect. Experimental digesters were fed with doses of heavy metal salts and stochiometricaly equivalent doses of sulfate salts. The ionic heavy metal concentration reached a level of several hundred mg/l but no toxic effect was observed and all fermenters performed satisfactorily. When sulfate salts were substituted with chloride salts, the gas production was almost completely inhibited. It was suggested that sulfate salts were biologically broken down to sulfides which in turn reacted with - and precipitated - heavy metal salts.

A controversy still exists about the role of volatile organic acids in toxicity and its control. Methane producing bacteria grow only at a pH range of 6.4 to 7.5. In properly balanced fermenters, pH is controlled automatically by the biochemical reactions. Volatile organic acids produced during the breakdown of complex organic substrates tend to reduce the pH, however, this is counter-balanced by their degradation and regeneration of bicarbonate buffer during the methane formation. Under unbalanced conditions, the rate of acid formation

exceeds that of its utilization resulting in decreased pH and thus inhibiting methane formation.

2.4.3 Biochemistry of the Non-Methanogenic Phase

The composition and complexity of food processing wastes varies with the type of industry, raw starting materials, etc. The following is a brief commentary on major biochemical pathways and reactions involved in the breakdown of complex organic substrates by the microorganisms (Lehninger, 1972).

(i) Carbohydrates

Carbohydrates are polyhydroxylic aldehydes or ketones with the empirical formula $(CH_2O)_n$. For classification purposes, they are divided into three basic groups:

(a) Monosaccharides or simple sugars consisting of a single polyhydroxy-aldehyde or ketone unit.

(b) Oligosaccharides containing from two to ten monosaccharide units joined in glycosidic linkages.

(c) Polysaccharides consisting of long chains of monosaccharide units.

The overwhelming bulk of carbohydrates found in nature is in the form of high molecular weight polysaccharides, namely starch and cellulose.

Starch, the predominant storage polysaccharide in plants occurs in two forms, alpha-amylose and amylopectin.

Alpha-amylose is composed of long, unbranched chains of D-glucose joined by alpha 1-4 glycosidic linkage. Depending on the origin, molecular weight varies from a few thousand up to 500 000 daltons. Amylopectin, on the other hand, is branched via the alpha 1-6

glycosidic linkage. Both forms of starch can be broken down enzymatically or by acid to individual monosaccharides or their derivatives.

Cellulose is the most abundant structural material in plants. Its backbone consists of D-glucose units joined by the beta 1-4 linkage.

McCarty et al. (1962) demonstrated, using C^{14} labelled substrate that carbohydrates are utilized via Embden-Meyerhof and hexose-monophosphate pathways during the course of anaerobic digestion. Using the presence and activities of different enzymes from glycolytic and TCA cycles, Hattingh et al. (1967) observed proportional increases in fructose-6-P kinase activity with the increase in gas production. Since this enzyme is considered to be rate limiting in the glycolitic pathway, its presence indicated that EMP pathway was functional. Study by Kotze et al. (1968) established the presence of glycolytic and glyoxylate pathway enzymes. Glucose-6-P-dehydrogenase activity, indicating the presence of hexose-monophosphate shunt, was detected only in digesters receiving synthetic substrate.

(ii) Lipids

The term lipids refers to a heterogenous group of water insoluble organic substances found in cells. They are extractable by non-polar solvents such as ether or chloroform. Some of the most prominent groups are:

(a) Fatty acids, characterized by long hydrocarbon chains and a terminal carboxyl group. The chain may be saturated or it may have up to three double bonds. Nearly all have even number of carbon atoms, usually ranging from 14 to 22. The most common are 16 to 18 carbon units.

(b) Neutral fats (acylglycerols) are fatty acid esters of the alcohol glycerol. Depending upon the degree of esterification, they are present as mono-, di-, or triacyl glycerols. Triglycerols are most common in nature and serve as energy and/or food storage.

(c) Phosphoglycerides are characterized by having one of the primary hydroxyl groups of glycerol esterified to phosphoric acid instead of the fatty acid. In addition, they contain two fatty acid residues esterified to the other hydroxyl groups of glycerol.

Other forms of lipids include waxes, steroids, terpenes, sphingolipids and glycolipids.

For fatty acids to undergo activation and oxidative degradation. they must be in the free, non-esterified form. Neutral fats and phosphoglycerides are thus hydrolyzed by specific enzymes, lipases and/or phospholipases, to free fatty acids and glycerol. The long chain fatty acids are in turn activated by thiokinases to form fatty acyl carnitine esters. Once within the mitochondrial membrane, fatty acyl coenzyme A units are removed via beta-oxidation mechanism. This last process is repeated until the fatty acid is completely broken down to individual acetate units, as exemplified by the degradation of palmitic acid:

$$CH_3(CH_2)_{14}COOH + 7\ H_2O \quad 8\ CH_3COOH + 14\ H^+$$

The presence of this two stage degradation mechanism has been demonstrated by Stadtman and Barker (1951). This was later confirmed by McCarty et al. (1962) by using C^{14} tracers of palmitic and

octanoid acids.

Chynoweth and Mah (1971) reported the formation of acetate from chemically pure palmitate at a rate similar to that of the lipid extract of raw sludge suggesting the presence of a beta-oxidation system. The addition of C^{14} palmitic acid to a $CHCl_3$ - inhibited system resulted in formation of butyrate at almost twenty times the rate of acetate and five times the rate of propionate. It was suggested that some secondary reaction involving acetate and/or propionate acted as hydrogen removal, since methanogenesis was inhibited. This assumption was partially confirmed when radioactively labelled acetate was found to be converted much faster into formate and butyrate in an inhibited rather than uninhibited sludge (Chynoweth and Mah, 1971). It leaves a possibility that butyrate formation indicates some alternative to the beta-oxidation mechanism.

(iii) Proteins

Proteins are considered to be a basic requirement of life. They constitute more than fifty percent of dry weight of biological cells and are intimately connected with all chemical and physical functions and activities of the cell.

Proteins are made up of one or more polypeptide chains containing alpha-amino acid residues covalently linked together by peptide bonds. Regardless of their origin, all proteins are made from a basic pool of 20 amino acids, arranged in specific sequences. It is the sequence that gives protein its functional properties.

For proteins to be utilized by microorganisms, they must undergo extracellular enzymatic hydrolysis by proteases and peptidases. The resulting amino acids are then transported into the cell where

they are further degraded.

Jeris (1962) reported acetic acid to be one of the major organic acids formed during anaerobic fermentation of proteinaceous material. Thiel and Hattingh (1967) reported high activities of the enzyme glutamate dehydrogenase which catalyzes oxidative deamination of glutamate to alpha-ketoglutarate and ammonia. The glutamate-pyruvate transaminase was also found quite active indicating high rate of release and/or fixation of ammonia and rapid turnover in the amino acid metabolism in a digester receiving several different substrates.

2.4.4 Biochemistry of the Methanogenic Phase

First attempts to reveal the actual biochemistry of bacterial methane production were met with only limited success.

Initially, whole cells with their complex intracellular content were used for experiments thus interfering with some reactions.

The first theory of CO_2 reduction to CO_4 was proposed by Van Niel (in Barker, 1956) using ethyl alcohol as a substrate for Methanobacillus omelianskii. In this, ethyl alcohol is oxidized to acetate and 8 electrons are released to participate in reduction of CO_2 to CH_4 and H_2O.

The advancement in separation and purification of active cell-free extracts of methanogenic organisms allowed researchers to concentrate on the actual mechanisms of CH_4 formation and transfer within the cell without any interference.

Bryan et al. (1967) demonstrated that the oxidation of ethyl alcohol and subsequent reduction to CO_2 and CH_4 was a result of two symbiotic organisms. The first organism ("S" organism) oxidized ethyl alcohol to acetate and hydrogen, while the second organism,

Methanobacillus omelianskii strain MOH, oxidized hydrogen and reduced CO_2 to methane. The carboxyl group of the pyruvate was indicated as a precursor of methane via free CO_2 as an intermediate.

Wood et al. (1965) investigated the role of tetrahydrofolate (FH_4) in the transfer of carbon 3 of serine to methane, using cell-free extracts of Methanobacillus omelianskii. When present, FH_4 acted as the acceptor of the beta-carbon atom of serine to yield glycine and ^{5}N-^{10}N-methylenetetrahydrofolate and finally CH_4 and FH_4. The role of ATP in the mechanism is not well understood, however, its presence is required. In the absence of ATP, $^{5}N^{10}N$-methylenetetrahydrofolate was oxidized via ^{10}N-formyltetrahydrofolate to formate and FH_4.

Blaylock and Stadtman (1966) reported that the amount of labelled methane formed by crude extract of Methanosarcina barkeri was proportional to the methylcobalamine added. The reaction was almost completely dependent on the addition of ATP and greatly stimulated by the coenzyme A. Removal of H_2 and pyruvate from the substrate resulted in halting methane formation from methylcobalamine as they both acted as reducing agents. Although all of the ribonucleotide triphosphates had a stimulatory effect on methane production, when added to the reaction mixtures, only ADP was found to have a similar effect of all ribonucleotide diphosphates. This would suggest that ATP rather than ADP was the active compound and was generated in the cell extract from endogenous ADP.

When 1-^{14}C pyryvate was used as a substrate, a rapid labelling of CO_2 and subsequently CH_4 was observed indicating that the methane was derived from 1-C of the pyruvate via CO_2. When 2-^{14}C or 3-^{14}C pyruvate

were used, no labelled methane was detected.

Roberton and Wolfe (1969) demonstrated that the conversion of the methyl group of methylcobalamine to methane required catalytic amounts of ATP. Once ATP had reacted, free ATP was no longer required.

The effect of ATP, added at different physiological stages of the methanogenic culture, one methane production from methyl-B_{12} was studied by Pantskhava and Bukin (1972). In their experiment, methane production was greatly induced when ATP was added during the active fermentation stage while ADP and AMP additions had very little effect. The opposite result was obtained when ATP, ADP and AMP were added to cell-free extract from the slow fermentation stage. In this case, AMP had stimulatory effect while ATP acted as an inhibitor. Suggestion that nucleotides might have regulatory rather than energy donor f[illegible] was forwarded to explain the results.

2.5 Temperature Effect in Anaerobic Waste Treatment

The effect of temperature on treatment efficiency was studied by Basu and LeClerc (1973). Working with molasses distillery wastes at mesophilic ($35^{o}C$) and thermophilic ($55^{o}C$) temperatures, they concluded that although the treatment efficiency was slightly higher at $55^{o}C$, the difference was not sufficient to prove it economically feasible.

Pfeffer (1973) examined the degradation of raw domestic solid refuse under a variety of temperatures ranging from $35^{o}C$ to $60^{o}C$. The gas production and the rate of BOD reduction at the thermophilic rate greatly exceeded that at the mesophilic rate, however, the methane content of the digester gas was somewhat lower, ranging from 55 to 62%, as compared to 53 - 70% at the mesophilic temperature.

The increase in digester temperature from 50° to 60°C resulted in proportionally higher gas production. At the mesophilic range, higher gas production was observed at 40° C rather than at 45°C. This agrees with the results published earlier by Golueke (1958) who reported the greatest gas production from raw domestic sewage at 40°C. At thermophilic conditions, the greatest gas production was achieved at 50°C (Figure 3). Results from both experiments indicate the possibility of a different group of methane producing organisms for each temperature range.

In the second part of his study, Pfeffer (1973) examined the effect of temperature and pH on the rate of cellulose utilization under anaerobic conditions. At the mesophilic temperature range, the rate of utilization was greatest at 40°C followed by [illegible] and 45°C when the pH was lower than 7.0. With a pH increase [illegible] and higher, the rate of cellulose utilization at 45°C exceeded that at 35°C with 40°C still being the optimum.

A full scale plant study on feasibility and performance of an anaerobic digester at 49°C were carried out at Hyperion treatment plant in Los Angeles, California, over the period of one year (Garber, 1954). Operationally, the thermophilic digester has posed no problem with the exception of difficulties associated with maintaining proper temperature during winter months. Once the microbial culture was established, the digester was quite stable even when the temperature varied as much as 5°C in 48 hours. Organic loading as high as 3.2 kg COD/m^3/day presented no problem even though the volatile acid content of the thermophilic digester was about 6 times of that for the mesophilic process.

