

METHANE PRODUCTION FROM HIGH RATE
ANAEROBIC DIGESTION OF HOG AND DAIRY CATTLE MANURE

A Thesis Submitted to
The Faculty of Graduate Studies and Research
University of Manitoba

In Partial Fulfillment of the
Requirements for the Degree
Master of Science

by

Wang Y. FONG

May, 1973



ABSTRACT

Hog and dairy cattle wastes were digested anaerobically in intermittently-mixed units at two retention times, 10 and 15 days. Three levels of organic loading, 0.15, 0.20 and 0.25 pounds of volatile solids per cubic foot digester volume per day were applied. Temperatures for digestion ranged from 32 to 52° C. The treatment response was measured in terms of volatile solids reduction, decrease in chemical oxygen demand (COD) and in gas production. Other parameters measured were volatile acids, gas composition, alkalinity as CaCO₃, organic nitrogen and ammonia-nitrogen. Optimum yields of methane were attained at a loading rate of 0.25 lb.VS/ft³/day at 52° C for both type of wastes. However, for dairy cattle manure, lower yields were noted.

ACKNOWLEDGEMENTS

The writer expresses his thanks to Dr. N.E.R. Campbell, Professor of the Department of Microbiology for his advice during the course of this investigation and in the preparation of this manuscript. Appreciation is also extended to Professor H. Lapp, Department of Agricultural Engineering, for his encouragement and assistance throughout this project.

Special thanks must be accorded to Mr. E.G. Phillips of the Biomass Energy Institute for his kind advice and assistance in collecting raw manure samples.

TABLE OF CONTENTS

	<u>PAGE</u>
Abstract -----	i
Acknowledgement -----	ii
Table of Contents -----	iii
List of Tables -----	v
List of Figures -----	vi
INTRODUCTION -----	1
HISTORICAL -----	4
MATERIALS AND METHODS -----	10
Apparatus -----	10
Collection of raw manure samples -----	10
Hog manure -----	10
Dairy cattle manure -----	10
Operation -----	12
Sampling and analyses -----	13
RESULTS AND DISCUSSION -----	15
Some chemical characteristics of hog and dairy cattle manure -----	15
Some physical attributes of digested manure effluent -----	15
Volatile solids and COD reductions -----	18
pH, alkalinity and volatile acids concentration -----	24

	<u>PAGE</u>
Ammonia and organic nitrogen relationship -----	33
Gas production and methane composition -----	39
General effect of temperature on anaerobic digestion -----	44
Economics of recovery of energy from hog and dairy cattle wastes -----	47
GENERAL CONCLUSION -----	50
REFERENCE -----	53

LIST OF TABLES

<u>TABLE</u>		<u>PAGE</u>
I	Characteristics of hog manure -----	16
II	Characteristics of dairy cattle manure -----	17

LIST OF FIGURES

<u>FIGURES</u>	<u>PAGE</u>
1. Experimental digester system -----	11
2. a. Percent volatile solids reduction as a function of temperature in hog manure digestion -----	20
b. Percent volatile solids reduction as a function of temperature in dairy cattle manure digestion -----	21
3. a. Percent COD reduction as a function of temperature in hog manure digestion -----	22
b. Percent COD reduction as a function of temperature in dairy cattle manure digestion -----	23
4. a. Alkalinity as CaCO_3 as a function of temperature in hog manure digestion -----	26
b. Alkalinity as CaCO_3 as a function of temperature in dairy cattle manure digestion -----	27
5. a. pH as a function of temperature in hog manure digestion -----	28
b. pH as a function of temperature in dairy cattle manure digestion -----	29
6. a. Volatile acids concentration as a function of temperature in hog manure digestion -----	31
b. Volatile acids concentration as a function of temperature in dairy cattle manure digestion -----	32
7. a. $\text{NH}_3\text{-N}$ concentration as a function of temperature in hog manure digestion -----	34
b. $\text{NH}_3\text{-N}$ concentration as a function of temperature in dairy cattle manure digestion -----	35

<u>FIGURES</u>	<u>PAGE</u>
8. a. Organic-N concentration as a function of temperature in hog manure digestion-----	36
b. Organic-N concentraion as a function of temperature in dairy cattle manure digestion -----	37
9. a. Gas production, ml./gm.volatile solids destroyed/ day as a function of temperature in hog manure digestion -----	40
b. Gas production, ml./gm.volatile solids destroyed/ day as a function of temperature in dairy cattle manure digestion -----	41
10.a. Percent methane as a function of temperature in hog manure digestion -----	42
b. Percent methane as a function of temperature in dairy cattle manure digestion -----	43
11.a. Gas production in litres/day/4 litres sludge as a function of temperature in hog manure digestion -----	45
b. Gas production in litres/day/4 litres sludge as a function of temperature in dairy cattle manure digestion -----	46

INTRODUCTION

INTRODUCTION

The explosive growth of power technology in the last two decades accounts for much of the increase in energy consumption. It has been estimated (1) that by the year 2000 the annual United States requirement for energy in all forms is expected to double that required at the present time; the worldwide demand is expected to triple.

Modern society is almost totally dependent on non-renewable fossil fuels in the form of natural gas, petroleum and coal. Various estimates have been made as to the date when known reserves of fossil fuels will be depleted (1). The consensus of opinion seems to be that within 30 years we will have consumed almost all fuels of this type. Two possible alternatives to fossil fuels are indicated, nuclear energy on one hand and, secondary, a more effective use of renewable biomass as an energy source.

The development of power from nuclear processes is proceeding at a much slower rate than was anticipated ten years ago. Prospects for power generated by nuclear fission reactions have encountered considerable opposition from environmentalists who are concerned with the proper disposal of highly radioactive wastes inherent in this mode of operation. Power generated by fusion reactions would provide a much cleaner method but current progress in controlled fusion technology indicates a delay of

20 to 30 years before this form of power generation is practical. Besides, nuclear processes are also dependent upon a limited quantity of non-renewable resources such as uranium, thorium and plutonium which can also be depleted over certain period of years.

The United States is currently experiencing an energy crisis and, being largely dependent on imported fuels, i.e. oil and natural gas, is entering a critical period. Recently, interest has turned toward the use of renewable biomass as a fuel source. The list of materials normally considered as waste products that can be converted to or consumed as fuels is very broad, ranging from sawdust wastes from lumber mill operations to livestock residues from agricultural and food industry operations. It is the latter category of livestock wastes with which this dissertation is concerned.

At the present time, livestock waste production in the United States alone has been estimated at 1.7 billion tons annually (2). This immense quantity has created an important and costly problem in terms of handling, treatment and disposal. Current practice for disposal of this material usually involves its dispersal, without further treatment, as a fertilizer over agricultural lands. A considerable portion of the constituent hydrogen in these wastes is lost to the atmosphere through microbial decomposition and assimilation activities in the amended soils. This represents a loss of energy which, in light of the continental energy deficit position can ill be afforded.

For a considerable period of time in Europe and in Asia animal wastes have been treated on a small scale by high rate anaerobic digestion as a means of recovering a useful fuel, methane, and for conserving if not enhancing the subsequent fertilizer value. It has been estimated that one ton of farm animal wastes yields approximately 50 to 60 cubic meters of gas with a 55 percent methane content at a caloric value equivalent to 10 imperial gallons of gasoline (3). With the annual amount of animal wastes currently disposed of in the United States, approximately 17 billion gallons of gasoline or its equivalent in heating fuels could be recovered. In light of the energy imbalance, this figure is not insignificant and methane gas produced in this way could form an important alternative to reduce the depletion rate of current reserves of fossil fuels until such time as controlled fusion energy production becomes practical. Concomittant with methane production by anaerobic digestion, the sludge residues remaining retain their nutritive values as fertilizers and, in some cases, the conversion of organic nitrogen to an inorganic form as ammonia improves the residues in terms of their fertilizer efficiency.

HISTORICAL

HISTORICAL

Methane production by the anaerobic digestion of farm animal wastes and compost materials has been practiced for a number of years. As early as 1895, a Donald Cameron in England, collected methane gas produced by his 'carefully designed' septic tank and used it for street lighting in the vicinity of the plant in the town of Exeter (4).

Lord Lveagh, in 1919-1920, on his personal interest and initiative, built five digesters in several different localities in England for the production of methane gas (5). Briefly, these primitive digesters each consisted of a strong metal cyclinder, 28 feet high and 16 feet in diameter, with a heating device in the bottom, a paraffin engine for the mixing of the sludge and a gas collection chamber. The gas produced was piped to nearby dwellings for use in cooking. The digester built at Pyrford, England in 1929 operated for a number of years with a minimum of attention. When fully loaded, the digester held 165 cart-loads of manure suspended in 3000 gallons of water. Daily gas production was approximately 450 cubic feet with a load retention time of six months for each 16 cart-loads of fresh manure. This was believed to be one of the first domestic methane producing plants in the world but because little was understood at that time about the optimum conditions necessary for efficient methane production,

the maximum yields of combustible gas theoretically possible for the loading rates used were never attained.

Apart from its use as a cooking fuel, methane gas produced by anaerobic fermentation has also been used as a fuel for heating farm buildings, for drying grain crops and for the production of electricity by thermal generating plants (6). Schmidt et al (7) in 1951 used digester methane as a fuel for farm tractors and other equipment. In this case, the gases evolved were stored under 5000 psi in large tanks from which supplies were provided as required to 2840 psi pressurized fuel tanks fitted to each tractor. More recently in England (8) digester gas produced from chicken manure has been used successfully as a fuel for automobiles.

Operation of single farm-scale digesters is underway in India and in Taiwan. In India, the Gobar Gas Research Station in Ajitmal, Etawah (U.P.) has designed hog and poultry manure digesters suitable for operation under Indian climatic conditions (9). In Taiwan under the supervision of the Chinese-American Joint Commission on Rural Re-construction, 6000 farm-waste digesters have been constructed (10). These produce methane which is used primarily as a fuel for cooking. In view of the moderate oceanic climate of Taiwan, residual heat lost during the cooking process provides for heating the small dwellings. After fermentation, the residual sludge provides an ideal substrate for the growth of a Chlorella

species, a unicellular algae rich in proteins and vitamins. The algae crop is used as a supplement to feed rations for poultry and hogs. It has been reported that such practices provide savings of up to \$1000 NT. per month in the farm operation and, moreover, the market quality of the poultry and hogs has been improved.

In the light of the successful Taiwan operation, several advantages of anaerobic fermentation of farm wastes are obvious. According to Schmidt (7) these are: one, mechanization of manure handling is encouraged; two, weed seeds and animal parasites are destroyed during digestion; and three, loss of nitrogen and fertilizer values from raw manure is markedly reduced. These advantages, together with the methane produced as a fuel, are usually sufficient to provide for the recovery of total capital costs of the digester installation within 10 years. Taiganides et al (1962) estimates that a hog producer averaging about 10000 hogs per year can increase his profit significantly by the utilization of combustible gases produced by manure digestion while at the same time largely eliminating the odour problem associated with operations of this kind (11).

Despite the increasing use of anaerobic digesters, the fundamental microbiology of the process is still not well understood. McKinney (12) has commented that anaerobic digestion is 'the enchanted wilderness of sanitary engineering'.

Buswell (13) and Barker (14) in the mid-1930's described the process as a two-stage fermentation. The first stage consists of hydrolyses of organic materials with the production of volatile acids and alcohols and is catalysed by a heterogeneous group of microorganisms that are largely anaerobic or micro-aerophilic saprophytes. This stage is referred to as liquefaction and is characterized by the rapid proliferation of a mixed microorganic population relatively insensitive to changes in environmental and chemical conditions (15). O'Shaughnessy in 1914, reported that the acid-formers in sludge fell into several groups that included coliforms, proteus species, denitrifiers, lipolytic and cellulolytic bacteria (16) while Hotchkiss in 1924, indicated the presence of denitrifying, albumen digesting and H₂S producing bacteria in sludge (17). Ruchhoft et al (18) indicated the presence of different proteolytic bacteria, such as gelatin liquefiers and protein digesters. Hungate in 1950 isolated strains of cellulolytic bacteria in pure culture that were present in anaerobic digesters (19). In stage two which is referred to as gasification, volatile acids produced during the primary stage are converted to methane and carbon dioxide by a group of highly oxygen-sensitive methanogenic bacteria (20). According to Cookson et al (21), the methanogenic bacteria belonged to the genera Methanobacterium (22,23), Methanococcus (24), Methanobacillus (25), Methanosarcina (26), and Clostridium (27).

These organisms are slow-growing and are extremely sensitive to changes in pH and temperature (28). According to McCarty (29), the methanogenic population represents the key organisms in the overall digestion process, i.e. the gasification stage is the rate-limiting one in the process. Under unbalanced digester conditions, methane producing organisms which use carbon dioxide as a terminal electron acceptor, do not remove the volatile acids as quickly as they are formed by the primary population group. This acid concentration build-up may take place very rapidly and leads to a precipitous drop in pH with a consequent failure of digester operation (30).

Laboratory investigations of the anaerobic digestion of livestock wastes seem to have begun only in the last decade. Taiganides et al (11) in laboratory studies concluded that at a temperature of 35°C with continuous mixing of reactor contents, hog manure was satisfactorily digested at a loading rate of 0.20 lbs volatile solids¹ (VS) per cubic foot digester.

¹The volatile solids measurement is conventionally adopted by sanitary engineers for the determination of combustible solid materials in anaerobic sludge and waste water. It is done by igniting the sample to a constant weight at 550°C after evaporation using an electric muffle furnace. The loss of weight is reported in terms of pounds of volatile solids per cubic foot of the sample.

capacity per day. The average yield of gas per pound of volatile solids in the digesters ranged from 7.8 to 10.3 cu. ft. per day with a composition of methane (59%), carbon dioxide (40%) and trace amounts of hydrogen, hydrogen sulfide and oxygen.

Similar investigations have been carried out at the Rowett Agriculture Research Institute, Aberdeenshire, Scotland where Hobson (31) has operated hog-manure digesters at 35°C with a loading rate of 0.216 lbs. of volatile solids per cubic foot per day using a retention time of 14 days. These experiments indicated a reduction of 56% in biological oxygen demand (BOD), a 37% reduction in chemical oxygen demand (COD) and a reduction in total solids in the influent organic material of 26%. Under these conditions, the digester gave no indication of failure and it may be concluded that maximum loading rates had not been attained. The effluent produced is more stable and less odorous than the original material and could be discharged to domestic sewers. Gas production showed a 70% methane content with the remainder being largely carbon dioxide.

METHODS AND MATERIALS

METHODS AND MATERIALS

APPARATUS

Anaerobic digestion of hog and dairy cattle manure was investigated using six laboratory-scale digester units each consisting of three 4.5 litre bottles as shown in Fig. 1. These were maintained in a deep water bath whose temperature was controlled by a thermoregulator.

Gases evolved in the digester unit were collected in the gas collection bottle by displacement of water which, in turn, was collected in the reservoir. Prior to use, each bottle was calibrated and marked as to volume.

COLLECTION OF RAW MANURE SAMPLES

HOG MANURE

Raw hog manure was collected monthly from the Glenlea Research Station, University of Manitoba, which is located some 15 miles south of Winnipeg, Manitoba on Highway 75. To ensure representative samples, the manure was well mixed prior to its removal by pump from the holding pit. The collected waste was diluted with tap water to yield a slurry of approximately ten per cent total solids by weight.

DAIRY CATTLE MANURE

About 120 pounds of dairy cattle manure was also

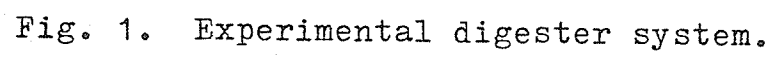
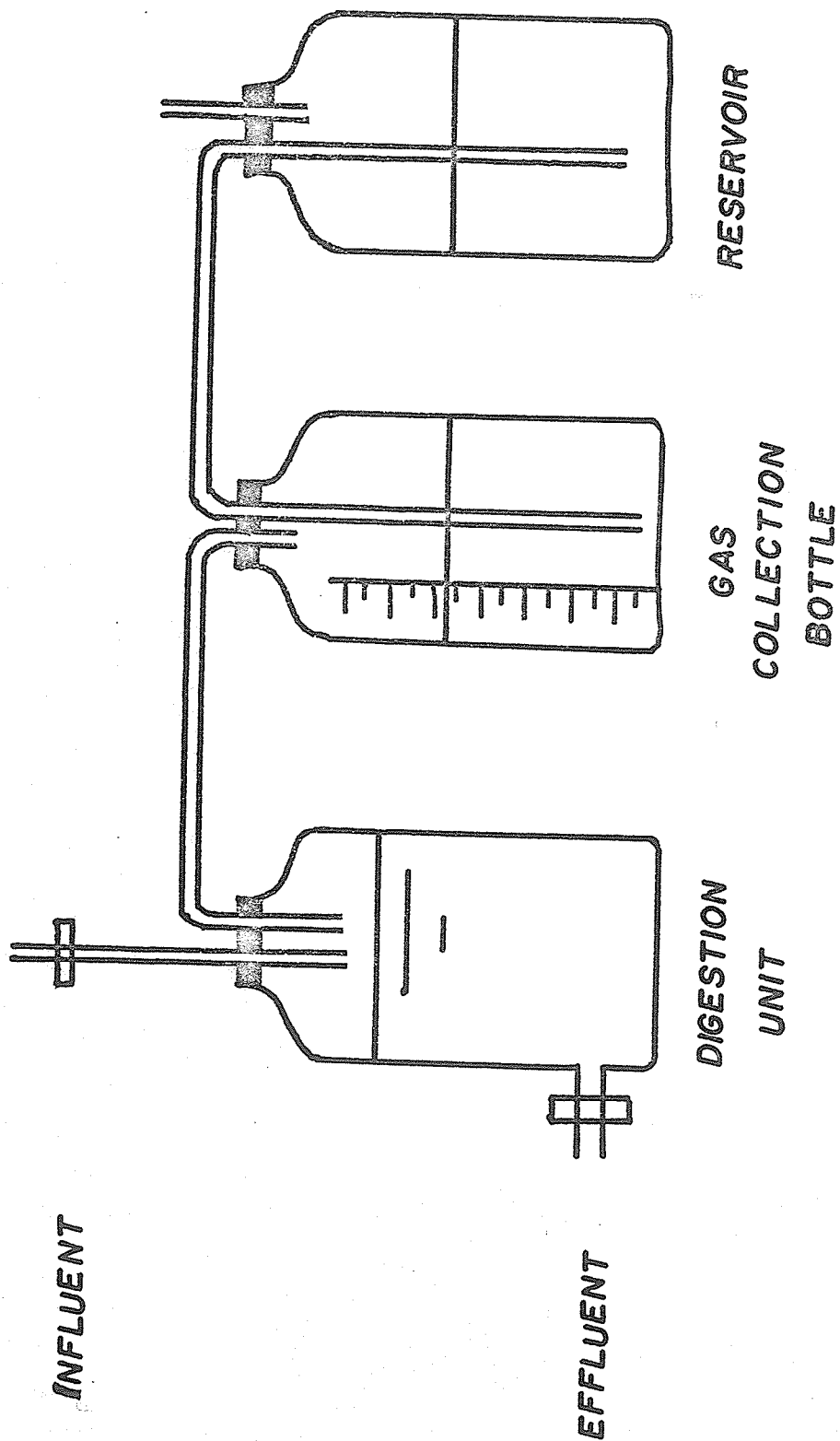


Fig. 1. Experimental digester system.



obtained from the Glenlea Research Station. The waste was also diluted with tap water to give a slurry having a total solids content of 10 per cent by weight.

Volatile solids content of each manure type was determined in triplicate according to the Standard Methods for the Examination of Water and Waste Water (32). Then, the waste slurries were stored at 4°C until required in five-gallon polypropylene containers fitted with screw-caps.

OPERATION

Initially, the digesters were seeded with approximately four litres of digested domestic sludge obtained from the No. 10 digester at the North End Waste Disposal Treatment Plant, City of Winnipeg. Digesters were acclimated at a constant temperature of 32°C until gas production, pH and alkalinity (as calcium carbonate) were constant.

Loading rates of 0.15, 0.20, 0.25 lb./VS/ft³/day and retention times of 10 and 15 days were arbitrarily selected for this study. The four litre capacity digesters were fed daily with a mixture of tap water and manure in proportion to yield the desired loading rates and retention times. An equal volume of digested manure was removed prior to each feeding. Because of the large, settleable solids content of the wastes under study, homogenization by means of a blender was necessary before the materials were fed into the

digesters.

Temperatures selected for the digestion operation covered the upper level of the mesophilic range and the lower level of the thermophilic range. These were arbitrarily selected as 32, 37, 42, 47 and 52°C. The initial temperature used in the study was 32°C.

Once equilibrium was established after each loading, analyses were carried out at regular intervals. Equilibrium conditions were based mainly on the constancy of gas production (as ml. gas/gm. VS destroyed/day) and on the pH value measured. When an equilibrium state was reached, the digester temperature was raised to the next highest value selected and analyses were again carried out once equilibrium at the new temperature had been reached.

Measurements of pH, COD, $\text{NH}_3\text{-N}$, organic-N, volatile acids and gas composition were routinely taken during the course of this study. The total daily volume of gases evolved was also recorded. Digester contents were mixed mechanically three times daily on week days and once on Sundays. Prior to each sampling, digester contents were thoroughly mixed to provide homogeneity of sample.

SAMPLING AND ANALYSIS

Obtaining representative samples of the digester

material required careful sampling techniques. Because of the extreme heterogeneity of suspended particles in terms of size, a thorough mixing of the contents was imperative. Uniformity of sample proved to be quite difficult to attain and some of the variations noted in the analyses data likely reflected this difficulty.

The various chemical analyses noted above were carried out in accordance with the procedure outlined in Standard Methods for the Examination of Water and Waste Water (32). Since the digested manure may be subjected to further changes, i.e. a gain or loss of carbon dioxide when exposed to the air during sampling, pH, alkalinity as calcium carbonate and free ammonia content were measured without delay.

Methane content of the evolved gas was determined using a Pye Series 104 chromatograph fitted with a flame ionization detector. Separation and identification of methane was provided by a 3/16" eight foot stainless steel column packed with Porapak R. Nitrogen at a flow rate of 20 ml./min. was used as the carrier gas, column temperature was maintained at 27°C.

RESULTS AND DISCUSSION

RESULTS AND DISCUSSION

SOME CHEMICAL CHARACTERISTICS OF HOG AND DAIRY CATTLE MANURE

Some pertinent chemical characteristics of hog and dairy cattle manure are summarized in Tables I and II respectively. When volatile solids concentrations in both type of wastes are equal, they exhibit similar chemical oxygen demand, percentage of volatile solids and alkalinity. Hog manure, however, contains approximately six times the amount of ammonia nitrogen and twice the organic nitrogen content as compared to dairy cattle manure.

Differences particularly in ammonia nitrogen are quite significant in terms of digester operation. High ammonia values tend to create an unfavorable alkaline environment and, if as a consequence of primary fermentation activity, ammonia values tend to increase even further, the failure of the secondary phase methanogenic process is likely (33,34,35).

SOME PHYSICAL ATTRIBUTES OF DIGESTED MANURE EFFLUENT

Properly digested hog and dairy cattle manure effluents were black, thick free-flowing liquids with little offensive odour. As such, they can be disposed of by field application as a fertilizer without creating an environmental odour nuisance or may be stored temporarily in farm buildings without the gas problems often associated with the storage

of fresh manure (36).

VOLATILE SOLIDS AND COD REDUCTIONS

The percent reduction of volatile solids in hog wastes was good when compared to that obtained in digestion of domestic wastes (37). The highest reduction observed, 76 percent, was obtained at an operating temperature of 52°C with a loading rate of 0.15 lb. VS/ft³/day during a 15-day retention period as noted in Fig. 2a. Similar data have also been reported by Gramms et al (38). For dairy cattle waste, the percent reduction in volatile solids was considerably lower, 28.6 percent, for the same temperature, loading rate and retention time, Fig. 2b.

These data suggest that rates of volatile solids destruction are critically dependent on the rate of digester loading. For hog manure, for example, an increase in loading rate of 0.1 lb.VS/ft³/day effectively decreased volatile solids reduction at 37°C from 44.8 percent to 40.3 percent in 15 days. Similar decrease in volatile solids reduction with increasing loading rates were evident for cattle manure digestion as noted in Fig. 2b.

Increasing the retention time in the digester gave a corresponding increase in volatile solids destruction as might be expected. An increase of five days, for example, resulted in volatile solids destruction some 10 percent greater

in the case of hog wastes and a 2 percent increase in the case of cattle manure. Again, these findings support the observations reported by Gramms et al (38).

At retention times of 10 and 15 days, there was a substantial increase in volatile solid destruction for both hog and dairy cattle manure as the digester temperature was increased from 32°C to 42°C. With a further temperature increase to 47°C a slight decrease in volatile acid destruction was noted but as the digester temperature was increased still further to 52°C a marked increase in volatile solids destruction was observed. This phenomenon has been reported by other investigators (39, 40, 41, 42, 43) and will be discussed in detail in the latter part of this section.

The relationship of temperature to the reduction of COD at different loading rates and retention times for both types of wastes are shown in Fig. 3a and 3b. Again, a marked difference is apparent between hog and dairy cattle wastes as was the case for volatile solids reduction. Hog wastes digestion at 52°C gave the maximum COD reduction of 60 percent at a loading rate of 0.15 lb.VS/ft³/day for a 15 day retention time whereas with dairy cattle waste under these conditions reached a COD reduction maximum of 20.2 percent. Similar results have been reported by Hobson (31). In both cases, COD reduction increased with retention time and decreased with

Fig. 2a. Percent volatile solids reduction as a function of temperature in hog manure digestion, Solid lines indicate 15-day retention time; dotted lines indicate 10-day retention time.

Units 1 and 2 have $0.15 \text{ lb.VS/ft}^3/\text{day}$ loading rate.

Units 3 and 4 have $0.20 \text{ lb.VS/ft}^3/\text{day}$ loading rate.

Units 5 and 6 have $0.25 \text{ lb.VS/ft}^3/\text{day}$ loading rate.