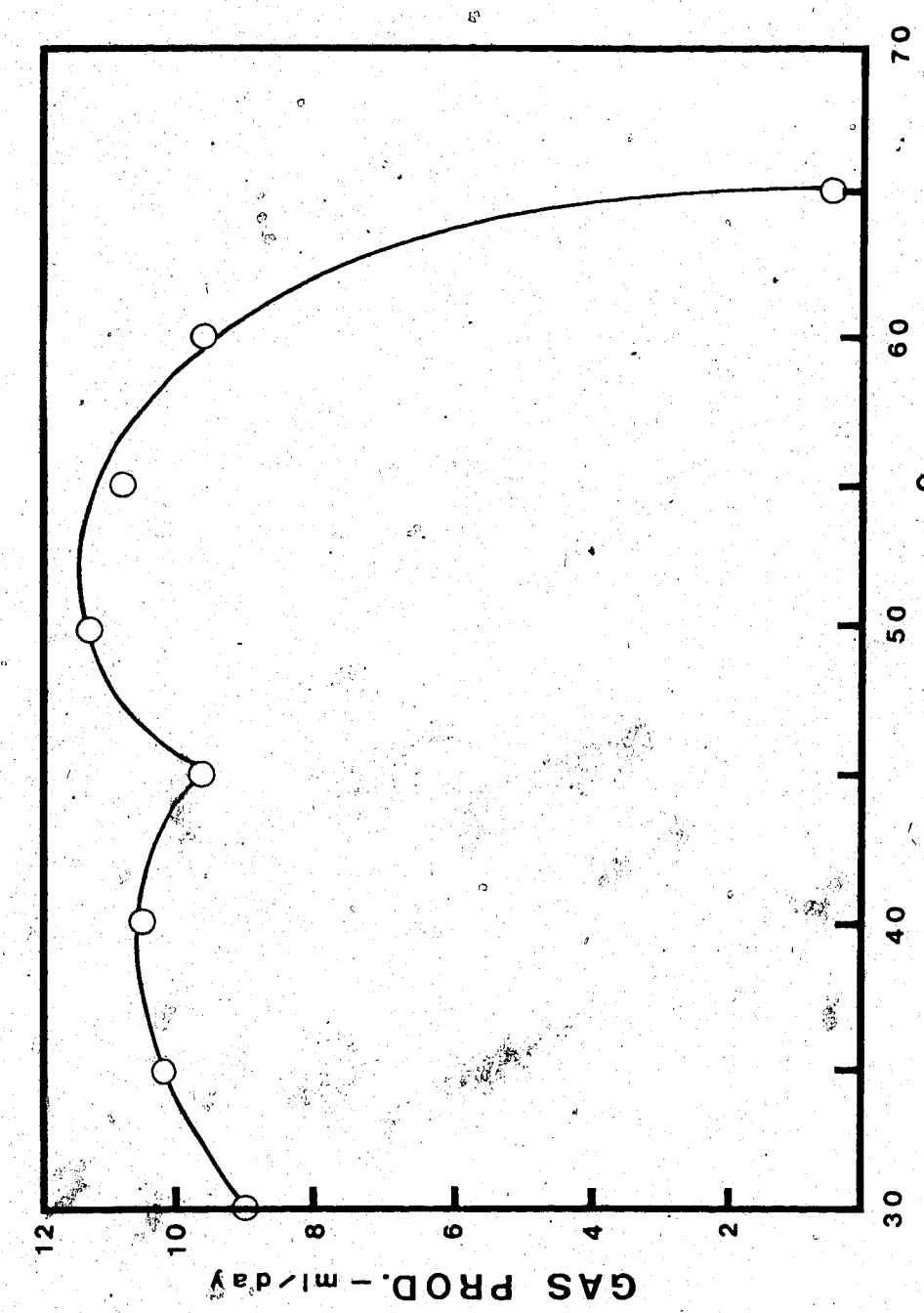


Figure 3: Effect of Temperature on Gas Production (after Golueke, 1958)

Total gas production and composition were almost identical for both temperatures. The method of mixing and gas recirculation had little effect on the course of digestion, provided adequate mixing was achieved.

Although not studied in detail, a change in the general microflora was observed. At 20°C to 37°C the flora consisted mainly of cocci-organisms encapsulated by a gelatinous sheet, while at 49°C, rod organisms with no gelatinous sheet were predominant.

The second report published by Garber et al. (1975) describes the performance of the same treatment plant over 2 years of operation at thermophilic temperatures. In this period, loading rates of 2.0 kg VS/m^3/day had been successfully treated at about 50°C. The actual temperature of reactor fluctuated from 46 to 51°C with no adverse effect on treatment efficiency.

In contrast to the first report, treatment efficiency at the thermophilic range was lower than at the mesophilic range for the same loading rates. COD reduction of 21% at 50°C as compared to 49% at 35.5° was reported. Gas production was slightly higher at the thermophilic range at 1.18m^3/kg of volatile solids destroyed compared to 1.06m^3/kg VS destroyed at 35.5°C with CH_4 gas content being almost equal.

Popova and Bolotina (1964) reported some data on the anaerobic treatment of municipal sewage at Moscow, USSR. The treatment plant was operated at a mesophilic (32.6°C) temperature, loading rate of 2.4 kg VS/m^3/day at 12.5 to 20 days retention time. Under these conditions 43-48% destruction of volatile acids was achieved. Gas production ranged from 0.15 to 0.18m^3/m^3 of the reactor, with CH_4 content

ranging from 62.8 to 64.1%. A pilot plant experiment (Popova and Bolotina, 1964) at 51.2°C resulted in the increase of organic load to 3.6 kg VS/m^3/day and a decrease in retention time from 18 to 9 days. Gas production decreased by 3-4% for the same period of time. No difference in BOD reduction (92-93%) was observed.

O'Rourke (1968) studied the kinetics of reaction and the rate of utilization of primary sludge at 15°, 20°, 25°, and 35°C. Treatment units were operated from 60 days retention time to 2.5 days or until such time at which process failure occured. Minimum solids retention time to achieve at least 80% BOD reduction were approximately 7.5, 15, and 30 days at 35°C, 25°C and 20°C respectively. At 15°C the BOD reduction was considered to be insignificant even at 60 days solids retention time. The gas production was markedly influenced by the digester operating temperature. At 35°C maximum production was achieved at 12 days while at 25°C, the same production level was reached at 30 days retention period. Retention period in excess of 60 days was required to reach same level of gas production at 20°C.

2.6 Economics of Methane Production

The selection of a particular treatment method is largely dictated by the total capital expenditures required. The survey of different meat packing waste treatment plant installations (Table 3) indicates that the least expensive treatment method is a lagoon (aerobic or anaerobic). Next is a combination aerobic/anaerobic contact system. The capital cost for trickling filter and activated sludge treatment were reported to be twice and five times higher than that for the lagoon system (Jones, 1974).

Table 3: Capital and Operating and Maintenance (O & M) Costs of Meat Packing Waste Treatment Facilities (after Jones, 1974)

Type of Treatment Facility	Medium Plant 2454m^3/day Waste		Large Plant 7646m^3/day Waste	
	Capital Cost($)	O & M Cost($)	Capital Cost($)	O & M Cost($)
Lagoon System	215,000	11,000	415,000	21,000
Trickling System	700,000	30,000	900,000	35,000
Activated Sludge			1,900,000	150,000
Anaerobic Contact (followed by activated sludge or trickling filter)	410,000	20,000	630,000	30,000

Chittenden et al. (1979) evaluated the cost of aerobic versus anaerobic/aerobic system for the treatment of meat packing plant wastes with an average flow of 13090 m^3/day (Table 4).

Capital cost expenditures for the aerobic treatment plant were almost one million dollars higher than for the combined anaerobic/aerobic system. Operating costs for the aerobic treatment were estimated to be almost 80,000 $/year higher than for anaerobic/aerobic system. In addition, utilization of the gas produced during the anaerobic stage would result in further savings of approximately 84,000 $/year.

Table 4: Capital Cost Comparison - 13090 m^3 Waste/day (after Chittenden et al. 1979)

	Aerobic System	Anaerobic/Aerobic System
First Stage Aeration	$1,256,000	$ 0
Anaerobic Lagoon	0	194,000
Second Stage Aeration	779,000	688,000
Sludge Handling Facil.	375,000	83,000
Irrigation Storage Ponds - 30 Acres	314,000	314,000
Subtotal	$2,734,000	$1,279,000
Anaerobic Lagoon Cover and Burner	0	369,000
Total Cost	$2,734,000	$1,648,000

Garber et al. (1975) reported the increased ease of separation of solids from the thermophilic, as compared to mesophilic, digester effluent resulting in savings of 300,000 $/year thus more than offsetting the additional cost of 5,200 $/year associated with extra heating.

Detailed economic evaluation of the anaerobic fermentation of

domestic solid refuse at different temperatures was presented by Pfeffer (1973). Temperatures and retention times were the major variables. The cost of the treatment at 35°C and 30 days retention time was estimated to be 93,800 $/year. Capital cost accounted for 69.3% of the annual cost. When the same substrate was subjected to digestion at 60°C and 4 days retention time, the total annual cost estimate decreased to 23,900 $/year. As a result of high temperature of the digester, the heating cost accounted for 39.3% of the total cost.

To calculate the net annual cost, savings resulting from the gas production were included. Operating at 60°C and 4 days retention time was found to be the most economical alternative. The total net benefit was estimated to be 113,000 $/year (Pfeffer, 1973).

The anaerobic treatment offers medium to low cost means of waste stabilization. The research data on the comparative treatment at mesophilic and thermophilic temperatures indicate that shorter retention time is required at a thermophilic range to achieve the same degree of BOD/COD removal. The extra cost of heating associated with high temperatures of a digester could be offset by reduced capital cost of the installation.

From this brief literature survey it appears that the anaerobic waste treatment is an economically and technically viable method for the industrial waste treatment. Its suitability for the various types of food processing wastes must still be established in terms of the waste volume, strength and potential improvements in both the waste stabilization and gas production aspects.

CHAPTER 3

EXPERIMENTAL PROCEDURES

3.1 Apparatus and Experimental Program

Fermenters utilized in experiments described in this thesis constituted two bench-scale, completely mixed, anaerobic digestion systems operated under controlled conditions. Each unit was operated at one of the two temperatures selected with Solids Retention Time (SRT) decreasing from 20 days until fermenter failure. This occured at the SRT of 1.5 days at both temperatures.

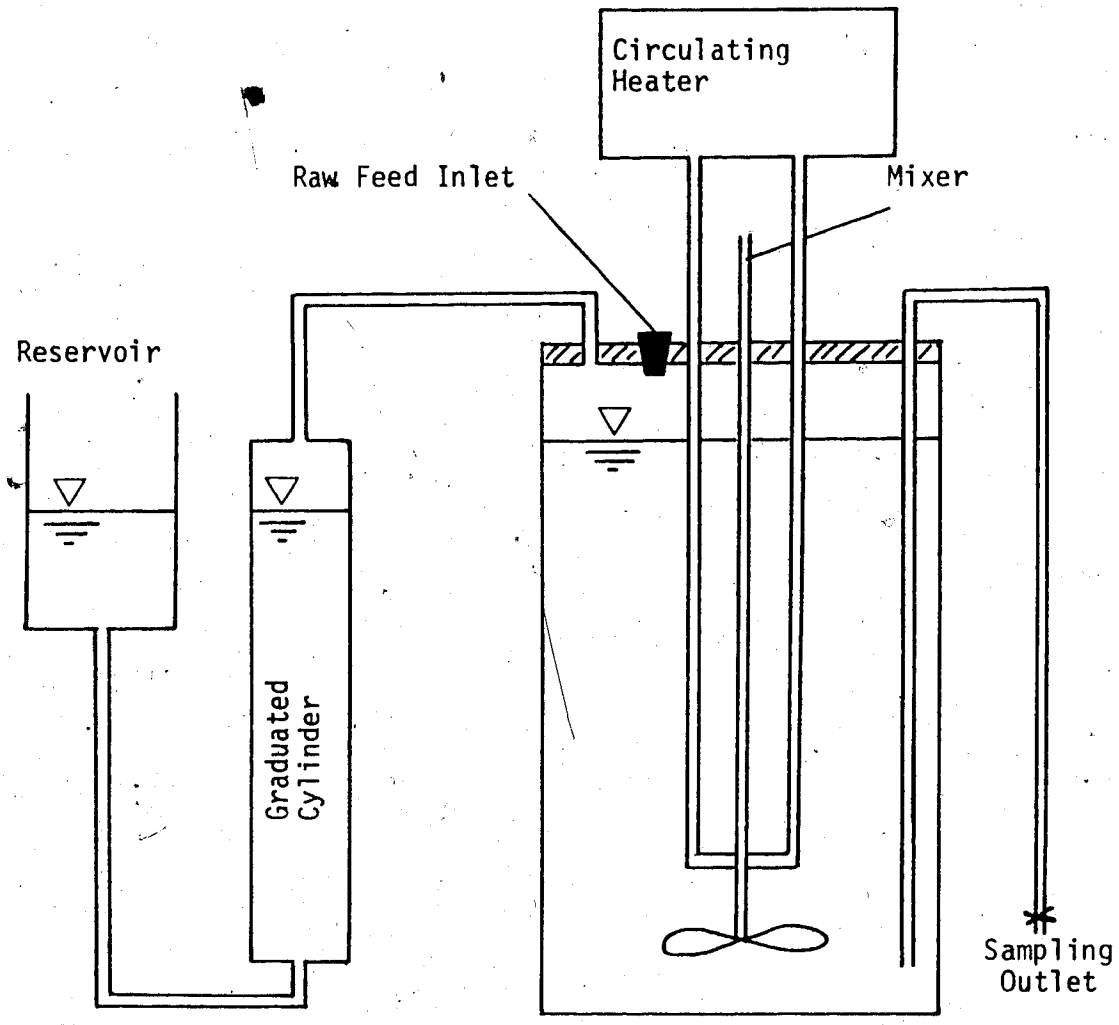
3.1.1 Experimental Reactors

Two "Microferm" fermenters, (New Brunswick Sc. Co., Edison, New Jersey) were used throughout this study. Both were equipped with precision temperature controllers and variable speed agitators. The 6 litre fermenting vessel was made of glass. The assembly lid was equipped with 1.25 inch diameter raw feed inlet. A 0.25 inch diameter tube that extended to about 1.5 inches above the fermenter bottom was used for effluent withdrawal. Another 0.25 inch assembly lid opening was connected to a calibrated tube, 2.0 inches in diameter, that was used to measure the gas volumes produced. The calibrated collection tube was connected to a reservoir of acidified salt solution which served as a retaining fluid. Figure 4 is a schematic view of the experimental reactor.

3.1.2 Substrate

In designing this experimental study, meat packing waste was selected as an example of a diluted food processing waste. The choice of the substrate was based on its suitability for the

Figure 4: Schematic of the Digester Unit



anaerobic process; continuous problems with its disposal, and its availability.