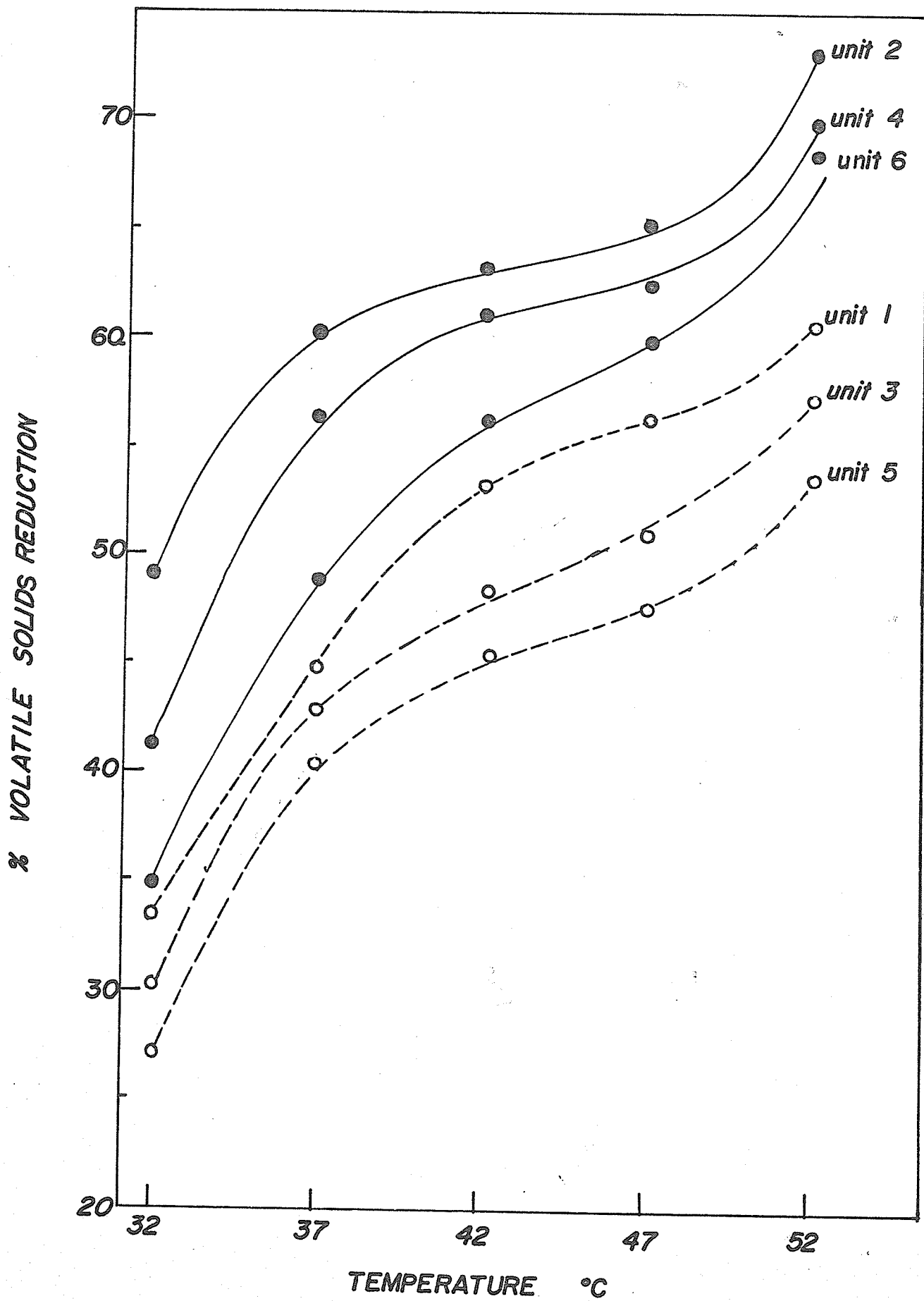


Fig. 2b. Percent volatile solids reduction as a function of temperature in dairy cattle manure digestion. Solid lines indicate 15-day retention time; dotted lines indicate 10-day retention time. Units 1 and 2 have $0.15 \text{ lb.VS/ft}^3/\text{day}$ loading rate. Units 3 and 4 have $0.20 \text{ lb.VS/ft}^3/\text{day}$ loading rate. Units 5 and 6 have $0.25 \text{ lb.VS/ft}^3/\text{day}$ loading rate.

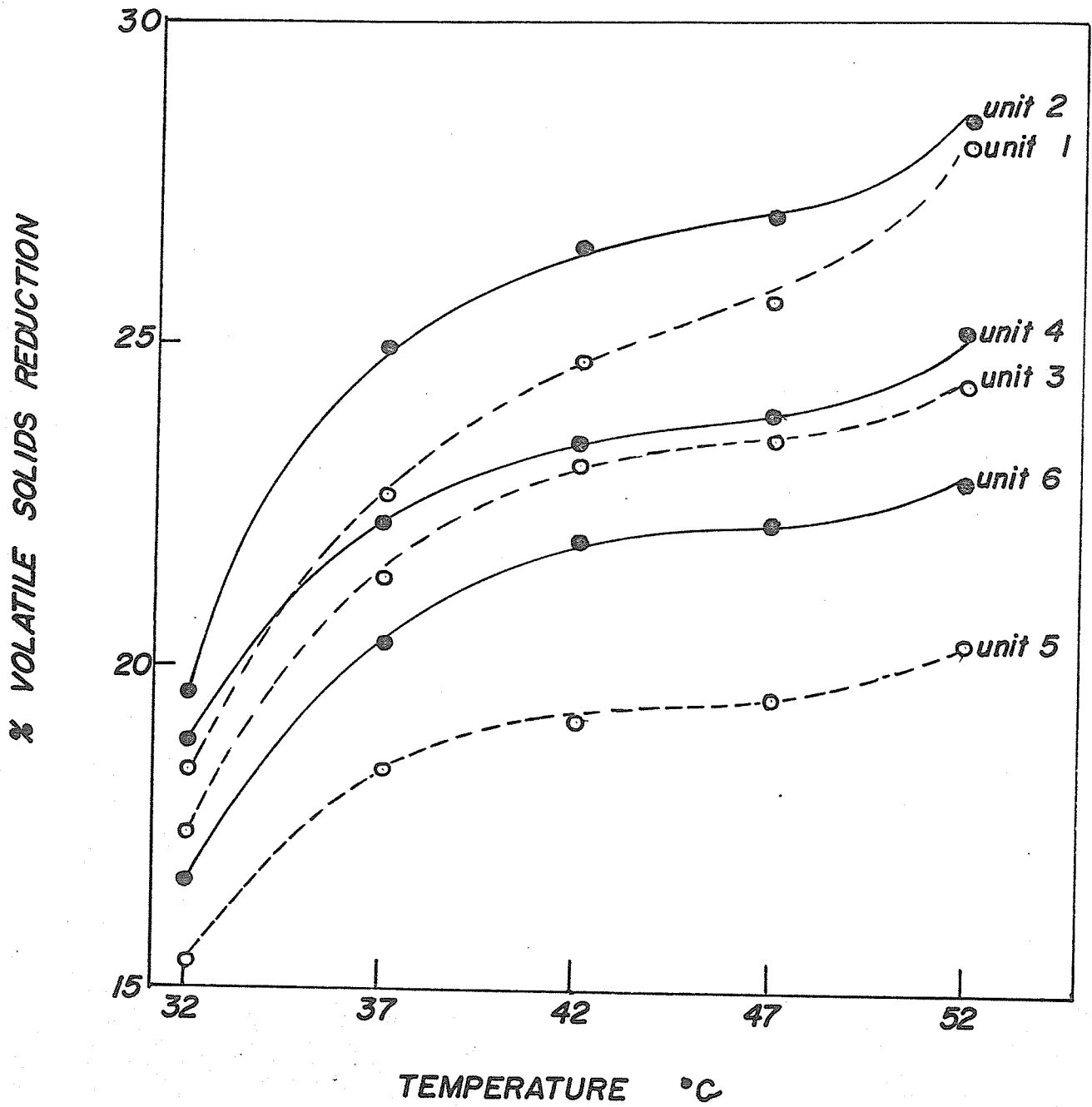


Fig. 3a. Percent COD reduction as a function of temperature in hog manure digestion. Solid lines indicate 15-day retention time; dotted lines indicate 10-day retention time.

Units 1 and 2 have $0.15 \text{ lb.VS/ft}^3/\text{day}$ loading rate.

Units 3 and 4 have $0.20 \text{ lb.VS/ft}^3/\text{day}$ loading rate.

Units 5 and 6 have $0.25 \text{ lb.VS/ft}^3/\text{day}$ loading rate.

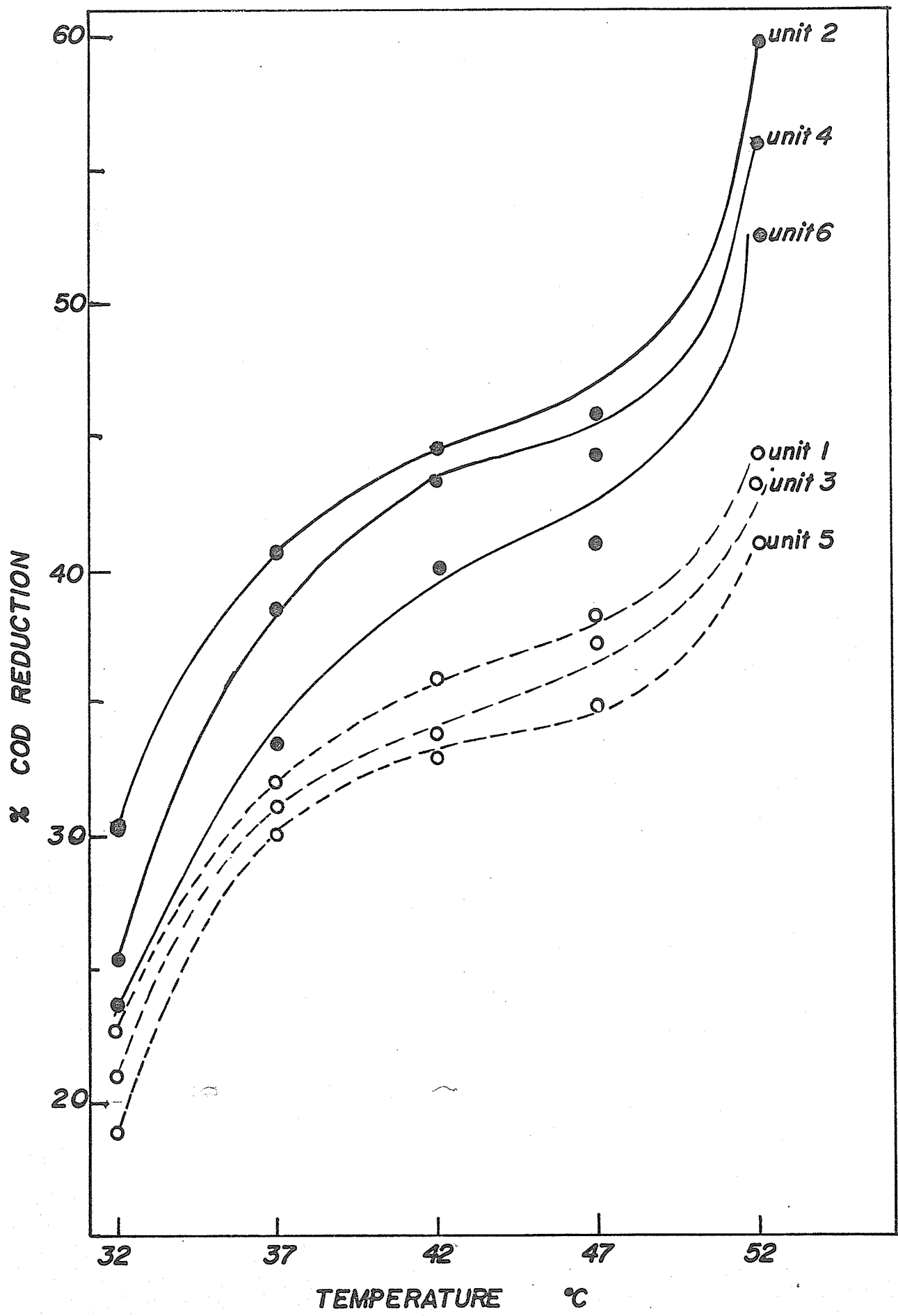
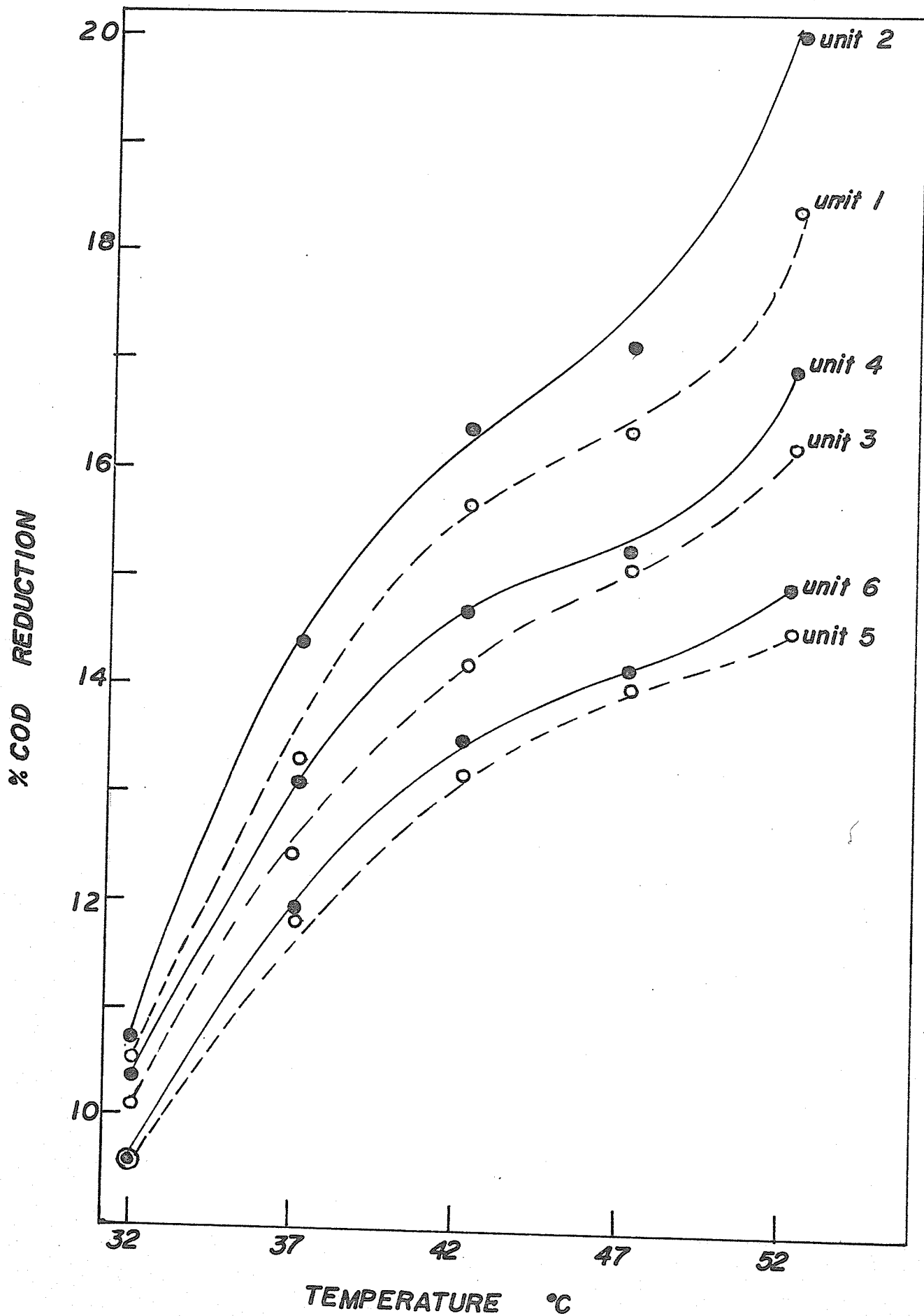