In the spring of 1975, approximately 1,000 litres of composite meat packing waste was collected in 60 litre individual lots. All but one lot were immediately frozen and kept until required. When needed, one lot was thawed at a time and stored in a 2°C cooler. The thawed liquid was thoroughly mixed for 10 minutes before use.

A complete analysis for chemical oxygen demand, total solids, total volatile solids, total volatile acids, lipids, ammonia and total nitrogen, alkalinity and pH was performed on each lot to detect any deviations from the original sample. The analytical data collected are presented in Table 5.

3.1.3 Start-Up of Anaerobic Digester

Seed sludge was obtained from the secondary digester at the City of Edmonton Waste Treatment Plant at Clover Bar.

The fermenting vessels were all sterilized and purged with a mixture of nitrogen and CO_2 gas prior to addition of sludge. Three litres of seed sludge were then added to each vessel and diluted with deareated warm water at 35°C to 6 litres. This step was used to acclimatize the sludge microflora to higher operating temperatures, as the raw sludge seed was obtained from about 20°C digester.

Starting the following day, 100 mls of effluent was withdrawn every day and replaced by meat packaging waste to slowly adapt the organisms present to the new substrate. A limited gas production was observed in 3 days.

After the evaluation of work of Pfeffer (1973) and Golueke (1958) two temperatures selected for this study were 40°C and 50°C.

Maximum rate of waste utilization and gas production in the mesophilic temperature range was reported to be at 40°C (Pfeffer, 1973, Golueke, 1958). Furthermore, the temperature of the outcoming wastes from the meat processing plant concerned averaged 39°C over 24 hour period. Almost no energy would be required to raise the temperature to 40°C. The second temperature selected, 50°C, corresponds to the lowest feasible point at the thermophilic range. Pfeffer (1973) demonstrated that the total gas production at 60°C is about 14% higher than at 50°C. In this case the digestion at 60°C was not considered to be practical as we were attempting to develop a model for a large scale industrial operation.

3.1.4 Analytical Sampling Program

The fermenters were fed once per day with raw sewage. Based on the Solids Retention Time (SRT), a pre-determined amount of digested liquid was withdrawn and subsequently replaced with equal amounts of fresh feed. The effluent was used for analytical measurements.

The volume of gas produced per 24 hour period was measured and a gas analysis was performed daily.

The analysis for COD, pH, total solids, volatile solids, ammonia and organic nitrogen, alkalinity and volatile acids were performed daily. Lipid analysis by gas chromatography was carried out on weekly basis.

3.2 Analytical Procedures

Analytical techniques used in the course of this study as well as the apparatus used are outlined below. Most of the analytical procedures were according to Standard Methods for the Examination of Water and Waste Water, 12th edition (USPHA, 1965), in further referred

to as Standard Methods (1965).

3.2.1 pH

The pH was determined daily using a Beckman pH meter-model 600 as described in Standard Methods (1965).

3.2.2 Chemical Oxygen Demand

The chemical oxygen demand (COD) of the feed and the digester effluent was determined by the reflux-method as described in Standard Methods (1965).

3.2.3 Total Solids

The evaporating dish was dried in an oven at 103°C for one hour, and subsequently cooled in a dessicator. A 50 ml sample was placed into pre-weighed, dried dish and evaporated to dryness at 103°C overnight. Samples were then cooled in a dessicator and weighed.

$$\text{Total Solids (mg/l)} = \frac{\text{Wt. of dry residue x 1,000}}{\text{ml sample}}$$

3.2.4 Total Volatile Solids

Total Volatile Solids (TVS) were determined according to the method described in Standard Methods (1965). The sample from total solids analysis was used for the determination.

3.2.5 Volatile Acids

The determination of Total Volatile Acids (TVA) was accomplished by the chromatographic column extraction method as described in Standard Methods (1965), and modified according to O'Rourke (1968).

The principal difference of the modified method is the use of the solvent CB_{20}. It consists of 320 mls of chloroform, 80 mls of butanol, and 80 mls of 0.5 N H_2SO_4. After mixing in a separatory funnel,

a top layer is withdrawn into a reservoir bottle and 65 mls of this solvent is used for each analyzed sample.

3.2.6 Grease/Lipid Analysis

Modified method of Loehr and Rohlich (1962) as described by O'Rourke (1968) was used.

3.2.7 Total Nitrogen

Total nitrogen was determined by the Kjeldahl method as described in the Standard Methods (1965).

3.2.8 Ammonia Nitrogen

Ammonia nitrogen was determined by the distillation method outlined in the Standard Methods (1965). Sample used was 25 ml and the distillate was titrated to pH 4.5.

3.2.9 Organic Nitrogen

The liquid after the ammonia distillation was analyzed for organic nitrogen using the Kjeldahl method (Standard Methods, 1965).

3.2.10 Alkalinity

Alkalinity was determined by titrating 50 ml sample to potentiometric end point of pH 4.5, using 0.02 N HCl (Standard Methods, 1965).

3.3 Chromatographic Procedures

3.3.1 Gas Analysis

The composition of the digester gas was determined by gas chromatography using a 12 foot long copper column packed with Porapak T 50-80 mesh size (Van Huyssteen, 1967).

Experimental conditions were:

Column - 12' x 3/16" O.D. copper

Packing - Porapak T, 50-80 mesh size

Temperature - Oven - 50°C

Injection port - 60°C

Detector - 100°C

Carrier gas - Helium - 30 ml/minute

Bridge current - 200 mA

Sample size - 0.1 - 0.2 ml

Under these conditions, clear separation of CO_2 from (N_2 + O_2) and CH_4 was achieved in about 7 minutes. The injection of a pure O_2, N_2, CO_2 and CH_4 gases was used to identify the individual peaks and the actual concentration (calibration) of CO_2 and CH_4.

3.3.2 Individual Volatile Acids

Centrifuged effluent supernatant was qualitatively analyzed for individual volatile acids, i.e. fatty acids with 6 or less carbon atom chains, using the gas-liquid chromatography method of Andrews et al. (1964).

Experimental conditions :

Column - 12' x 3/16" O.D. copper

Packing - 15% Carbowax 20 + 5% H_3PO_4 on 60-80 mesh A/W Chromabsorb P

Temperature - Oven - 145°C

Injection port - 210°C

Detector - 180°C

Carrier gas - N_2 - 20 ml/minute

H_2 - 25 ml/minute

Air - 300 ml/minute

Sample size - 3 µl

3.3.3 Fatty Acids

The fatty acid composition of the fermenter effluent was determined by gas-liquid chromatography. A sample from the lipid analysis (Section 3.2.6) was transesterified according to the following sequence:

(i) Incubation of 0.1 g of crude lipid + 2 ml of 2% (w/v) H_2SO_4 in absolute methanol at 75°C overnight in a sealed glass ampule.

(ii) Dilution with 3 ml of distilled water and extraction with 3 ml of hexane, repeated twice.

(iii) Combined hexane extracts were washed twice with 3 ml of 0.2 N Na_2CO_3 and twice with 3 ml of distilled water.

(iv) Small amount of anhydrous sodium sulfate was added to remove residual water.

(v) Samples were concentrated to a volume of 1 ml by a stream of nitrogen.

(vi) The concentrated solution was injected into a gas-liquid chromatograph, under the following experimental conditions:

Column - 6' x 1/8" I.D., U-shaped glass

Packing - 15% ESG on 100-120 mesh A/W Chromabsorb P

Temperature - Oven - 185°C

Injection port - 215°C

Detector - 215°C

Carrier gas - N_2 - 60 ml/minute

H_2 - 40 ml/minute

Air - 750 ml/minute

Sample size - 4 µl

CHAPTER 4

EXPERIMENTAL RESULTS

4.1 Preliminary Studies

Initial analysis of the waste from the meat packing plant (Table 5, page 55) revealed that ammonium nitrogen was present in insufficient quantities to support good growth of the methanogenic organisms. Consequently, a further investigation into the rate of ammonia addition and its effect on the treatment efficiency during the anaerobic digestion was carried out. Several 300 ml Erlenmayer flasks were filled with 100 ml of substrate and 50 ml of innoculum. Ammonia, in the form of NH_4Cl, was added in varying amounts to give a range of concentrations from 50 to 500 mg/l. The top of the liquid in each flask was flushed for 5 minutes with nitrogen and closed with a rubber stopper that had plastic tubing attached to a graduated gas measuring tube. Flasks were placed into a 40°C incubator/shaker bath. Effluent samples (10 ml) were withdrawn daily and replaced with equal amount of fresh substrate containing ammonia salt in desired concentration. During the sampling, the top of the liquid was purged with nitrogen gas to prevent exposure to oxygen. The same experiment was repeated at 50°C. Gas production and composition were monitored (Table 6).

To evaluate the effect of phosphorus addition on gas production, similar experiments were conducted at 40°C and 50°C with PO_4^{2-} being added together with ammonia at 5:1 ration of N:P (Table 7).

Table 5: Raw Waste Composition

	Average (mg/l)	Standard Deviation
COD	4356	±68.0
Total Solids	2554	±292.0
Total Volatile Solids	1982	±53.0
Total Volatile Acids (as HoAC)	63.2	±10.8
Grease/Lipids	1470	±119.0
Total Nitrogen	118	±17.0
NH_3 - Nitrogen	15.8	±1.9
Protein (Kjedahl x 6.25)	126.5	-
PO_4 - Phosphorus	35.0	±4.2
Alkalinity (as $CaCO_3$)	225	±23.0
pH	6.8	
Temperature (^oC)[1]	39.2	

[1]Discharged Waste at time of sample collection

Table 6: The Effect of Ammonia Concentration on CH_4 Production at 40^0C and 50^0C

Ammonia Concentration (mg/l)	Average CH_4 Production* (ml/day) 40^0C	50^0C
Control	5.2	5.9
50	4.9	6.5
100	5.2	7.5
150	5.1	9.3
200	4.7	11.8
250	5.5	17.5
300	13.8	21.8
350	20.8	27.1
400	15.6	28.2
450	10.4	26.8
500	6.6	25.0

*Average of four replicates

Table 7: The Effect of Ammonia/Phosphorus Concentration on CH_4 Production at 40^oC and 50^oC

Ammonia/Phosphorus Concentration (mg/l)	Average CH_4 Production* (ml/day)	
	40^oC	50^oC
Control	5.2	5.9
50/10	5.4	6.2
100/20	6.0	6.8
150/30	6.1	7.3
200/40	6.7	9.0
250/50	12.6	10.7
300/60	25.0	16.4
350/70	24.6	21.0
400/80	20.9	24.0
450/90	14.6	24.3
500/100	7.4	23.2

*Average of four replicates

The results from the experiments conducted at 40°C (Figure 5, Table 6 and 7) indicate that the addition of ammonia and phosphorus at 300/60 mg/l, had the greatest stimulatory effect on the gas production. When ammonia was added alone, the gas production reached only about 74% of that when both ammonia and phosphorus were added.

The results from 50°C digestion (Figure 6, Table 6 and 7) are somewhat surprising. Not only had the nutrient addition a more gradual effect on CH_4 production, but in this case, ammonia, when added alone, exhibited a greater stimulatory effect. Further, maximum CH_4 production at 50°C was actually reached at concentration levels that were partially inhibitory at 40°C.

To explain the difference in results at the two temperatures, two main factors should be considered:

(i) The actual microflora of the digester was probably different at different treatment temperatures. Methane gas production is a two-stage process involving first the facultative anaerobic organisms that break down complex organic molecules to substrates that can be utilized by the methane forming organisms. The microflora of the first "acid-producing" stage consists mainly of a facultative psychrophilic and mesophilic soil bacteria (McKinney, 1958). Some of these organisms might not be able to survive at 50°C: thus the change in a general microflora might result in profound changes in the nutritional requirements.