Fig. 3b. Percent COD reduction as a function of temperature in dairy cattle manure digestion. Solid lines indicate 15-day retention time; dotted lines indicate 10-day retention time. Units 1 and 2 have 0.15 lb.VS/ft³/day loading rate. Units 3 and 4 have 0.20 lb.VS/ft³/day loading rate. Units 5 and 6 have 0.25 lb.VS/ft³/day loading rate.



increasing loading rates. Retardation of COD reduction was noted at 47°C.

pH, ALKALINITY AND VOLATILE ACIDS CONCENTRATION

Alkalinity, pH and volatile acids are accepted parameters for practical control of the anaerobic digestion (32, 44). These three factors however are interdependent and their effect on the process of anaerobic digestion will be discussed together. Usually, the pH of the contents of a digester depends on the relationship between the volatile acid and alkalinity. The main volatile acids produced during digestion are acetic, propionic and butyric acids (30) which provide the hydrogen ions for the reduction of carbon dioxide in the system. On the other hand, the alkalinity of an anaerobic digester is a measure of the buffering capacity of the digester contents. The principal form of alkalinity in an anaerobic digester of this type is bicarbonate. Bicarbonate alkalinity is developed in these digesters by the reaction of ammonia with carbon dioxide and water to form ammonium bicarbonate (33, 34, 35, 45). This natural production of alkalinity provides an essential buffer which, in most digestion systems, holds the pH in the desired range of 6.8 to 7.2. A high alkalinity is therefore considered to be an

indication that the system is safeguarded against pH fluctuation while a low alkalinity suggests that sudden increase in volatile acids may have lowered the pH to such an extent that biological activity is impaired (30). Mueller et al (46) and Pohland (47) emphasized the need for a balance between alkalinity and volatile acids concentration for normal waste digestion and implied that variations in pH occurred only after the volatile acids-alkalinity balance had been disrupted. It has been stated recently that if the ratio of volatile acid (expressed as mg./litre acetic acid) to total alkalinity (expressed as mg./litre CaCO_3) was lower than 0.8, unbalanced conditions in a digester are indicated (44).

The relationship of alkalinity to temperature at various loading rates and retention times for hog and dairy cattle wastes digestion are illustrated in Figs. 4a and 4b respectively. As shown in these figures, alkalinity increased with increasing loading rates, retention times and temperature. In dairy cattle manure digestion, for example, at a 10 day retention time with a temperature of 32°C , alkalinity increased by approximately 1500 mg./litre as CaCO_3 when the loading rate changed from 0.15 to 0.25 lb.VS/ft³/day (Fig. 4b). As the temperature was raised from 32 to 52°C , alkalinity increased almost by a factor of 2.5. An increase in retention time by 5 days also increased the alkalinity by an average of 300 mg./litre as CaCO_3 .

The relationship between alkalinity and loading is of great importance to the operation of these digesters. Since a substantial portion of this alkalinity is in the form of

Fig. 4a. Alkalinity as CaCO_3 as a function of temperature in hog manure digestion. Solid lines indicate 15-day retention time; dotted lines indicate 10-day retention time.

Units 1 and 2 have $0.15 \text{ lb.VS/ft}^3/\text{day}$ loading rate.

Units 3 and 4 have $0.20 \text{ lb.VS/ft}^3/\text{day}$ loading rate.

Units 5 and 6 have $0.25 \text{ lb.VS/ft}^3/\text{day}$ loading rate.

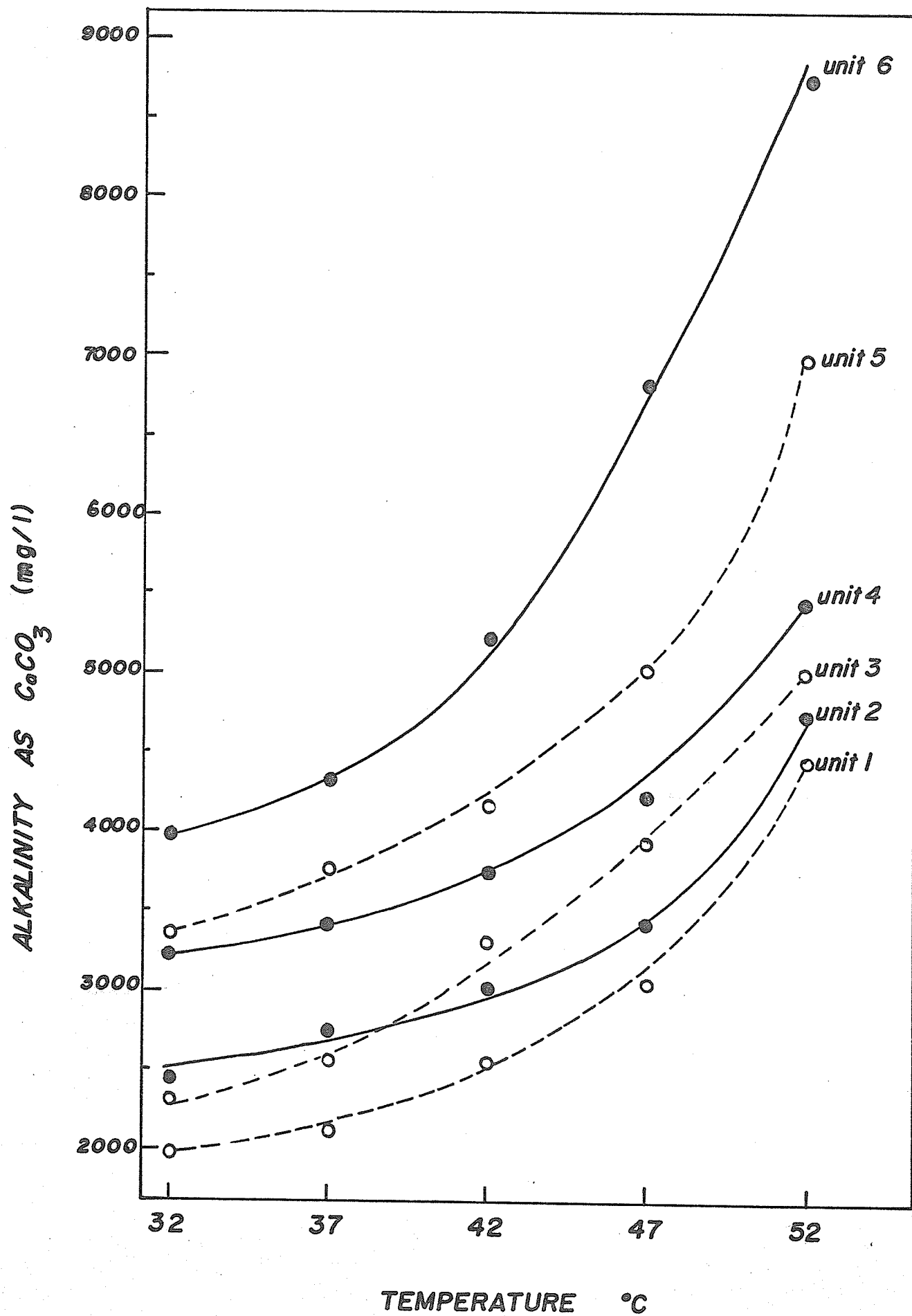


Fig. 4b. Alkalinity as CaCO_3 as a function of temperature in dairy cattle manure digestion. Solid lines indicate 15-day retention time; dotted lines indicate 10-day retention time.

Units 1 and 2 have $0.15 \text{ lb.VS/ft}^3/\text{day}$ loading rate.

Units 3 and 4 have $0.20 \text{ lb.VS/ft}^3/\text{day}$ loading rate.

Units 5 and 6 have $0.25 \text{ lb.VS/ft}^3/\text{day}$ loading rate.

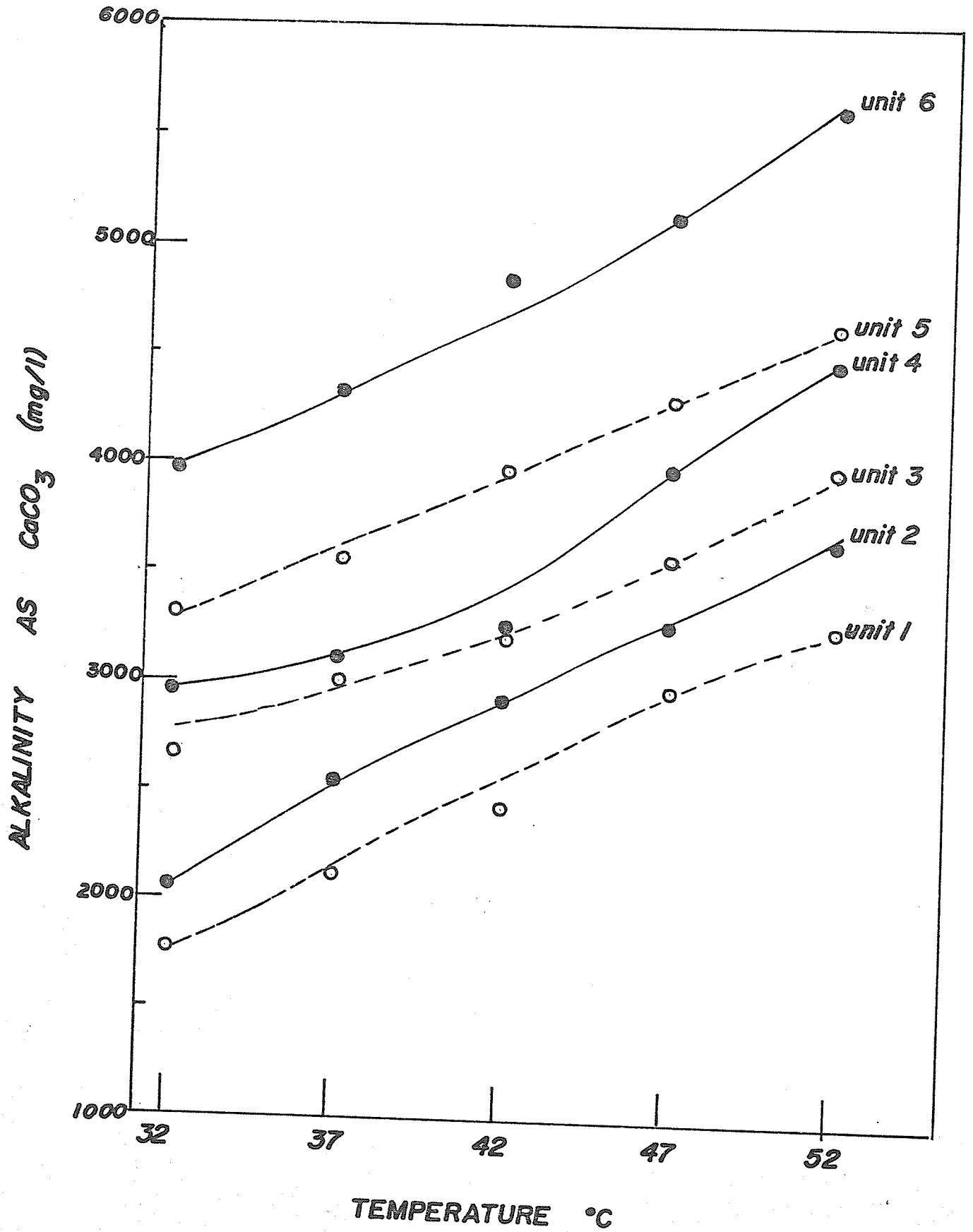


Fig. 5a. pH as a function of temperature in hog manure digestion. Solid lines indicate 15-day retention time; dotted lines indicate 10-day retention time. Units 1 and 2 have 0.15 lb.VS/ft³/day loading rate. Units 3 and 4 have 0.20 lb.VS/ft³/day loading rate. Units 5 and 6 have 0.25 lb.VS/ft³/day loading rate.

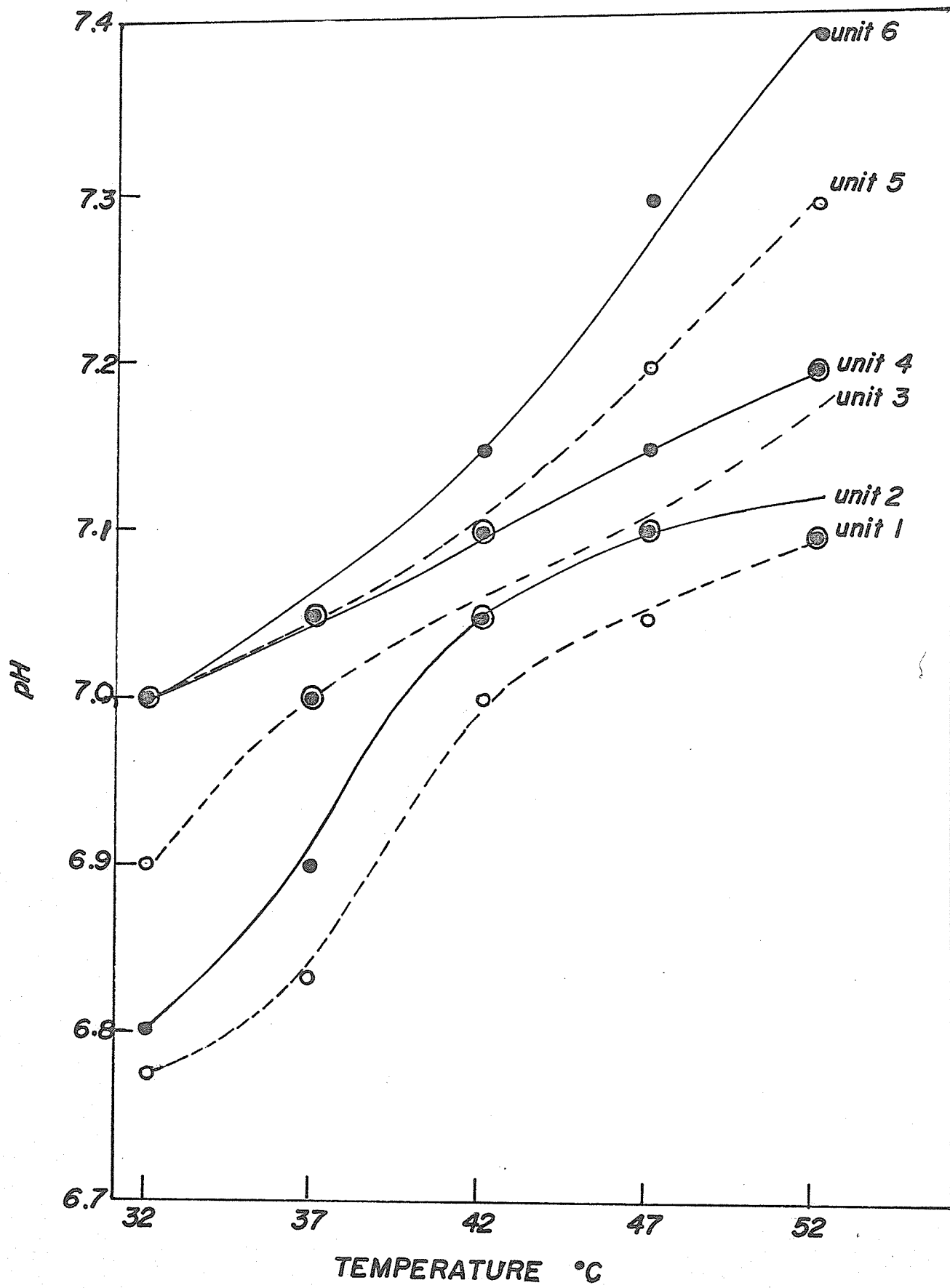
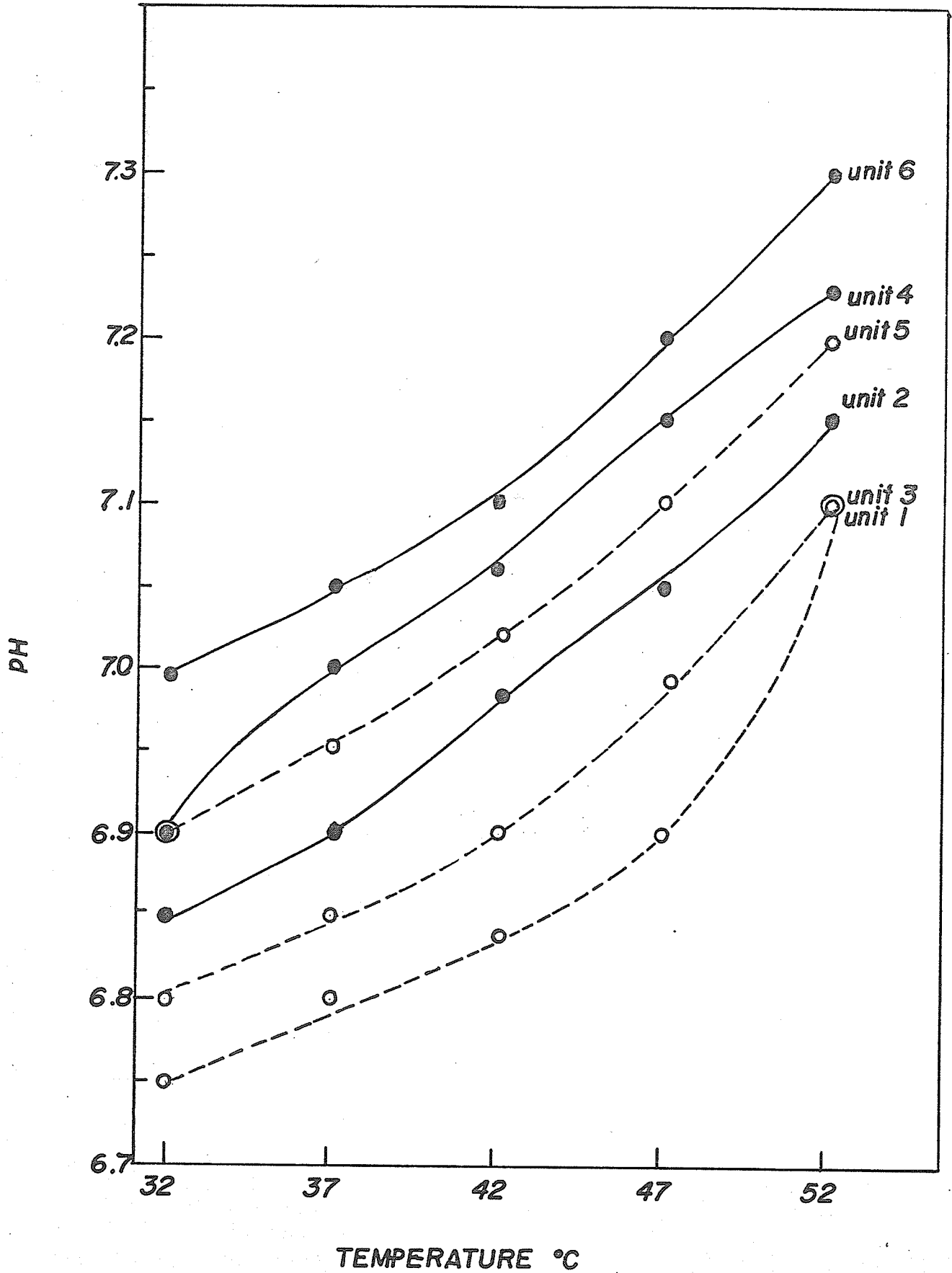


Fig. 5b. pH as a function of temperature in dairy cattle manure digestion. Solid lines indicate 15-day retention time; dotted lines indicate 10-day retention time.

Units 1 and 2 have $0.15 \text{ lb.VS/ft}^3/\text{day}$ loading rate.

Units 3 and 4 have $0.20 \text{ lb.VS/ft}^3/\text{day}$ loading rate.

Units 5 and 6 have $0.25 \text{ lb.VS/ft}^3/\text{day}$ loading rate.



ammonium bicarbonate (33, 34, 35), there is a potential problem of ammonium toxicity. McCarty and McKinney (35) reported that the free ammonia concentration increased with increasing pH and that 150 mg./litre of free ammonia produced a toxic condition in the digesters. This did not occur until the pH reached values of pH 7.6. However, the ammonia is related not only to pH, but also to the total ammonium ion concentration. Therefore, when digesting wastes at high loading rates and long retention time, it is possible to build up ammonia concentration to toxic levels.

Along with the increase of alkalinity which parallels increases in loading rates, retention times, and temperatures, the pH of the digesters increases correspondingly, Fig.5a and 5b. The pH of the hog waste digesters ranged from 6.75 to 7.40 (Fig. 5a) whereas the dairy cattle digesters had pH values ranging from 6.75 to 7.3 at various loading rates, retention times and temperatures. In general, the pH of both waste digesters was under control and was within the range recommended for optimal digestion of domestic sludge (37).

The hog waste digester had volatile acids content ranging from 204 mg./litre at 32°C to 1450 mg./litre at 52°C (Fig. 6a). It was indicated clearly from the graphs that retention time had a very large effect upon the volatile acids concentration in the digesters. For instance, at 42°C, Unit 5 with a 10 day retention time had a volatile acids concentration of 975 mg./litre as CH_3COOH . However, Unit 6 which had the same loading rate as Unit 5, with 15-day retention

Fig. 6a. Volatile acids concentration as a function of temperature in hog manure digestion. Solid lines indicate 15-day retention time; dotted lines indicate 10-day retention time.

Units 1 and 2 have 0.15 lb.VS/ft /day loading rate.

Units 3 and 4 have 0.20 lb.VS/ft /day loading rate.

Units 5 and 6 have 0.25 lb.VS/ft /day loading rate.

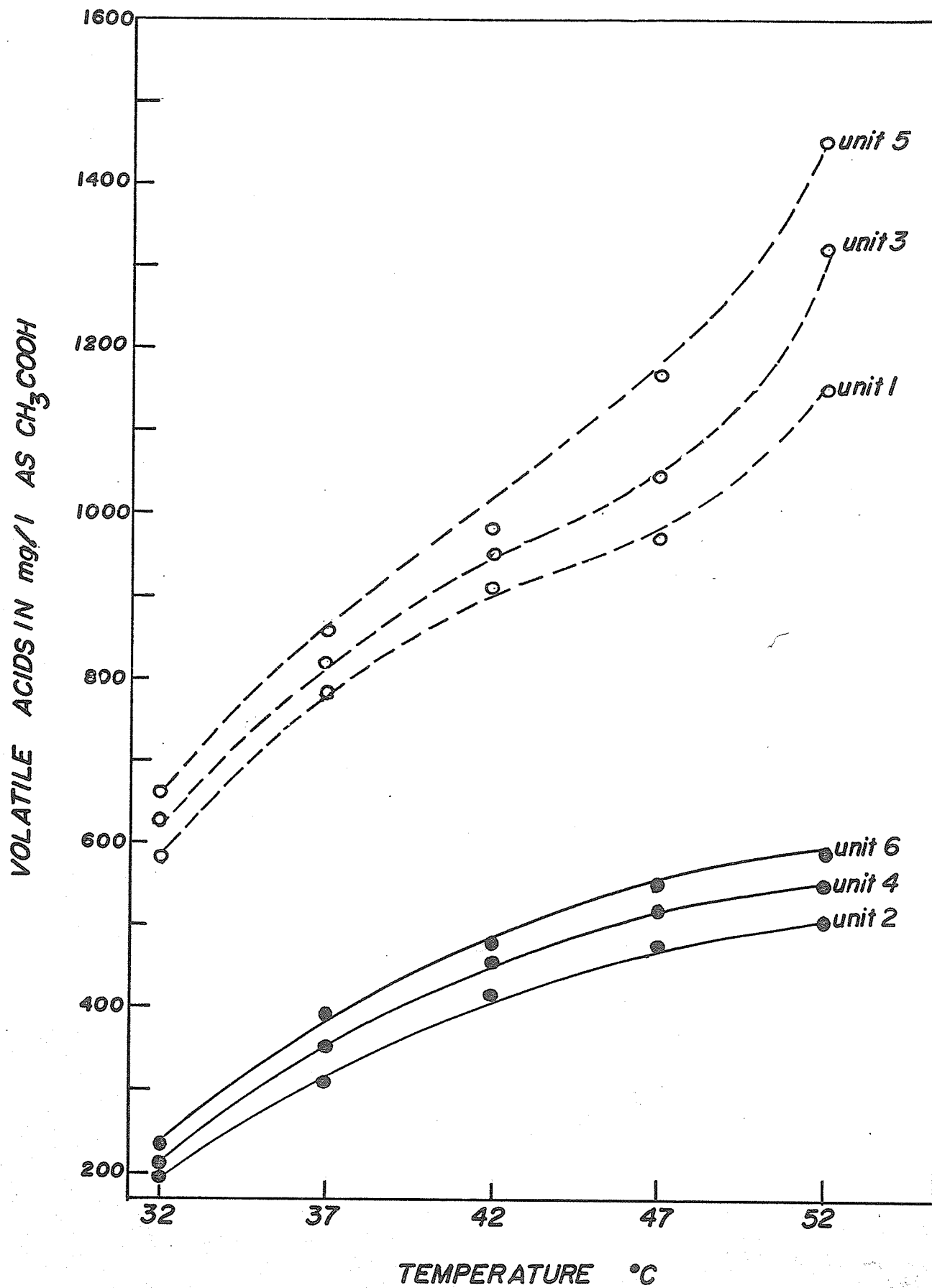
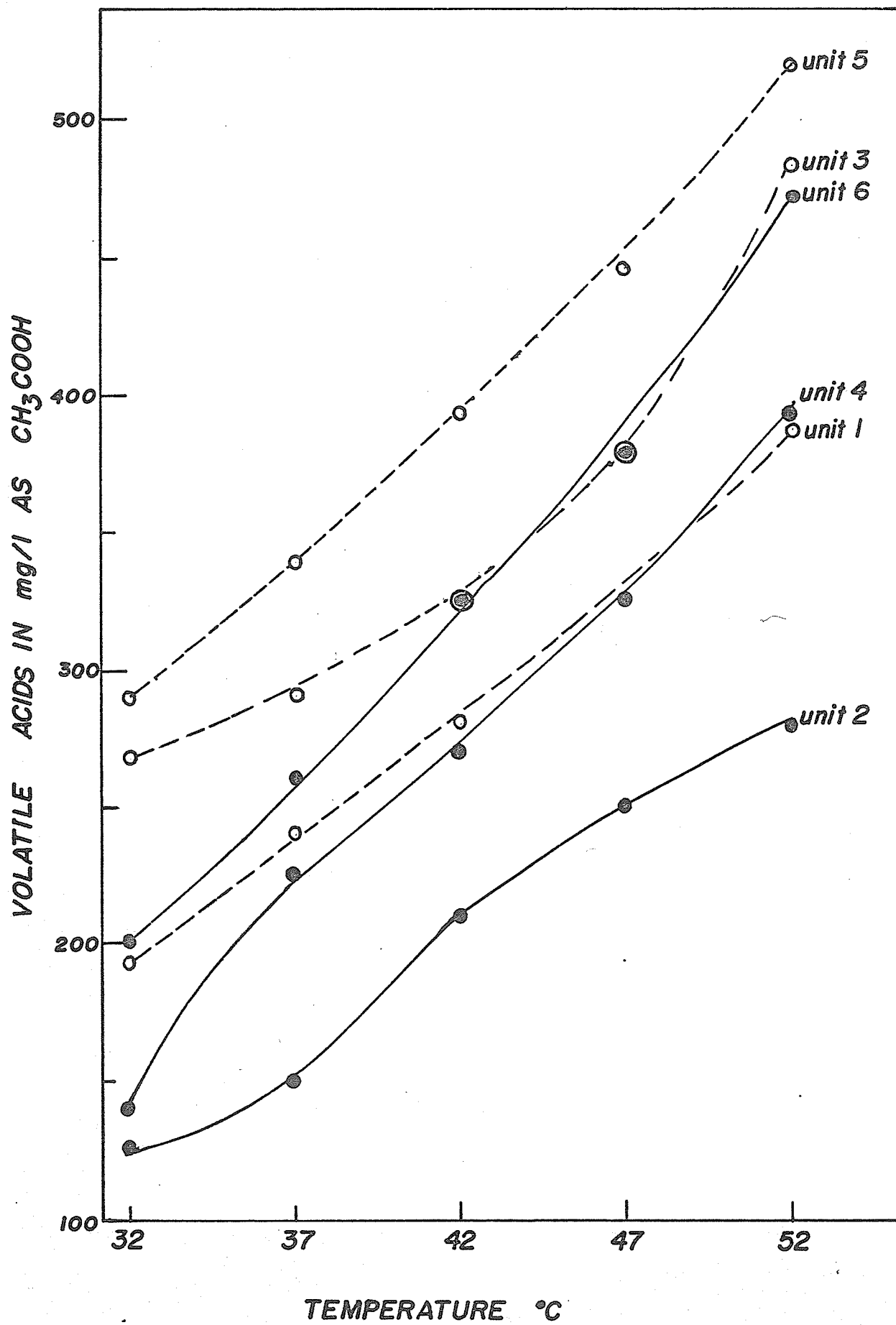