(ii) The reactions (chemical and/or) biochemical within the bacterial cell could be accelerated by the increase in treatment temperature. The possibility exists that with the change in microflora at the thermophilic temperature, different metabolical pathways

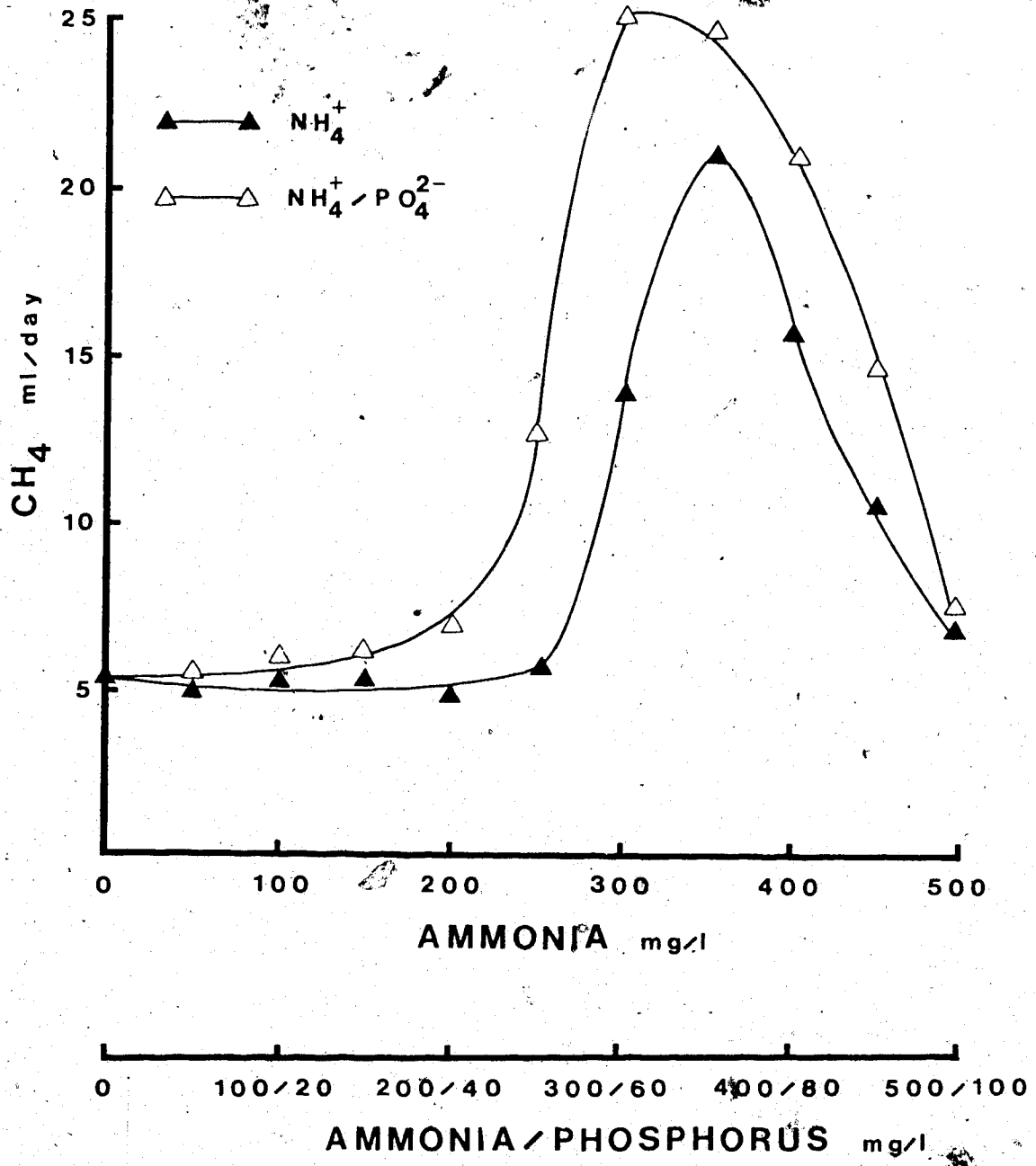


Figure 5: Effect of Ammonia and Ammonia/Phosphorus Concentration on CH_4 Production at 40^oC

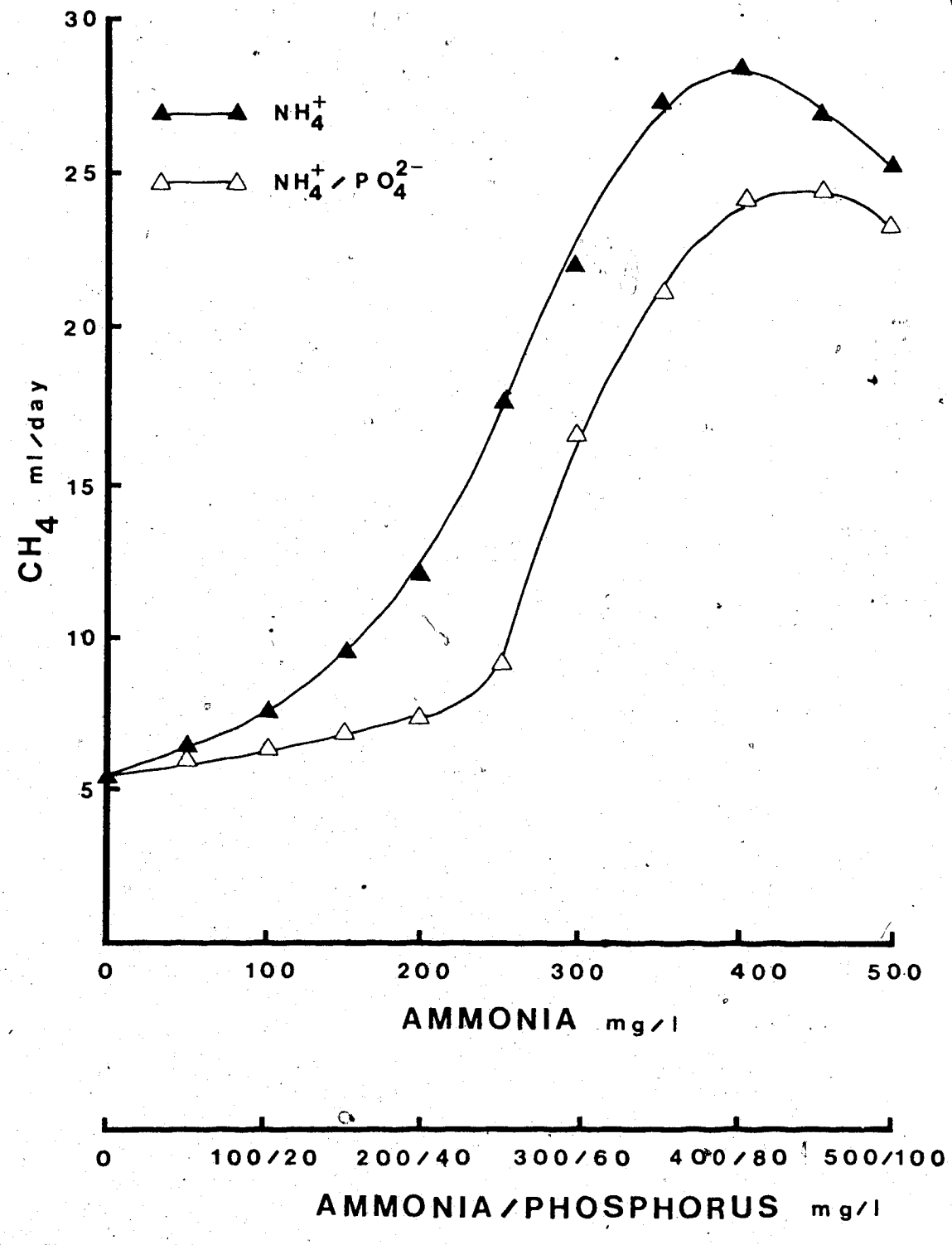


Figure 6: Effect of Ammonia and Ammonia/Phosphorus Concentration on CH_4 Production at 50°C

might be involved, resulting in different nutritional requirements.

Conditions that yielded the maximum CH_4 production in the preliminary study were applied to experimental digesters operating at 40°C and 50°C. Raw meat packing wastes that were used to feed the mesophilic (40°C) digester were supplemented only with ammonia to achieve 400 mg/l concentration. For the thermophilic (50°C) digester, ammonia and phosphorus were added to the substrate at 300 and 60 mg/l, respectively.

Both digesters were then subjected to a pre-determined programmed steady state operation. In this context, "steady state" refers to a condition where uniform reduction in COD was achieved. Once this condition was satisfied, the fermenter was operated for at least additional 30 days. During this time all operational parameters were measured.

The fermenters were started at 20 days solids retention time (SRT) and stepwise reduced to the point of failure as indicated by incomplete COD reduction to less than 60% of the original COD.

4.2 Anaerobic Digestion at 40°C

A summary of the results is presented in Tables 8 through 12. The overall effect of solids retention time on the reduction of COD, volatile solids, lipids, volatile acids and methane gas production is graphically presented in Figure 7.

Efficient treatment of the wastes studied was achieved at 4 days SRT at 40°C. At this point, COD reduction reached 91.8% (Table 8) and volatile solids were reduced by 57.1% (Table 9). Total volatile acids content at 4 days SRT was 205 mg/l expressed as acetic acid - about 3 times the concentration of the original raw sample (Table 10). This

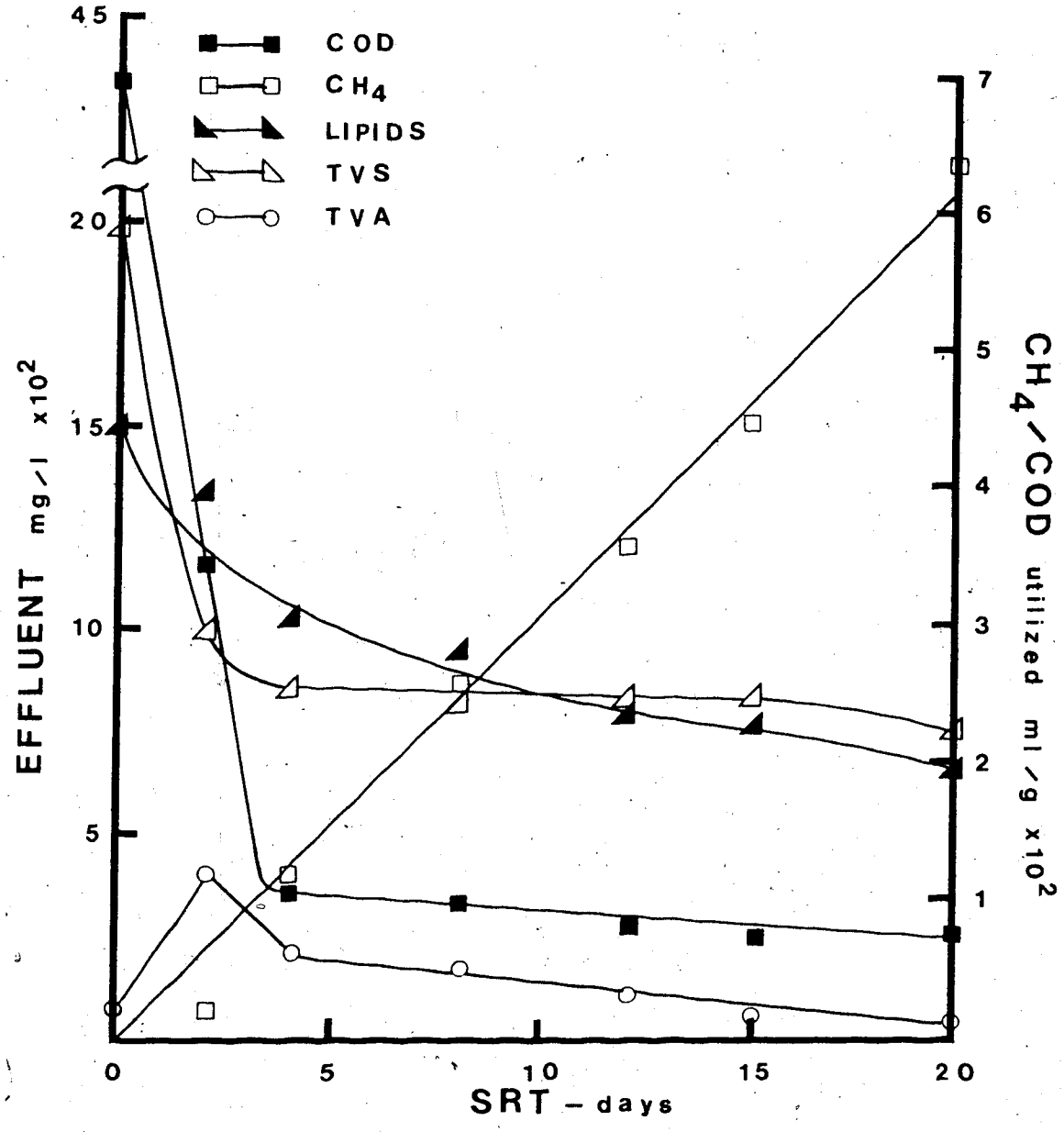


Figure 7: Effect of SRT on Utilization of the Individual Waste Components and Gas Production at 40°C

Table 8: The Effect of SRT on COD Reduction at 40°C and 50°C

SRT	COD (mg/l)				% Reduction	
(days)	40°C	Std. Deviation	50°C	Std. Deviation	40°C	50°C
20	253	±15	155	±23	92.4	96.4
15	255	±13	180	±22	94.1	95.9
12	279	±36	238	±37	93.6	94.5
8	332	±53	277	±29	92.4	93.6
4	358	±47	310	±54	91.8	92.9
2	1180	±259	824	±40.3	73.0	81.1

Table 9: The Effect of SRT on Total Volatile Solids Reduction at 40°C and 50°C

SRT	Total Volatile Solids (mg/l)				% Reduction	
(days)	40°C	Std. Deviation	50°C	Std. Deviation	40°C	50°C
20	736	±74	765	±47	62.8	61.0
15	823	±53	780	±54	58.5	60.6
12	816	±113	855	±75	58.9	56.9
8	821	±94	845	±77	58.6	57.4
4	850	±100	900	±71	57.1	54.6
2	990	±111	951	±91	50.1	52.0

Table 10: Digester Operating Conditions at 40°C

SRT		Alkalinity	Volatile Acids	
(days)	pH	mg/l $CaCO_3$	mg/l as HoAC	Std. Deviation
20	6.95	800	36	±5
15	7.00	640	44	±6
12	7.00	596	97	±15
8	6.95	542	170	±15
4	7.00	508	205	±25
2	6.85	282	430	±48

indicates that at a given loading rate, the actual breakdown of complex organic substrates by the facultative (and/or obligatory) anaerobic bacteria was proceeding at a higher rate than the utilization and transformation of the intermediates to methane gas. Equilibrium between volatile acids production and their utilization was reached between 12 and 15 days SRT. At this stage, the volatile acids concentration of digested liquid appeared to be equal to the volatile acids concentration of the raw sewage (Table 5 and 10).

Somewhat surprising was the finding regarding the low rate of lipid utilization. At 4 days SRT only about 31% reduction of total lipids was achieved. With the increase in SRT a slow increase in lipid utilization was observed, reaching about 48.3% at 15 days SRT and 55.8% at 20 days SRT (Table 11).

Gas production (Table 12) was erratic at 2 days SRT. The maximum gas production was reached at 4-8 days SRT. With the increase in SRT to 12 days or more, the gas production per digester volume (ml/day) decreased from 1198 ml/day to 997 ml/day.

Treatment efficiency at 2 days SRT was again erratic. At this point, the digester was close to its point of failure. Despite these conditions, 73% COD reduction was achieved. Volatile acids content increased to about 6.5 times its original concentration and maintenance of proper pH balance became increasingly difficult. Total alkalinity rapidly decreased from 508 mg/l (as $CaCO_3$) to 282 mg/l (Table 10). Gas production decreased to 43% of its maximum daily production and the methane content of the digester gas decreased from 66.5% at 4 days SRT to 48.6% (Table 12). A further decrease to 1.5 days SRT was attempted but met with failure as the organisms were

Table 11: The Effect of SRT on Lipid/Grease Reduction at 40°C and 50°C

SRT	Total Lipids (mg/l)				% Reduction	
(days)	40°C	Std. Deviation	50°C	Std. Deviation	40°C	50°C
20	650	±128	200	±97	55.8	86.4
15	760	±131	340	±103	48.3	76.9
12	780	±115	375	±88	46.7	74.5
8	946	±195	616	±145	35.6	58.1
4	1013	±151	700	±130	31.1	52.4
2	1331	±193	1128	±162	9.5	23.3

Table 12: Gas Production and Composition at 40°C

SRT	GAS PRODUCTION		METHANE CONTENT		METHANE PRODUCTION	
(days)	ml/day @ STP	Std. Deviation	% of Total Gas	Std. Deviation	ml/day @ STP	ml/g COD Utilized
20	985	±66	68.0	±3.1	670	636
15	980	±65	68.3	±3.4	670	450
12	997	±85	68.1	±3.3	680	358
8	1198	±65	63.4	±3.5	760	259
4	1156	±103	66.5	±5.1	750	121
2	514	±125	48.6	±8.5	250	21

washed out of the reactor.

4.3 Anaerobic Digestion at 50°C

Results from these experimental studies are included in Tables 8, 9, 11, 13 and 14. A graphical presentation of the overall effect of different SRT on the breakdown of individual waste components and gas formation is presented in Figure 8.

Steady operating conditions, together with efficient treatment and gas production, were again achieved at 4 days SRT. COD reduction reached 81.1% at 2 days SRT and increased to 92.9% at 4 days SRT. Longer SRT periods up to 20 days accounted for only a 3.5% increase in COD reduction (Table 8).

Sharp reduction in the volatile solids content from 1982 mg/l in raw waste to 951 mg/l (52% reduction) was achieved at 2 days SRT. The increase of SRT to 4 days and more had a very little effect on further volatile solids destruction (Table 9), indicating that approximately 39% of the total volatile solids was not degradable.

Volatile acids content at 2 days SRT increased to 360 mg/l. about 5.5 times its original value (Table 13). This increase would indicate an imbalance between the rate of degradation of long chain fatty acids and the utilization of free fatty acids to produce CO_2 and CH_4. Equilibrium between production and utilization of volatile acids was reached at about 8 days SRT (Tables 5 and 13).

Lipid degradation proceeded at almost twice the rate obtained at 40°C. 52.4% reduction was achieved at 4 days SRT with a steady increase to 86% at 20 days SRT (Table 11).

Contrary to the 40°C experiment, steady gas production was observed even at 2 days SRT (Table 14); however, the methane composition

Table 13: Digester Operating Conditions at 50°C

SRT (days)	pH	Alkalinity (mg/l $CaCO_3$)	Volatile Acids mg/l as HoAC	Volatile Acids Std. Deviation
20	7.00	767	35	±7
15	7.00	748	40	±10
12	7.00	617	69	±13
8	7.00	620	65	±21
4	6.95	415	110	±25
2	6.90	243	360	±75

Table 14: Gas Production and Composition at 50°C

SRT	GAS PRODUCTION		METHANE CONTENT		METHANE PRODUCTION	
(days)	ml/day @ STP	Std. Deviation	% of Total Gas	Std. Deviation	ml/day @ STP	ml/g COD Utilized
20	1110	±78	64.6	±0.9	717	662
15	1100	±94	64.9	±1.3	714	457
12	1133	±86	64.2	±0.65	727	374
8	1586	±164	62.7	±2.5	994	332
4	1520	±101	67.3	±7.1	1023	164
2	734	±60	62.6	±6.1	459	37

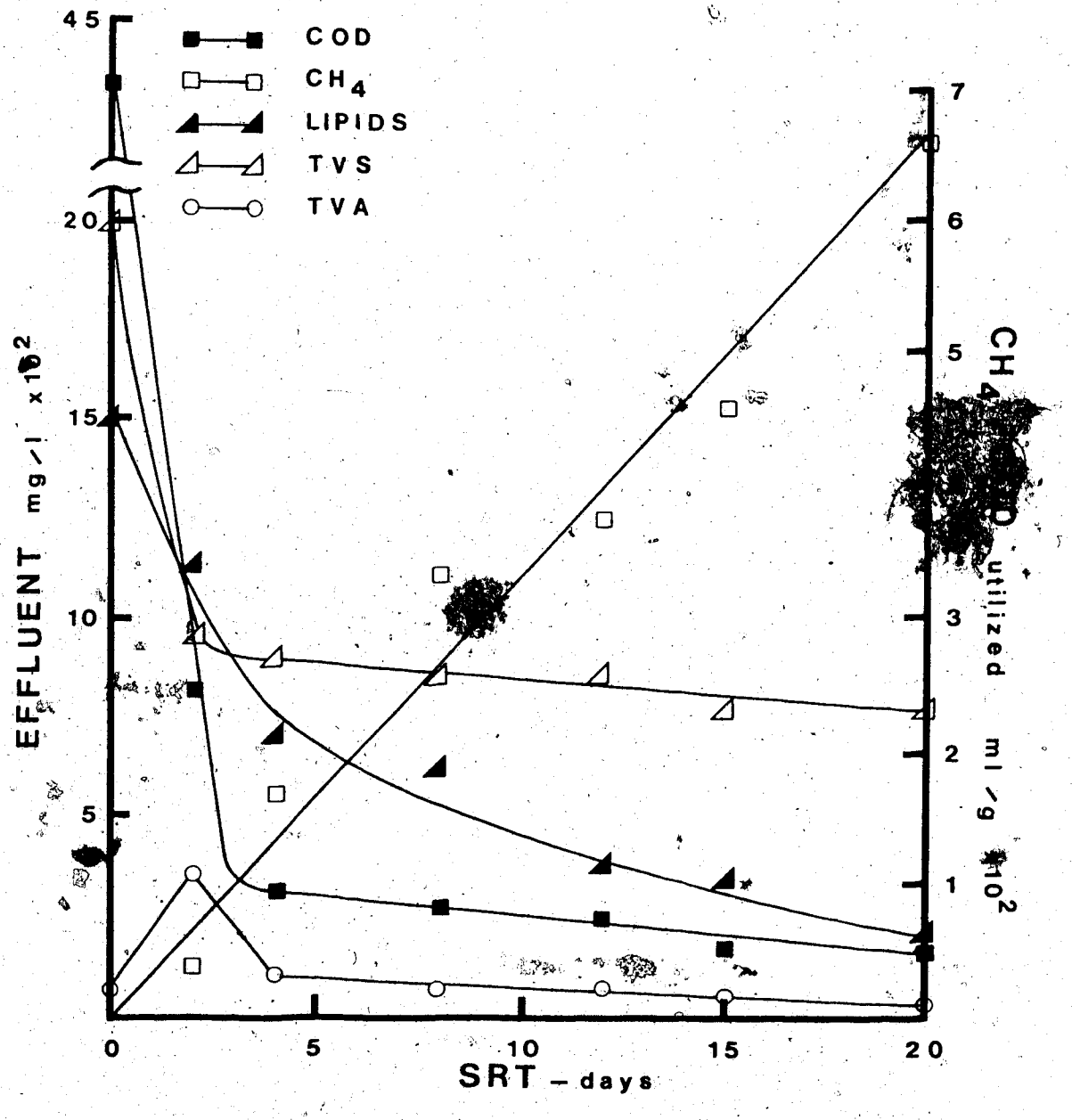


Figure 8: Effect of SRT on Utilization of the Individual Waste Components and Gas Production at 50^0C

of the digester gas was erratic, ranging from 55.9% to 70.2%. Maximum daily gas production per digester volume was reached at 4 to 8 days SRT and decreased with the increase in SRT. A further reduction of SRT to 1.5 days was attempted but failed to yield any meaningful data.

4.4 Discussion of Experimental Data

The basic objective of this study was to evaluate the efficiency of anaerobic treatment of meat processing plant waste at 40°C and 50°C. The main emphasis was placed on the rate and volumes of CH_4 production combined with efficient organic load reduction.

The data obtained in this study were also used for cost analysis evaluation of the process (Chapter 5).

4.4.1 Effect of Temperature and SRT on COD Reduction

Anaerobic treatment at both temperatures studied was effective in reducing COD (Table 8). At 40°C, COD reduction ranged from 94.2% at 20 days SRT to 73.0% at 2 days SRT. The decline in treatment efficiency was gradual up to 4 days SRT. The "break point" occured between 4 to 2 days SRT. A reduction from 4 to 2 days SRT resulted in a dramatic increase in residual COD and erratic operation.

A higher degree of COD utilization was achieved when the treatment temperature was increased to 50°C. COD reduction at 20 days SRT was 96.4% and even at 2 days SRT, 81.1% of the original COD was removed. A reduction in SRT to 1.5 days resulted in incomplete treatment, indicating that the "break point" is between 1.5 and 2 days SRT.

The maximum organic loading for the thermophilic (50°) digester to achieve at least 80% COD reduction was established at 2.18 kg COD/m^3/day, representing 2 days SRT. At 40°C 80% COD reduction would be achieved at

about 3 to 3.5 days SRT (Table 8), thus reducing the effective organic load to 1.64 and 1.36 kg COD/m^3/day respectively. This represents a 25-37% reduction in the digester utilization. It becomes important when considering the actual treatment design as it would necessitate an increase in the size (or numbers) of the anaerobic digesters required and this in turn would increase the capital investment. However, the above data for the mesophilic fermenter were obtained by extrapolating results of the COD reduction (Table 8) for comparison only. It may be that an 80% COD reduction combined with steady state digester operation would not be achieved at 3-3.5 days SRT. From a practical point of view, maximum organic loading rate for a digester operating at 40^{o}C appears to be 1.09 kg COD/m^3/day, representing 4 days SRT.

A difficulty arises when comparing the results of this study to other reports as our data are expressed in mg/l COD, while the majority of research literature data are expressed in mg/l BOD. Studies with meat packing plant wastes as a substrate indicate that majority of the organic loading rates range from 0.4 to 1.5 kg BOD/m^3/day. Organic [illegible] 4.0 kg BOD/m^3/day were successfully treated under laboratory conditions (Schroepfer and Ziemke, 1959).

The correction factor for the conversion of BOD to COD ranges from 1.6 to 1.[illegible] depending on the substrate. At these values, the maximum organic [illegible]ding rates obtained in this study, 2.18 kg COD/m^3/day at 50^{o}C and 1.09 kg COD/m^3/day at 40^{o}C, are well within the reported range.

The data obtained at 2 days SRT at 40^{o}C were somewhat erratic. Since our method of substrate feeding and effluent collection was discontinuous, the problem encountered at 2 days SRT may have been

caused by the feeding technique rather by the inability of the microorganisms to utilize the substrate. The effluent withdrawn for sampling contained a uniform mixture of sludge digested for 24 and/or 48 hours, thus not representing a true reflection of treatment efficiency related to the SRT. In fact, it might be perhaps more accurate to use a term "mean residence time", rather than solids retention time. Comparison between SRT and actual effluent "mean residence time" is presented in Table 15.

Table 15. Comparison Between SRT and Actual Effluent "Mean Residence Time"

SRT (Days)	Mean Residence Time (Days)
2	1.5
4	2.5
8	4.5
12	6.5

It appears that if the rate of the substrate addition can be controlled on a continuous basis and/or a part of the wasted sludge can be recycled, an efficient steady rate treatment could possibly be achieved at solids retention times of 1.5-2.0 days at 50°C, and 2.0-2.5 days at 40°C.

4.4.2 Effect of Temperature and SRT on Volatile Solids Reduction

Results obtained for total volatile solids (TVS) utilization indicate a very low rate of destruction. Even at the longest retention time (20 days) only about 61-62% reduction was achieved at both temperatures under the investigation. Throughout this study, temperature of digestion had very little effect on rate of TVS utilization (Table 9).

At 40°C, a rapid decrease in TVS from 2 to 4 days SRT was observed. This corresponds with erratic digester performance at 2 days SRT. A plateau was reached at 4 days SRT and TVS remained virtually constant up to 12 days SRT, then further decreasing rapidly from 12 to 20 days SRT.

The rate of TVS destruction at 50°C followed much smoother curve from 2 to 20 days SRT with the exception of results at 12 days SRT.

O'Rourke (1968) reported an 11% volatile solids destruction at 5 days SRT at 35°C, while 56% reduction in TVS was reached at 60 days SRT at the same treatment temperature.

4.4.3 Effect of Temperature and SRT on Lipid/Grease Reduction

Fatty acids are one of the main components of raw meat packing plant wastes. Therefore, the degradation of long chain and volatile fatty acids is critical in determining digester performance.