Fig. 6b. Volatile acids concentration as a function of temperature in dairy cattle manure digestion. Solid lines indicate 15-day retention time; dotted lines indicate 10-day retention time. Units 1 and 2 have $0.15 \text{ lb. VS/ft}^3/\text{day}$ loading rate. Units 3 and 4 have $0.20 \text{ lb. VS/ft}^3/\text{day}$ loading rate. Units 5 and 6 have $0.25 \text{ lb. VS/ft}^3/\text{day}$ loading rate.



time, had a volatile acids concentration of 470 mg./litre, indicating that an increase of 5 days in retention time reduced the volatile acids concentration approximately twice.

Results showed that volatile acids concentrations increased with increasing loading rate and temperature. A similar phenomenon had been reported by other investigators (29, 45). The explanation suggested is that at high loading rates and temperatures, the acid formers which are more numerous and more metabolically active than the methane formers, were producing volatile acids faster than the methane formers could convert these intermediates to methane and carbon dioxide. This accumulation of volatile acids may lead eventually to a drop in pH to such an extent that the digestion process is impaired. Further, while the existence of excessive amounts of volatile acids in the digester is, in itself, not harmful to the microorganisms, it tends to establish a condition in which the volatile acids are converted into their salts resulting in salt toxicity and thus retardation of the fermentation process (35).

AMMONIA AND ORGANIC NITROGEN RELATIONSHIP

The relationship of ammonia-nitrogen and organic nitrogen concentrations against temperature are presented in Figs. 7a, b, and 8a, b respectively.

Fig. 7a. $\text{NH}_3\text{-N}$ concentration as a function of temperature in hog manure digestion. Solid lines indicate 15-day retention time; dotted lines indicate 10-day retention time.

Units 1 and 2 have $0.15 \text{ lb.VS/ft}^3/\text{day}$ loading rate.

units 3 and 4 have $0.20 \text{ lb.VS/ft}^3/\text{day}$ loading rate.

Units 5 and 6 have $0.25 \text{ lb.VS/ft}^3/\text{day}$ loading rate.

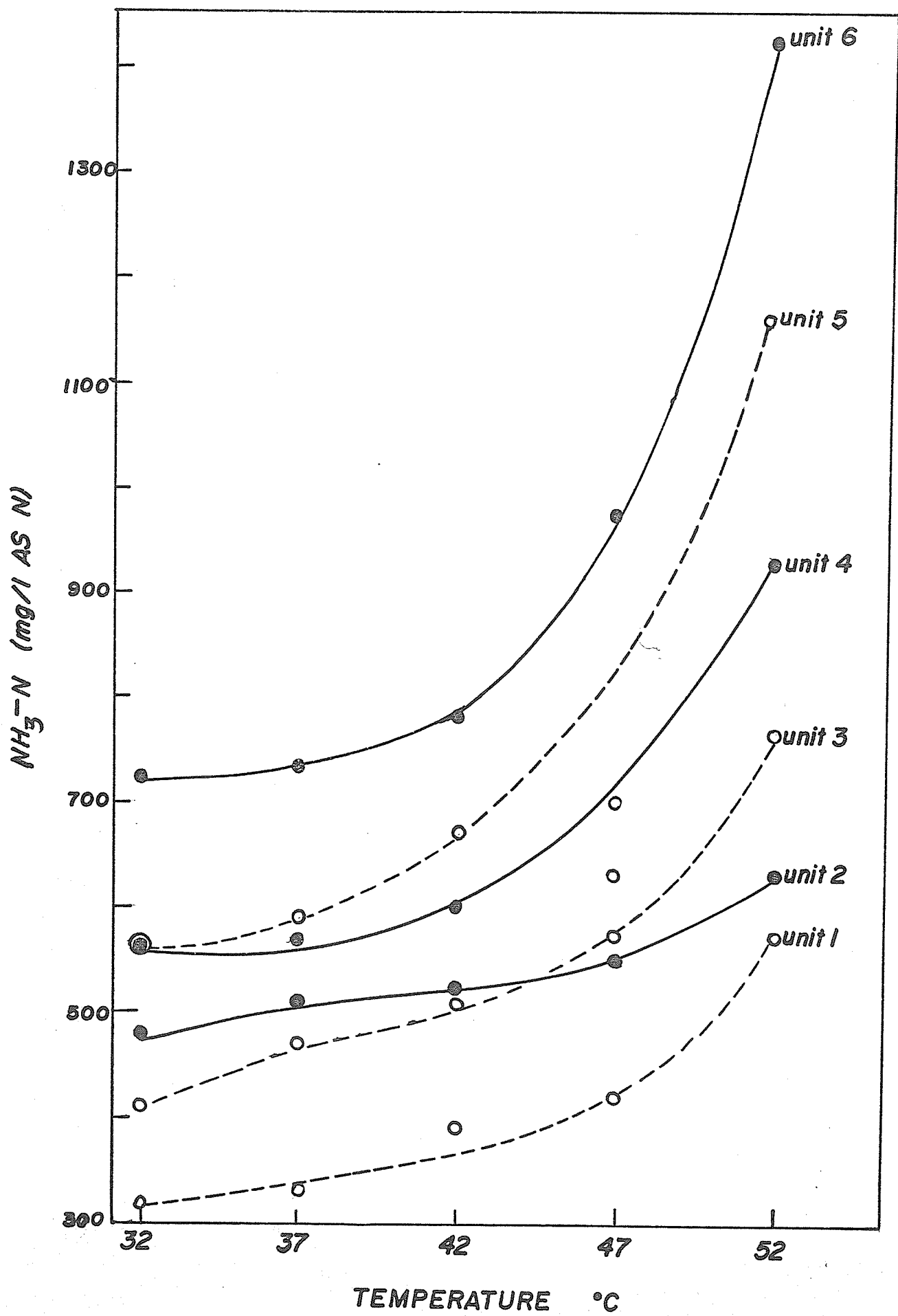


Fig. 7b. $\text{NH}_3\text{-N}$ concentration as a function of temperature in dairy cattle manure digestion. Solid lines indicate 15-day retention time; dotted lines indicate 10-day retention time.

Units 1 and 2 have $0.15 \text{ lb.VS/ft}^3/\text{day}$ loading rate.
Units 3 and 4 have $0.20 \text{ lb.vS/ft}^3/\text{day}$ loading rate.
units 5 and 6 have $0.25 \text{ lb.VS/ft}^3/\text{day}$ loading rate.

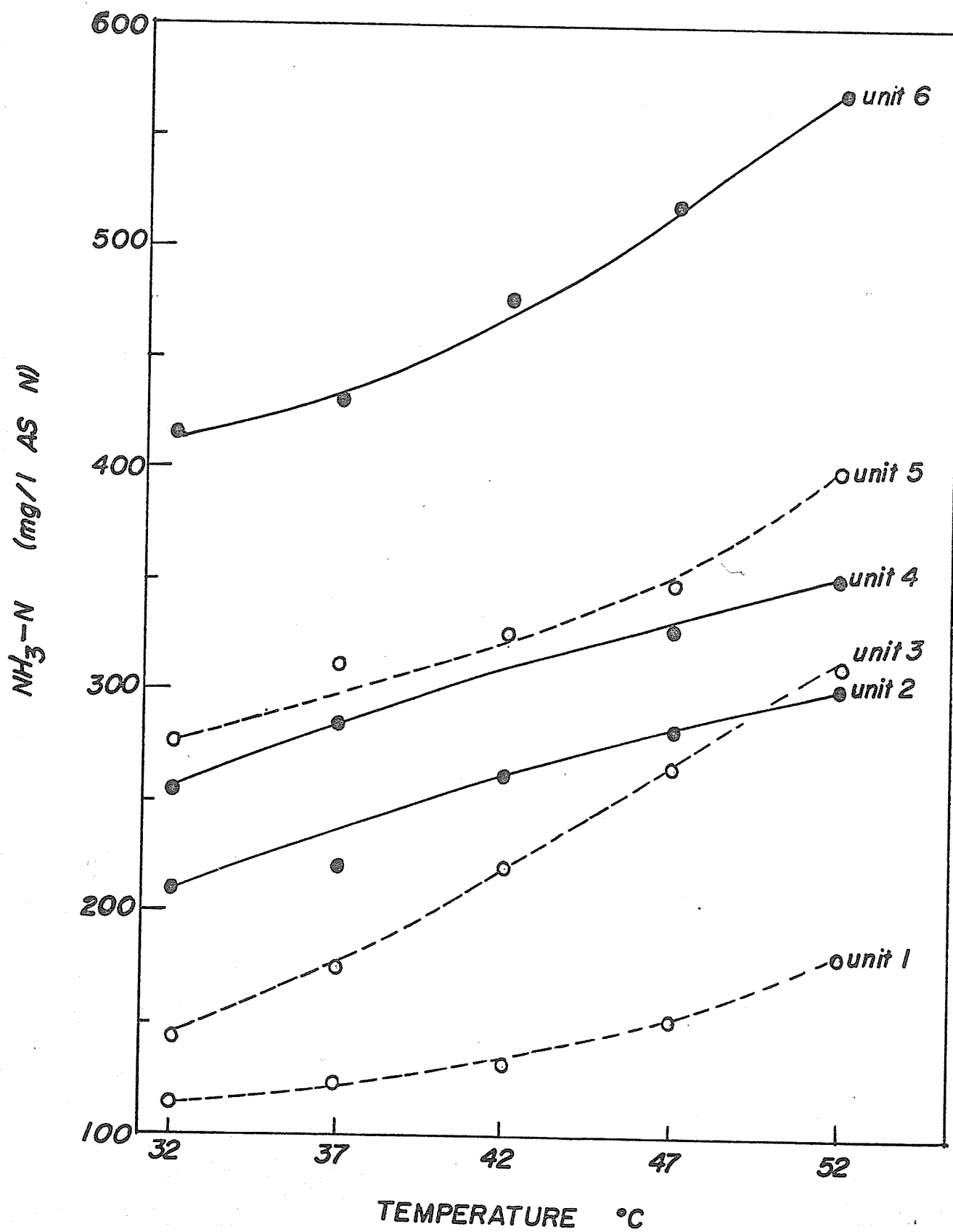


Fig. 8a. Organic-N concentration as a function of temperature in hog manure digestion. Solid lines indicate 15-day retention time; dotted lines indicate 10-day retention time.

units 1 and 2 have $0.15 \text{ lb.VS/ft}^3/\text{day}$ loading rate.

Units 3 and 4 have $0.20 \text{ lb.VS/ft}^3/\text{day}$ loading rate.

units 5 and 6 have $0.25 \text{ lb.VS/ft}^3/\text{day}$ loading rate.