The lipid portion of the raw waste was analyzed by gas chromatography. It contained 1.24% myristic acid, 28.9% plamitic acid, 24.2% stearic acid, 36.7% oleic acid, and 3.05% linoleic acid (Table 16).

In this study, the breakdown of long chain fatty acids proceeded at a rapid rate. At 2 days SRT only traces of C:18 were detected by gas chromatography effluents from both digesters, while accumulation of C:12 fatty acid was observed (Figure 9). A sharp decrease in

Table 16: Long Chain Fatty Acid Composition of Untreated Meat Packing Waste

Individual Fatty Acid	Composition % Total Lipids	Concentration mg/l Waste
Myristic (C:12)	1.24	14.0
Palmitic (C:16)	28.9	326.0
Stearic (C:18)	24.2	277.5
Oleic (C:18:1)	35.7	414.0
Linoleic (C:18:2)	3.05	34.4

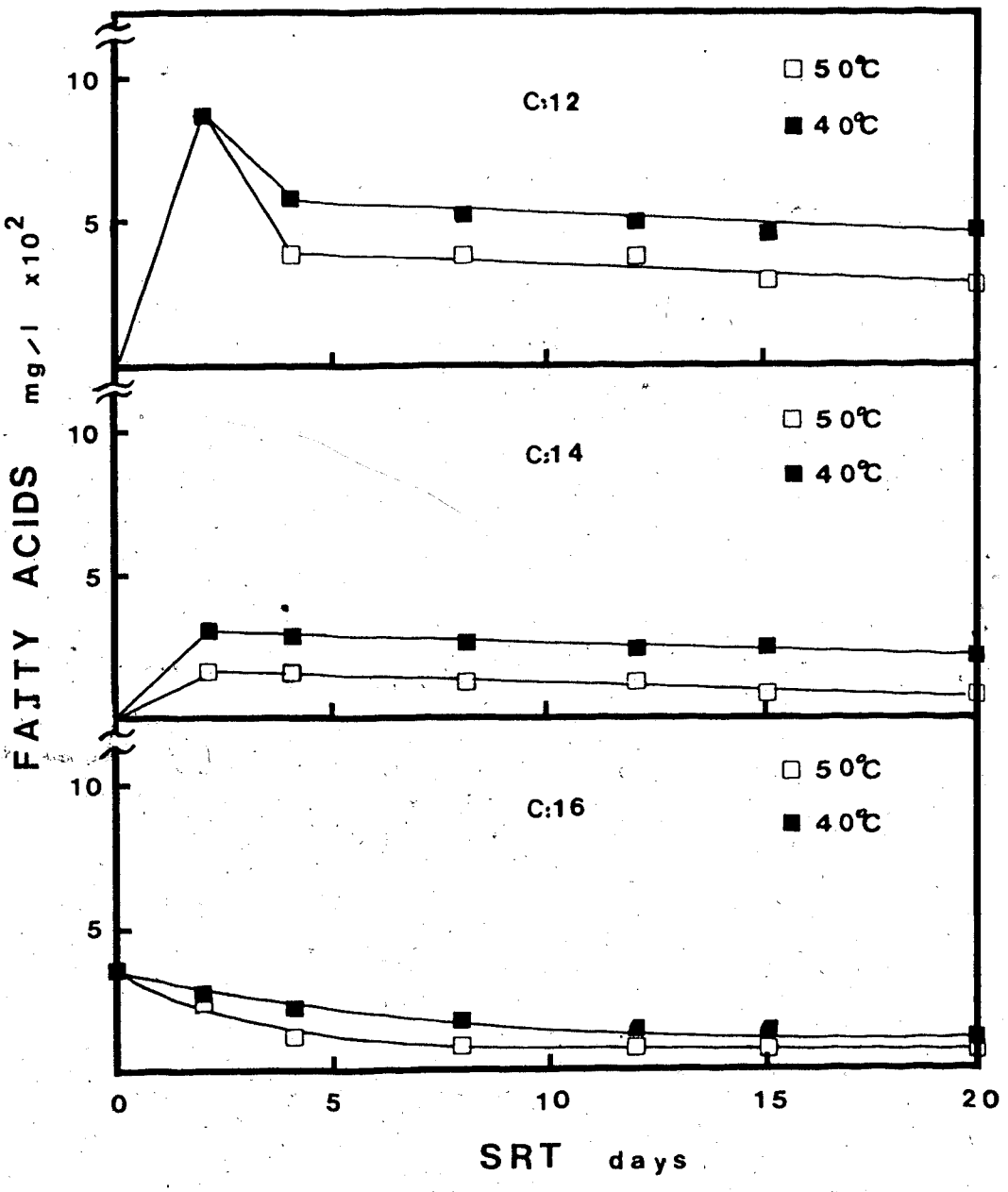


Figure 9: Effect of SRT on C:12, C:14 and C:16 Acids Effluent Concentration at 40°C and 50°C

C:12 fatty acid concentration occured between 2 and 4 days SRT, followed by a gradual decline at SRT longer than 4 days.

A similar increase, although not as notable, was observed for C:14 fatty acid concentration at 2 days SRT, reaching a plateau shortly thereafter (Figure 9). The constant level for both C:12 and C:14 at SRT longer than 4 days would indicate that approximately equilibrium conditions were established between long chain fatty acid degradation and C:12 and C:14 acid utilization.

This pattern of degradation follows that described by Novak and Carlson (1970). While studying the degradation of linoleic, stearic, palmitic and myristic acids, they observed that the unsaturated fatty acids were utilized at much faster rate than the saturated acids. The presence of the degradation intermediates indicated that the fatty acids were utilized via the beta-oxidation pathway as over 40% of the unsaturated fatty acids were converted to saturated acids, principally to palmitate.

The rate of lipid utilization proceeded almost twice as fast at 50°C than at 40°C. Although at 2 days SRT the total reduction was only 23.3%, it increased rapidly to 52% at 4 days SRT and reached 86.4% reduction at 20 days SRT. In contrast, at 40°C only 9.5% of the total lipids was utilized at 2 days SRT; the efficiency gradually increased to 55.8% at 20 days SRT.

The volatile acid concentration followed closely the rate of lipid utilization and gas formation (Figure 10, Table 10 and 13). From the original value of 65 mg/l as HoAC in the raw waste, volatile acid concentration increased to 430 mg/l and 360 mg/l in the 40°C and 50° reactors, respectively, at 2 days SRT. This buildup was

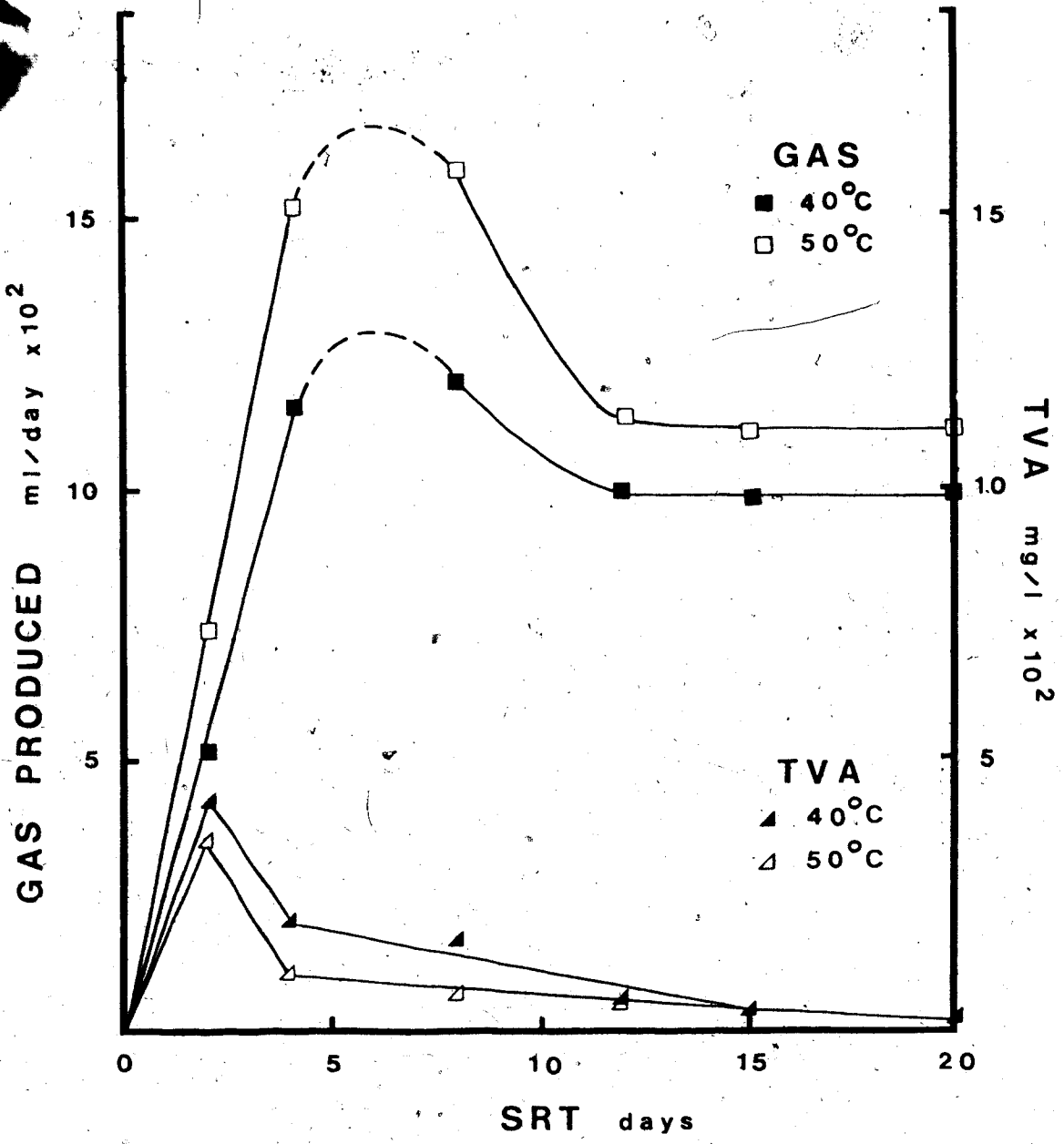


Figure 10: Effect of SRT on Effluent Total Volatile Acid Concentration and Gas Production at 40°C and 50°C

attributed to a lower rate of organic acids utilization as the fermenters were approaching the "break point" in treatment efficiency. At this stage, the volatile acid utilization appears to be the rate limiting step. Further increase in volatile acids concentration might have an inhibitory effect on methanogenic organisms.

At 50°C, after the initial increase, a gradual reduction in volatile acids concentration was observed, indicating that at SRT longer than 4 days the rate of utilization exceeded that of production.

4.4.4 Effect of Temperature and SRT on Gas Production and Composition

When comparing the total daily gas production (Figure 10), a maximum production period for both temperatures corresponds to 4-8 days SRT. At 2 days SRT a significant decrease in total gas production was observed for both temperature regimes. It corresponded closely to a marked increase in the volatile acid content of both fermenters and lower treatment efficiencies. As mentioned earlier, the volatile acids utilization appeared to be the rate limiting step.

A different pattern of gas production was observed at SRT longer than 8 days. Instead of an increase in the gas production, a sudden decrease to a constant plateau was observed. This behavior was similar for both temperatures. The lack of further increase in COD removal efficiency suggests a near depletion of the biodegradable substrate. This would indicate substrate availability to be the limiting step.

Theoretically, it would be expected that the greatest gas production would be at 4 days SRT as the indicators of the waste biodegradation are high (greater than 90% COD reduction at 50°C) and the orga-

nic load is the greatest. In practice, a slight decrease in total daily gas production was observed as compared to that at 8 days SRT.

Plotting the CH_4 production per unit of COD destroyed, versus the retention time (Figure 11) indicate near straight linear relationship between the substrate availability and the utilization.

As the organic substrate loading rate increased (by reducing the SRT), the rate of CH_4 and CO_2 production per gram of COD removed decreased. Even though there was an increase in total daily gas production, it was not proportional to the increase in organic loading rate (wt/wt). Since COD reduction is still in excess of 90% it appears that the complex organic material (organic carbon) is utilized within the system by some other means than conversion to CH_4 and CO_2. Due to the lack of experimental microbiological data, we can only theorize that this portion of organic carbon was utilized for the formation of new cell material. The fact that the rate of methane production - but not COD utilization - decreased would suggest that "acid-forming" rather than methane producing organisms were responsible for the organic carbon material utilization.

Temperature seemed to have very little effect on the gas composition (Tables 12 and 14). At the retention times greater than 8 days, the average methane content of the digester gas from the 40°C fermenter was marginally higher (68% vs 64.5%) than from the 50°C. Significant difference was observed at 2 days SRT as the methane content of 40°C digester gas decreased to 48% while that for the thermophilic digester gas remained almost unchanged at 62%.

A typical chromatogram showing separation of CH_4 and CO_2 is presented in the Appendix I.

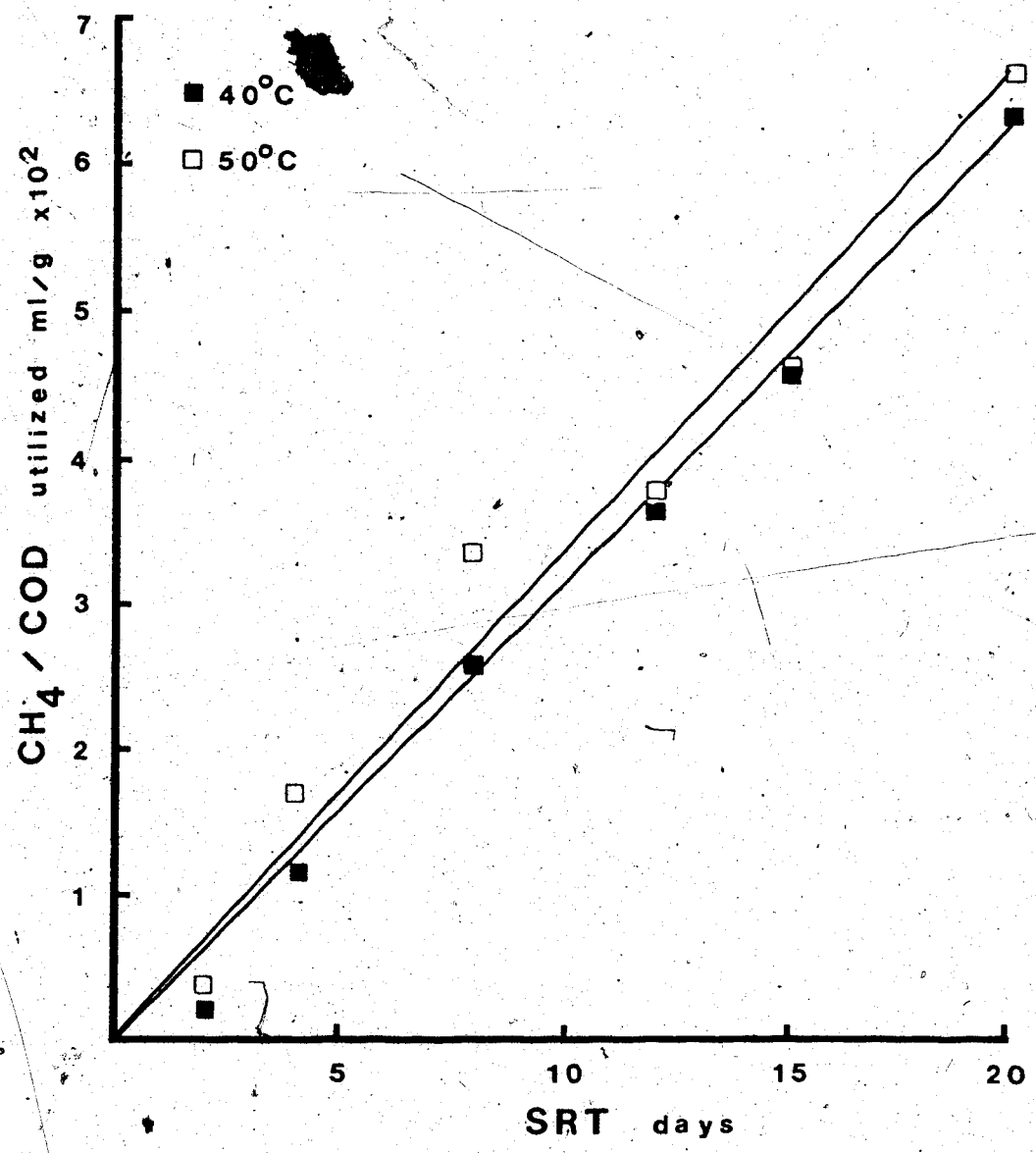


Figure 11: Effect of SRT on Methane Gas Production at 40°C and 50°C

At relatively long SRT (8 days or more) there was very little difference in treatment efficiencies between 40°C and 50°C digesters with the exception of higher rate of lipid utilization at 50°C. The difference between the two temperatures became apparent at short SRT, especially at 2 days. At this point, 50°C digester treatment efficiencies are clearly superior to 40°C digester, removing 81% COD vs 73% at 40°C. In addition, the CH_4 production (459 ml/day vs 250 ml/day and the lipid utilization (23.3 vs 9.3%) were also higher. At 4 days SRT, the 50°C was marginally more efficient than the 40°C treatment with both treatments being highly effective in terms of waste stabilization and methane production. Thus, SRT of 4 days was used for the cost analysis of the two systems.

CHAPTER 5

COST ANALYSIS

5.1 Introduction

Many food processing operations, such as meat, potato, and vegetable processing generate large amounts of organic waste. At present, neither useful by-products nor energy contained in this waste can be profitably recovered under most processing conditions.

Only relatively small portion of the energy available can be utilized in the form of "bio-gas", produced during the anaerobic fermentation. Most of the treatment units built so far are uneconomical mainly due to the large volumes of the digester required (i.e. high capital investments) and a relatively low gas production.

It is obvious that the volume of a digester has a marked effect on the capital cost of the treatment plant. Reduction in the digester size can be accomplished by concentration of waste liquid to reduce its volume or by increasing efficiency of the treatment by decreasing the necessary residence time.

Experimental results from Chapter 4 indicate that at the short SRT the rate of organic waste reduction was significantly higher at $50^{o}C$ than that at $40^{o}C$. Therefore, the objectives of this cost analysis were:

(i) To evaluate the economic feasibility of thermophilic ($50^{o}C$) as compared to mesophilic ($40^{o}C$) anaerobic digestion, and

(ii) to evaluate evaporation as one possible method of reducing the total daily volume of wastes that have to be treated.

5.2 Methodology

Total cost of the simple anaerobic waste treatment at two different temperatures was estimated in relation to the cost of the installation, operating and maintenance expenditures and benefit derived from utilizing the gas produced during the digestion.

The cost of waste concentration by evaporation to decrease the digester unit cost was also estimated. A comparison of costs at the two treatment temperatures was made for the simple anaerobic treatment as compared to the combined evaporation/anaerobic system to find the most economically feasible alternative.

5.2.1 Assumptions and Base Line Data

The method of evaluation was based on the following criteria:

(i) The cost of the construction and equipment obtained from technically valid but economically outdated sources was adjusted to December, 1981 using the Engineering News Record construction costs index of 344.93 (1967 = 100).

(ii) Cost of supplies, utilities, financing and labour (Table 17) were taken at current rates (December, 1981) for Edmonton, Alberta.

(iii) The process parameters used, such as the gas production, solids retention times and nutrient requirements (Table 18) were obtained from the experimental results described in the Chapter 4. An assumption was made that the nutrient requirement of concentrated waste would increase in relation to concentrating factor.

Table 17: Current Cost Data used for Analysis of the Anaerobic Waste Treatment

Parameters	Unit Rates/Cost	Source
Prime Interest Rate	16½%	Bank of Montreal
Water	0.10 $/m^3	Edmonton Utilities
Sewage	0.059 $/m^3	Edmonton Utilities
Union Labour Rate	10.50 $/hr	Manpower & Immigration Office, Edmonton, Alberta
Gas	0.07 $/m^3	Northwestern Utilities
Electricity	0.036 $/KWh	Edmonton Power
Ammonium Chloride	0.50 $/kg	CIL Chemicals Co.
Sodium Hydroxide	1.85 $/kg	Dow Chemicals
Sodium Phosphate	1.75 $/kg	CIL Chemicals Co.

Table 18: Technical Data used for the Cost Analysis of the Anaerobic Digestion

	Plant 1	Plant 2	Plant 3
Waste Flow (m^3/day)	1893	3785	7570
Solids Retention Time (days)	4	4	4
Methane Gas Production (m^3/day)			
Thermophilic Digestion	1290	2580	5160
Mesophilic Digestion	946	1892	3784
Digester Temperature (^oC)			
Thermophilic Digestion	50	50	50
Mesophilic Digestion	40	40	40
Nutrient Requirement (kg/day)			
Thermophilic Digestion			
Ammonium Chloride	757	1514	3028
Mesophilic Digestion			
Ammonium Chloride	568	1136	2272
Sodium Phosphate	114	228	456

Heating Value of Gas = 3.58×10^7 Joules/m^3

Operating and Maintenance Labour = 8% of Capital Cost

Equipment Life = 25 years

(iv) Three different sizes of the treatment units (Table 18) were selected on an average daily waste flow for small, medium and large meat processing plants as reported in literature (Jones, 1974).

5.2.2 Cost Estimation of the Anaerobic Digestion

The plant design considered was based on the conventional process with no provision for recirculation of solids. Technical data the calculation are given in Table 18.

The volume of the digester, based on a constant feed concentration was estimated according to Pfeffer (1973), as

$$V = \frac{M}{Cp} T$$

Where: V = Volume of reactor, m^3

M = Kg of solids/day

C = Feed concentration, %

p = Density of feed, kg/m^3

T = Retention time, days

To estimate the total capital cost of the digester unit, the formula developed by Smith (Eckenfelder and Adams, 1972) was selected and the cost was updated to December 1981, using the ENR cost index of 344.9; thus,

$$\text{Cost}(\$1000) = V(1.34 + \frac{13.8}{V^{0.87}})$$

Where: V = Volume of reactor, 1000 ft^3

The operating and maintenance costs were fixed at 8% of total capital cost (Eckenfelder and Adams, 1972). The amount and value of

gas produced was estimated from the results in Chapter 4 for incorporation into total costing procedure. The amount and cost of chemicals required for nutrient supplement was also estimated from the experimental results in Chapter 4. These data were used for estimation of the anaerobic treatment at 40°C and 50°C.

5.2.3 Cost Evaluation of the Combined Evaporation/Anaerobic Treatment

A survey of recent technical developments and economics of large scale evaporation methods was made to select a suitable process for the analysis of the waste concentration approach.

Since the multi-stage flash distillation (MSF) offers a high performance ratio (Spiegler, 1966) and ability to process large quantities of liquid, this process was evaluated for its feasibility as a mean of waste concentration.

The basis of calculations for evaporation for the three plant sizes described in 5.2.2 is given in Table 19. Operating data were calculated as described by Porteous (1975), Howe (1974) and Spiegler (1966). Basic assumptions, specific values and equations used are presented in Appendix II. Appendix III summarizes the mass flow for the smallest ($1893 m^3$/day) waste treatment unit.

The concentration factor of 10 was selected arbitrarily and at this stage the assumption was made that the gas production, solids retention time and the nutrient requirements of the subsequent anaerobic treatment will be identical to those from the experimental part (Chapter 4).

The costs of the evaporator, operating and maintenance costs and costs of utilities were calculated according to Clark et al. (1969). The cost of the anaerobic digester was estimated as described in 5.2.2.

Table 19: Basis of Calculations for Evaporation

General Factors			
Plant Size (m^3/day)	1893	3785	7570
Period of Operation		300 days/year	
Feed Characteristics		0.25% TS at 38°C	
Evaporator Factors			
Number of Stages	22	25	31
Maximum Temperature (°C)	120	120	120
Concentration Ratio	10	10	10
Performance Ratio	10	11.7	14
Electric Power Requirement (KWh/day)	9000	18000	36000

Financial Factors	Equipment Life	Interest
Equipment	25 years	16½%
Insurance	0.25% of Plant Investment	

5.3 Results

5.3.1 Cost Comparison of Anaerobic Treatment at 40°C and 50°C

When considering the total cost of any kind of waste treatment, one should not think in "money-making" or profit terms. The basic concept is "how can we decrease the necessary cost associated with waste treatment".

The cost of the anaerobic units and the benefit derived from the gas production are presented in Table 20 and 21. The cost of the digester is dictated by the volume of the waste to be treated and by solids retention time required to achieve at least 90% BOD and/or COD reduction. The cost of the supplemental heating is dependent on the temperature differential between the reactor and the outside air temperature and the amount and kind of insulation used to minimize the heat losses.

When comparing the efficiency of CH_4 gas production, the digester operating at 50°C produced almost 36% more of the methane gas. However, even at this rate, it still was not sufficient to account for the extra cost of heating required due to the digester heat losses and the energy required to increase the incoming sludge temperature to 50°C.

On the other hand, the digester operating at 40°C will be self sufficient with some extra energy available to be used elsewhere.

Examination of the cost comparison (Table 22) indicates that the greatest expenditure ($/day) is the cost of chemicals required as supplemental nutrients, followed by the operating and maintenance cost and the capital cost.

There is very little that can be done to offset the operating and maintenance cost except for limited automation. Alternative

Table 20: Cost Analysis of the Anaerobic Treatment at 40°C and 4 Days SRT

	Plant 1	Plant 2	Plant 3
Waste Flow (m^3/day)	1893	3785	7570
Reactor Volume (m^3)	7545	15090	30180
Capital Cost ($1000)	680	1319	2588
O & M Cost ($1000/year, at 8% C.C.)	54.5	105.5	207
CH_4 Gas Produced (m^3/day)	946	1892	3784
Gross Heat Value (J/day)	3.43×10^{10}	6.78×10^{10}	1.35×10^{11}
Estimated Heat Losses (J/day)	2.38×10^{10}	4.77×10^{10}	9.54×10^{10}
Net Energy (J/day)	1.05×10^{10}	2.01×10^{10}	3.96×10^{10}
Value of Available Energy ($/day, at -.07 $/$m^3$)	25	50	98

Table 21: Cost Analysis of the Anaerobic Treatment at 60°C and 4 Days SRT

	Plant 1	Plant 2	Plant 3
Waste Flow (m^3/day)	1893	3786	7570
Reactor Volume (m^3)	7545	15090	30180
Capital Cost ($1000)	680	1319	2588
O & M Cost ($1000/year, at 8% C.C.)	54.5	105.5	207
CH_4 Gas Produced (m^3/day)	1290	2580	5160
Gross Heat Value (J/day)	4.61×10^{10}	9.24×10^{10}	1.85×10^{11}
Estimated Heat Req.. (J/day)	1.16×10^{11}	2.20×10^{11}	4.40×10^{11}
Net Avail. Energy (J/day)	-6.99×10^{10}	-1.27×10^{11}	-2.55×10^{11}
Value of Avail. Energy ($/day, at 0.07 $/$m^3$)	-142	-285	-578

Table 22: Comparison of the Anaerobic Treatment Cost per day at 40°C and 50°C and 4 Days SRT

	Plant 1		Plant 2		Plant 3	
Waste Flow (m^3/day)	1893		3785		7570	
Temperature (°C)	40°C	50°C	40°C	50°C	40°C	50°C
Capital Cost ($/day)	74.5	74.5	144.5	144.5	283.6	283.6
O & M Cost ($/day)	149.3	149.3	289.0	289.0	576.1	576.1
Chemicals ($/day)	484.0	378.0	968.0	756.0	1936.0	1512.0
Gas Credit/Debit ($/day)	25.0	-142.0	50.0	-285.0	98.0	-578.0
Total Cost ($/day)	682.8	743.8	1351.5	1474.5	2697.7	2949.70

chemicals would have to be sought to provide cheaper source of ammonia and phosphorus. Despite high cost, these supplements may be necessary if maximum energy production is desired. However, the reduction in capital cost appears to be the principal variable that can be substantially influenced by technological improvements.