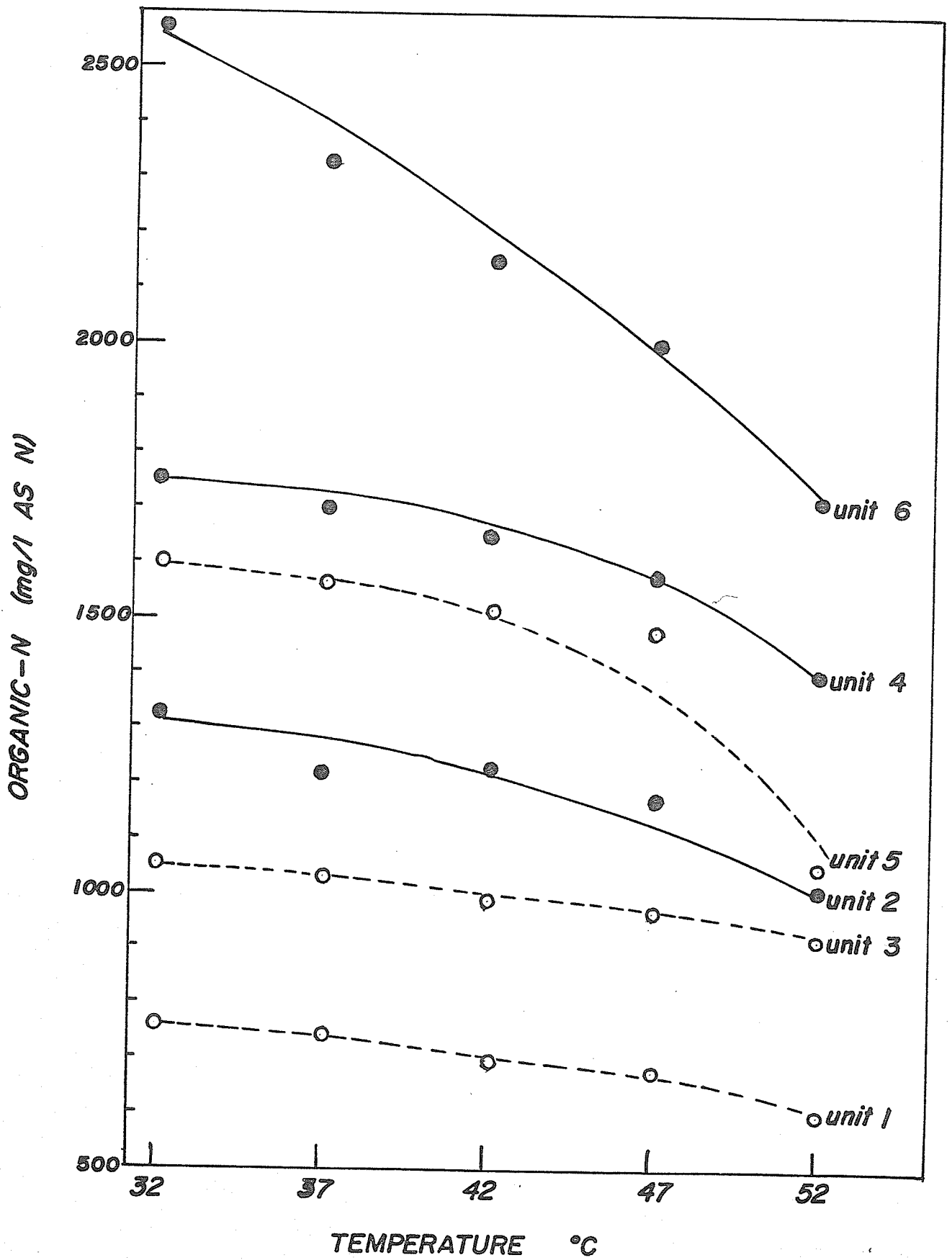
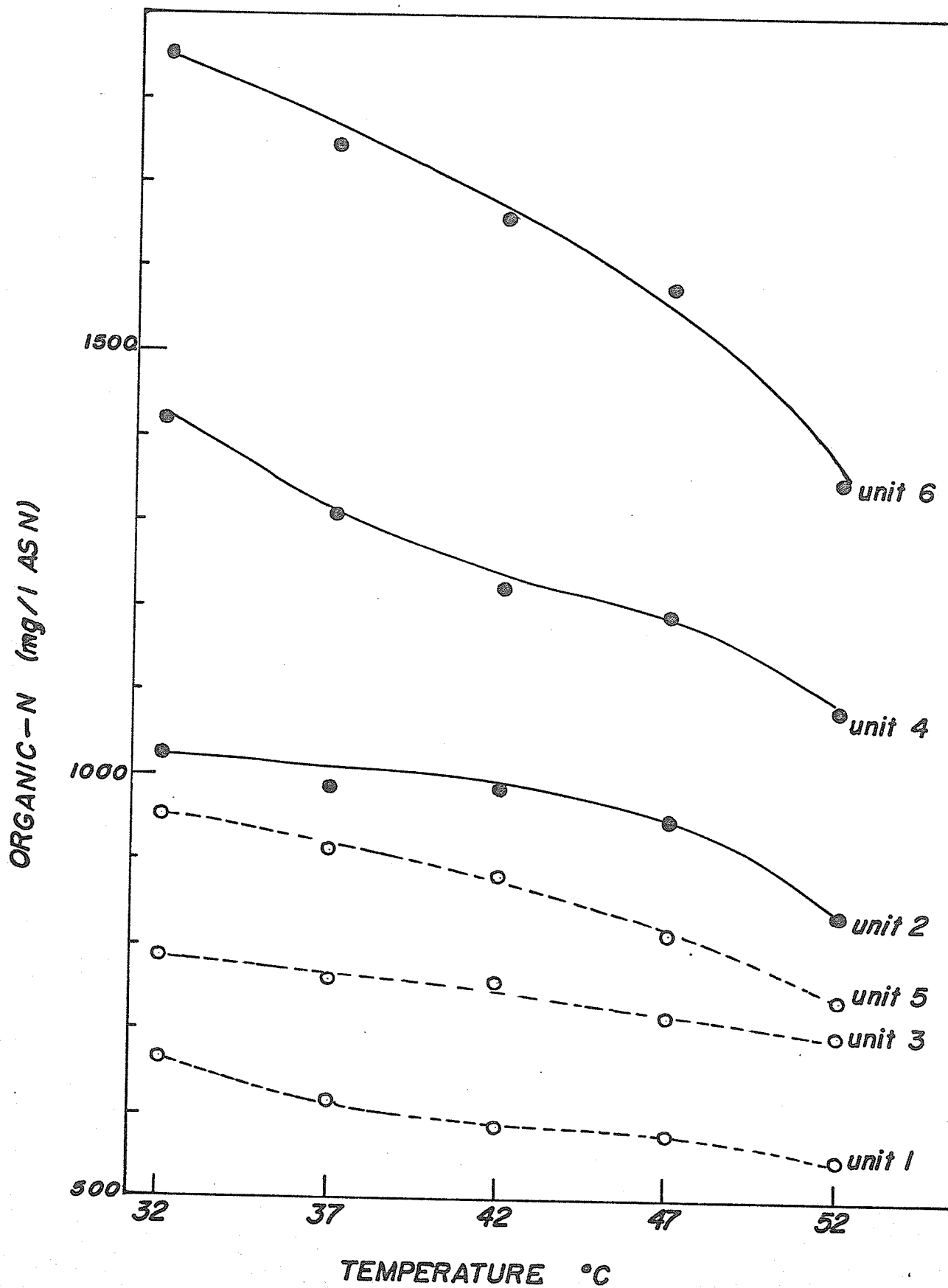


Fig. 8b. Organic-N concentration as a function of temperature in dairy cattle manure digestion. Solid lines indicate 15-day retention time; dotted lines indicate 10-day retention time.

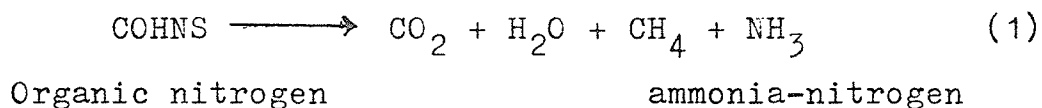
Units 1 and 2 have $0.15 \text{ lb.VS/ft}^3/\text{day}$ loading rate.

Units 3 and 4 have $0.20 \text{ lb.VS/ft}^3/\text{day}$ loading rate.

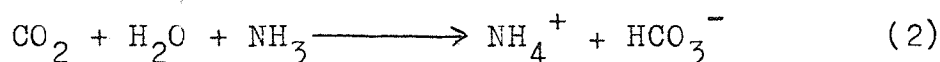
Units 5 and 6 have $0.25 \text{ lb.VS/ft}^3/\text{day}$ loading rate.



Data indicated that the accumulation of ammonia-nitrogen in the digester is associated with the decomposition of organic nitrogen mainly in the forms of protein and urea in the waste. According to Toerien et al (48), this decomposition can be represented in general terms by the equation:



The ammonia-nitrogen thus produced reacts with carbon dioxide to form ammonium bicarbonate.



The NH_4^+ and HCO_3^- remain in solution and contribute to the alkalinity and the buffering capacity of the substrate (45).

In both hog and dairy cattle waste digestion, the rate of accumulation of ammonia-nitrogen and the decomposition of organic nitrogen was parallel with increases in loading rates, retention times and temperatures (Figs. 7a, b and 8a, b). For example, in hog manure digestion at 42°C, Unit 3 had an ammonia-nitrogen concentration of 510 mg./litre while Unit 5 had 670 mg./litre (Fig. 7a). This indicated that an increase in loading rate of 0.5 lb. VS/ft.³/day increased the ammonia-nitrogen concentration by approximately 30 percent. Also, Unit 4 which had a 5-day longer retention time than Unit 3, show a correspondingly higher ammonia-nitrogen concentration.

At thermophilic temperatures (52°C), in both hog and dairy cattle waste digestion, the ammonia-nitrogen concentration was very much higher than that at mesophilic temperatures. Similar phenomenon have been reported by other investigators (41, 43). According to Gouleke (41), high ammonia production associated with thermophilic digestion is due to the more complete break down of proteins.

GAS PRODUCTION AND METHANE COMPOSITION

Gas production from hog manure digestion ranged from 47.0 to 104.3 ml./gm.VS destroyed/day, or 7.73 to 16.8 $\text{ft}^3/\text{lb.VS}$ destroyed/day (Fig. 9a). The sludge gas contained approximately 57 to 60 percent methane (Fig. 10a). These data are similar to those reported by other workers (11,38). Taiganides et al (36) reported that at 37°C the average gas yield per day per pound of volatile solids added to the digesters ranged from 7.8 to 10.3 ft^3 with a methane content of approximately 59 percent.

The dairy cattle wastes gas production ranged from 26.5 to 89.9 ml./gm. VS destroyed/day, or 4.2 to 14.5 $\text{ft}^3/\text{lb.VS}$ destroyed/day (Fig. 9b) at various temperatures, retention times and loading rates. The methane content of the gas in this case ranged from 57 to 68 percent (Fig. 10b), which was normal for domestic sludge gas (37). In general, these are in agreement with those reported by Gramms et al (38).

It was obvious from the results that gas production

Fig. 9a. Gas production, ml./gm.VS destroyed/day as a function of temperature in hog manure digestion. Solid lines indicate 15-day retention time; dotted lines indicate 10-day retention time. Units 1 and 2 have 0.15 lb.VS/ft³/day loading rate. Units 3 and 4 have 0.20 lb.VS/ft³/day loading rate. Units 5 and 6 have 0.25 lb.VS/ft³/day loading rate.

GAS PRODUCTION, ml./gm. VOLATILE SOLID DESTROYED / DAY

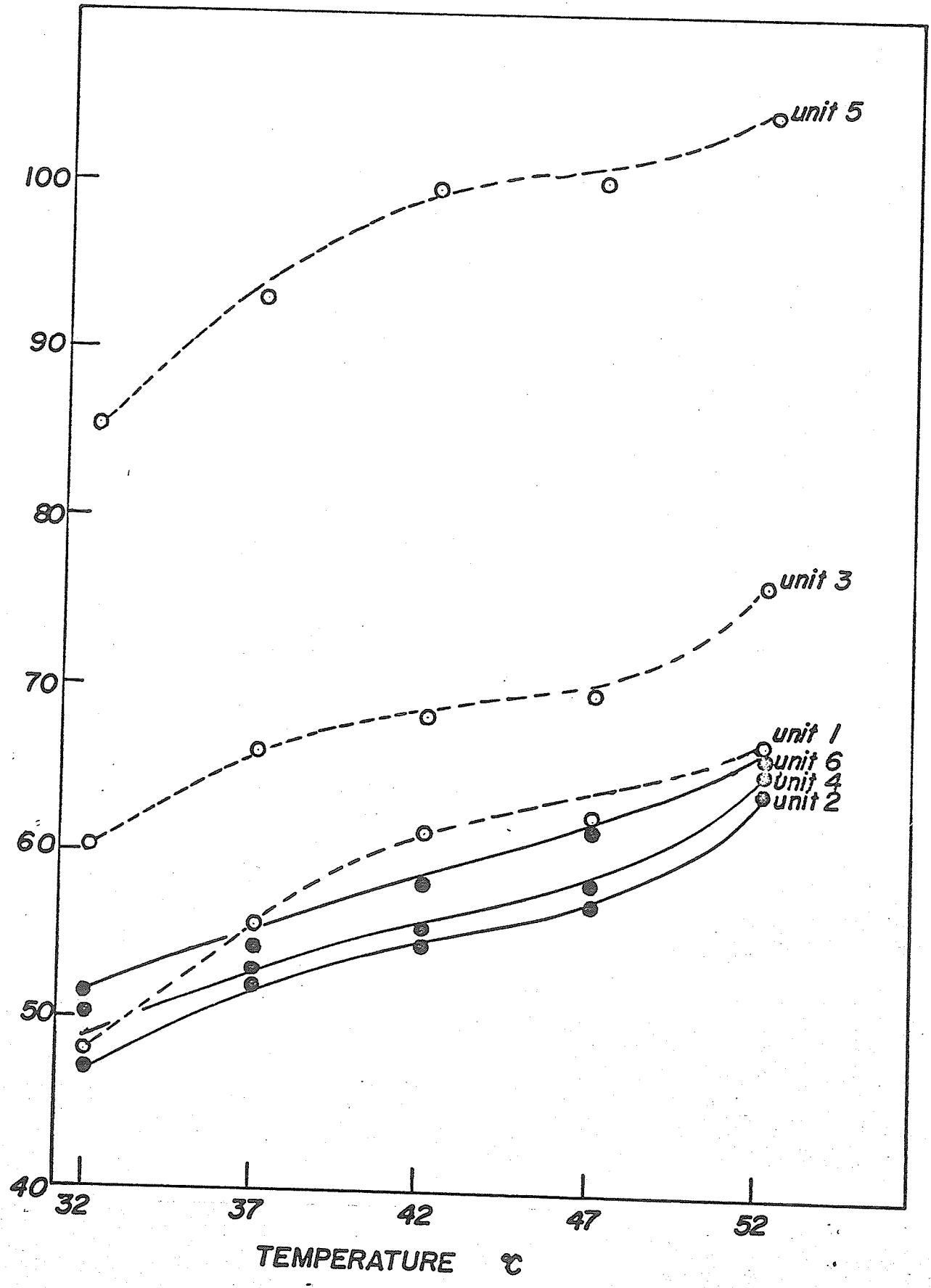


Fig. 9b. Gas production, ml./gm. VS destroyed/day as a function of temperature in dairy cattle manure digestion. Solid lines indicate 15-day retention time; dotted lines indicate 10-day retention time. Units 1 and 2 have 0.15 lb.VS/ft³/day loading rate. Units 3 and 4 have 0.20 lb.VS/ft³/day loading rate. Units 5 and 6 have 0.25 lb.VS/ft³/day loading rate.