As stated earlier, the capital cost is directly proportional to the waste volume and the solids retention time. A partial decrease in total volume can be achieved by careful management of water usage within the plant and by re-directing some water (if permissible) to the storm sewers.

The greatest effect can be achieved by increasing the biological activity of the digester, thus reducing the retention time and the reactor volume. This can be accomplished by increasing the treatment temperature from 40°C to 50°C. In previously described experiments (Chapter 4) a digester was operated at 50°C and 2 days SRT with 81% COD removal. This would reduce the total volume of digester for treatment plant with waste flow of 1893 m^3/day from 7545 m^3 to 4716 m^3, resulting in capital cost decrease from 74.5 $/day to 48.2 $/day. Under the same conditions, the capital cost for a medium size plant would be 74.5 $/day (vs 144.5 $/day) and for a large treatment plant 144.5 $/day vs 283.6 $/day.

However, marked reduction in the methane gas production was observed at 2 days SRT. As the retention time is nearing the treatment "break point", the efficiency of the organic matter removal is reduced and the digester is more sensitive to shock loadings; a condition unacceptable for the commercial application.

Cost comparison (Table 22) indicates that at 4 days SRT the

treatment at 40°C is more economical as it is not only self-sufficient in energy requirements, but additional energy in the form of CH_4 is available for other uses. Although the CH_4 gas production at 50°C was approximately 36% higher than at 40°C, it was not sufficient for the heat requirements of the digester. The large amounts of supplemental heat required would make this alternative uneconomical.

5.3.2 Evaporation/Anaerobic Digestion Treatment

The cost estimate of the MSF evaporation plant (Table 23) and the operating and maintenance estimate (Table 24) clearly indicate that the most important cost variables in decreasing order are:

(i) Cost of steam

(ii) Electric power cost

(iii) Operating labour

(iv) Capital cost of evaporator

With the exception of operating labour, there is an interdependence of other process variables, thus an optimum combination has to be found to yield the lowest costs.

Steam cost and the required surface area (therefore, capital cost) are interrelated by the performance ratio (PR); mass of distillate/mass of steam supplied. The increase in PR results in decreased steam consumption, however, at the same time, the total surface area required for evaporation increases, thereby increasing the capital cost. Simultaneously, as the number of stages increases, electric power requirements for pumping would increase.

The cost of evaporation was combined with cost required for a treatment of concentrated anaerobic waste for an economical analysis.

Table 23: Estimate of Multi-Stage Flash Evaporation Plant Capital Cost

Plant Size (m^3/day)	1893	3785	7570
Evaporator & Accessories	609,000	1,556,000	3,633,000
Indirect Capital Cost Eng. & Supervision	105,000	186,200	434,000
Building & site Improvement	110,000	290,000	684,000
Contingency (10%)	82,400	203,200	475,100
Total Cost	906,400	2,235,400	5,226,100

Table 24: Estimate of the Operating Cost of Multi-Stage Flash Evaporator ($/day)

Plant Size (m^3/day)	1893	3785	7570
O & M Cost	230	230	230
Supplies & Maint. Material (0.25% of C.C.)	74	18	49
Electric Power	266	533	1065
Steam Cost	1111	1455	2433
Total Operating Cost ($/day)	1614	2236	3777

5.4 Comparison of Anaerobic Digestion and Evaporation/Anaerobic Process

To evaluate the cost effectiveness of the reduction in total waste volume an economic comparison of the anaerobic versus evaporation/anaerobic treatment is necessary.

Taking as an example a plant with a total waste flow of 1893 m^3/day, the cost of evaporation will be in order of 2100 $/day. By decreasing the waste volume the capital cost for the anaerobic digester would be reduced by a factor of 10, resulting in savings of about 65-67 $/day. Furthermore, the distillate can be withdrawn at any desired temperature thus eliminating the need for heating of the digester incoming sludge. This would represent savings of 114 $/day in the case of 50^{o}C treatment.

From Appendix III, 7.11 x 10^4 kg of water/hour is recovered and can be reused within the plant. At a present rate of 0.10 $/$m^3$ (Table 17) this would represent approximately 210 $/day saving. Distillate produced can be considered bacteriologically safe, however, strong objectionable odor would require further treatment, thus decreasing the saving value. Since the above quantities of water will not be discharged into sewer, 125 $/day will be saved in sewer charges.

The above mentioned data are presented in Table 25. From this data it follows that evaporation as a mean of concentrating the raw waste prior to the anaerobic treatment would not be economical as it would result in additional increase in cost of approximately 963 $/day for 1893 m^3/day treatment plant. However, the condition of sewage discharge for each individual food processing plant changes from one

Table 25: Comparative Cost of the Anaerobic Waste Treatment and the Combined Evaporation/Anaerobic Digestion Treatment ($/day)

	Anaerobic Treatment	Evaporation/ Anaerobic Treatment
Waste Flow (m^3/day)	1893	1893
Capital Cost ($/day)	74.5	130
Operating Cost ($/day)	149	1963
Cost of Chemicals ($/day)	484	378
Value of Gas Produced	+25	+114
Water Cost/Saving ($/day)	237	+210
Sewer Charges/Savings ($/day)	140	+125
Total Cost ($/day)	1059.5	2022.0

location to another. Before deciding on the specific type of treatment, each waste stream must be judged on its own merit depending on local conditions.

CHAPTER 6

CONCLUSION AND RECOMMENDATION FOR FUTURE WORK

The waste from meat processing plant was successfully treated under anaerobic conditions at 40°C and 50°C. COD reduction of at least 90% was achieved at both temperatures at 4 days SRT, combined with satisfactory methane gas production.

The treatment at 50°C resulted in approximately 36% increase in methane gas production as compared to that at 40°C. However, large quantities of an external heat were required to raise the temperature of the incoming sludge to maintain the digester at 50°C, thus making it uneconomical.

The increase in the treatment temperature from 40°C to 50°C resulted in the increase of maximum loading rate from 1.64 to 2.18 kg COD/m^3/day respectively.

From the economical point of view, the optimum cost-treatment efficiency combination was achieved at 40°C and 4 days SRT. The concentration of waste by evaporation and subsequent treatment by anaerobic digestion would result in additional cost of 963 $/day, mainly due to the high cost of steam (1350 $/day) required.

As a result of this study, the following topics are suggested for future research:

1. A comprehensive study of the digester microflora at mesophilic and thermophilic temperatures
2. Study of the nutritional requirements of the microorganisms at 50°C that would result in increased biogas production.

3. Anaerobic digestion at mesophilic and thermophilic temperature using continuous substrate feed system (e.g. anaerobic filter application).
4. Two stage anaerobic digestion system - to separate "acid forming" stage (at 35-37^{o}C) from methane producing stage (at 50^{o}C).

In today's food manufacturing, waste, because of its unproductiveness and the cost associated with the treatment, is looked at as the "necessary evil". The increased cost and need for energy will undoubtedly make the anaerobic process more and more acceptable not only as an effective treatment but also as a contributor of a valuable energy that could be utilized by the particular plant concerned.

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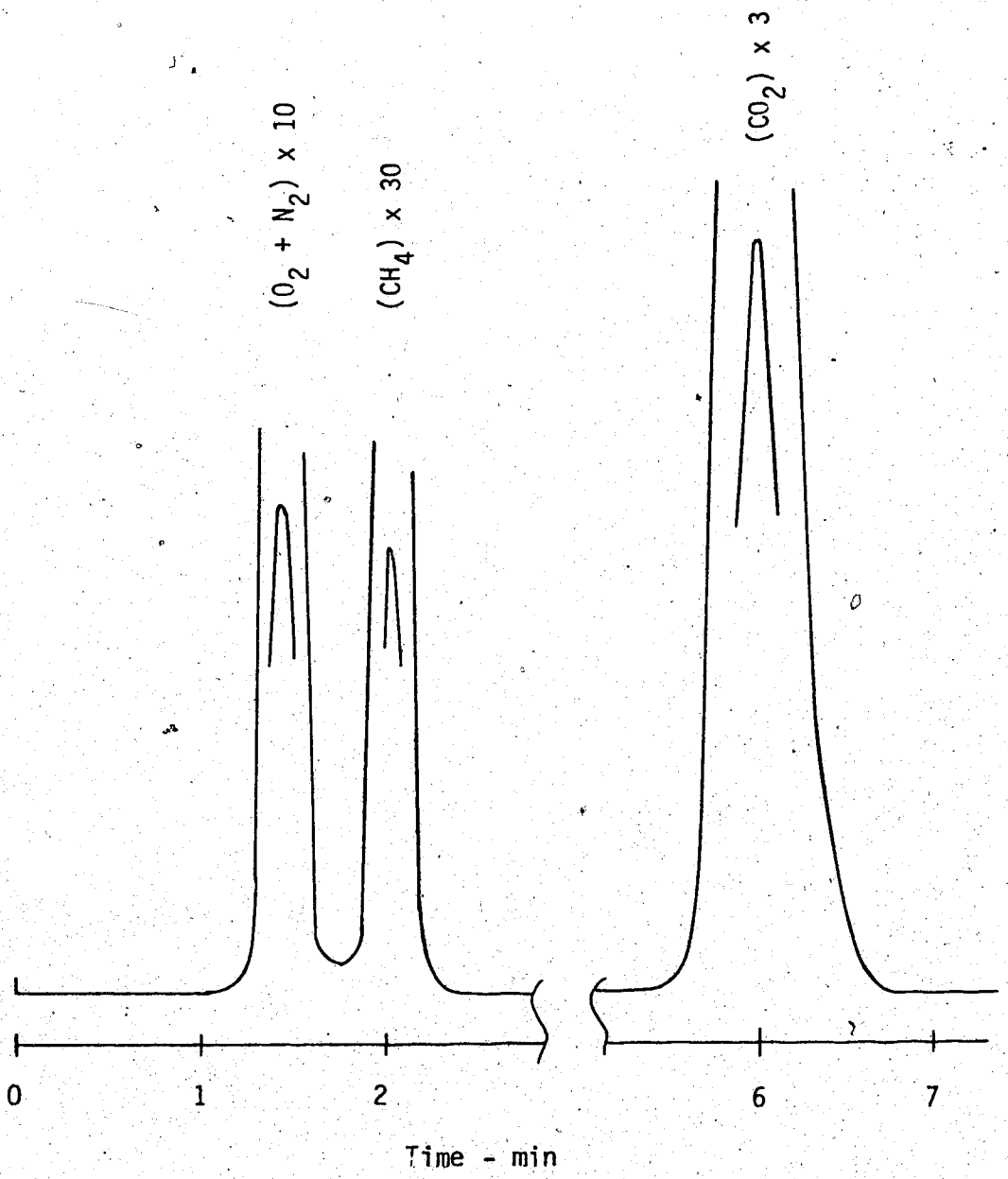
APPENDIX I



A Typical Chromatogram Showing Separation of $(O_2 + N_2)$, CH_4 and CO_2 Fractions

APPENDIX II

Values used for calculations:

C = Spec. heat of waste = 4200 J/kg ^{o}C

L = Latent heat of vaporization = 2.33×10^{6} J/kg

U = Overall heat transfer coeff. = 2830 W/m^{2} ^{o}C

Waste recirculated/kg of distillate:

$$r = \frac{L}{C(Tmax - Tdis)} = \frac{Wr}{Wd}$$

Performance ratio:

$$R = \frac{Tmax - Tdis}{Tmax - Tin}$$

Total distillate produced:

$$Wd = WR \frac{C(Tmax - Tdis)}{L}$$

Number of stages:

$$n = 19 + (6 \times mgd)$$

Area required per unit mass of distillate made per unit time:

$$As = \frac{L}{U} \frac{n}{(Tmax - Tdis)} \ln \left(\frac{n}{n-r} \right)$$

Enthalpy input in the heater:

$$H = \frac{1000}{R} Wd = W_r s \, (Tmax - Tin)$$

Heat balance on brine heater:

$$W_s h_{fs} = (W_f + W_r) \, (Tmax - Tin) \, C$$

W_r - Mass of waste recycled

W_d - Mass of distillate

W_s - Mass of steam

W_f - Mass of feed

Tmax - Maximum temperature of waste

Tin - Temperature of waste before entering heater

Tdis - Temperature of discharged waste

APPENDIX III

Plant Waste Flow - 1893 m^3/day

Input	Flow Rate (10^4 kg/hr)	Temperature (^oC)
Waste Feed	7.9	38
Recycled Waste from Heater	57.0	120
Output		
Distillate	7.11	50
Waste Recycle to Heater	57.0	113
Concentrated Waste	0.79	50
Stean to Heater	0.705	125