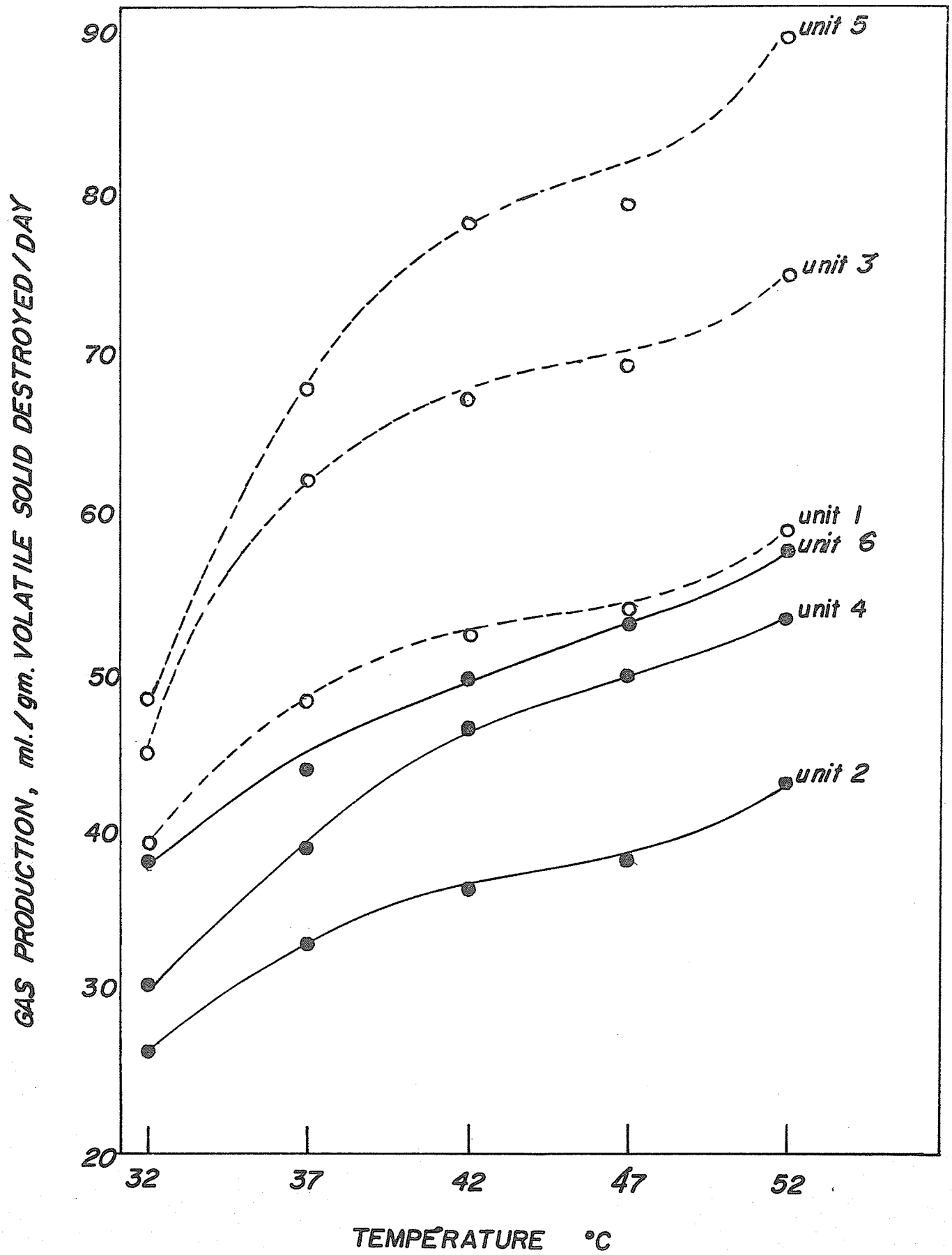


Fig. 10a. Percent methane as a function of temperature in hog manure digestion. Solid lines indicate 15-day retention time; dotted lines indicate 10-day retention time.

Units 1 and 2 have $0.15 \text{ lb.VS/ft}^3/\text{day}$ loading rate.

Units 3 and 4 have $0.20 \text{ lb.VS/ft}^3/\text{day}$ loading rate.

Units 5 and 6 have $0.25 \text{ lb.VS/ft}^3/\text{day}$ loading rate.

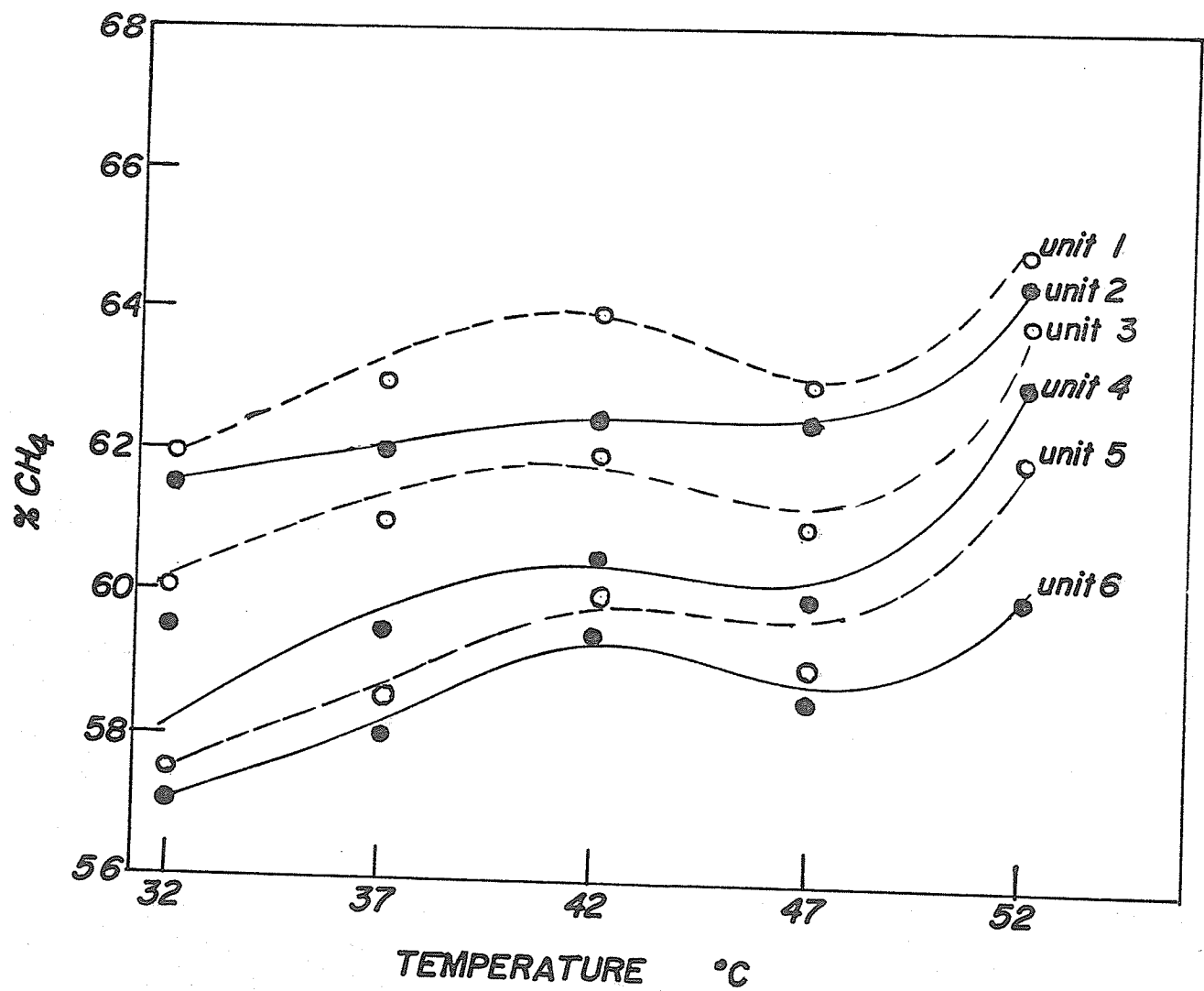
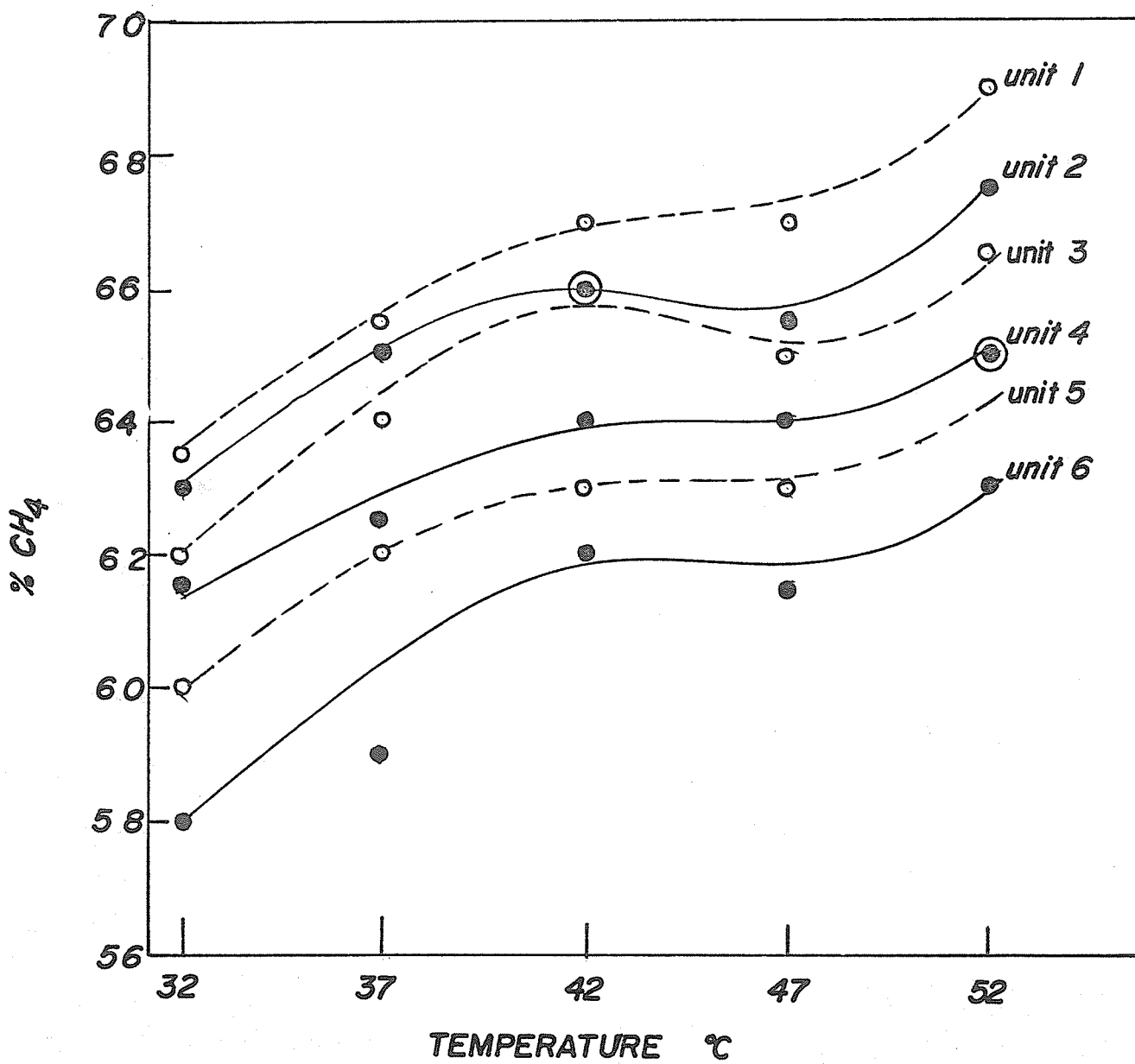


Fig. 10b. Percent methane as a function of temperature in dairy cattle manure digestion. Solid lines indicate 15-day retention time; dotted lines indicate 10-day retention time.

Units 1 and 2 have $0.15 \text{ lb.VS/ft}^3/\text{day}$ loading rate.

Units 3 and 4 have $0.20 \text{ lb.VS/ft}^3/\text{day}$ loading rate.

Units 5 and 6 have $0.25 \text{ lb.VS/ft}^3/\text{day}$ loading rate.



and its methane content increased with temperature, loading rate and retention time, (Fig.10a, b, 11a, b) from 32 to 42°C, but decreased slightly at 47°C. However, at 52°C, gas production again increased very considerably. In hog manure digestion, for example, the amount of gas produced at 52°C was about 2.5 times greater than that produced at 32°C. Similar results were also noted for cattle waste digestion. This phenomenon is common in domestic sludge digestion (41, 42, 43) and will be considered in detail.

GENERAL EFFECT OF TEMPERATURE ON ANAEROBIC DIGESTION

The reactions taking place in these anaerobic digestion are a result of the activity of a most heterogeneous population. The effect of temperature, therefore, likely is a reflection of the behaviour of bacteria at different temperatures (48). Goleuke (41) and Malina (42) reported that three temperature ranges existed for the anaerobic digestion process: a thermophilic range above 45°C, a mesophilic range of 28 to 45°C, and a psychrophilic range below 10°C. In each of these temperature zones, a different group of bacteria predominates (48). Therefore, a particular population can thus be described as psychrophilic, mesophilic or thermophilic depending on the temperature region in which optimal growth is obtained. Any drastic changes in temperature in the digester may impair the activity or may even be lethal to the component microorganisms. Generally, however,

Fig. 11a. Gas production in litres/day/4 litres sludge as a function of temperature in hog manure digestion. Solid lines indicate 15-day retention time; dotted lines indicate 10-day retention time. Units 1 and 2 have $0.15 \text{ lb.VS/ft}^3/\text{day}$ loading rate. Units 3 and 4 have $0.20 \text{ lb.VS/ft}^3/\text{day}$ loading rate. Units 5 and 6 have $0.25 \text{ lb.VS/ft}^3/\text{day}$ loading rate.

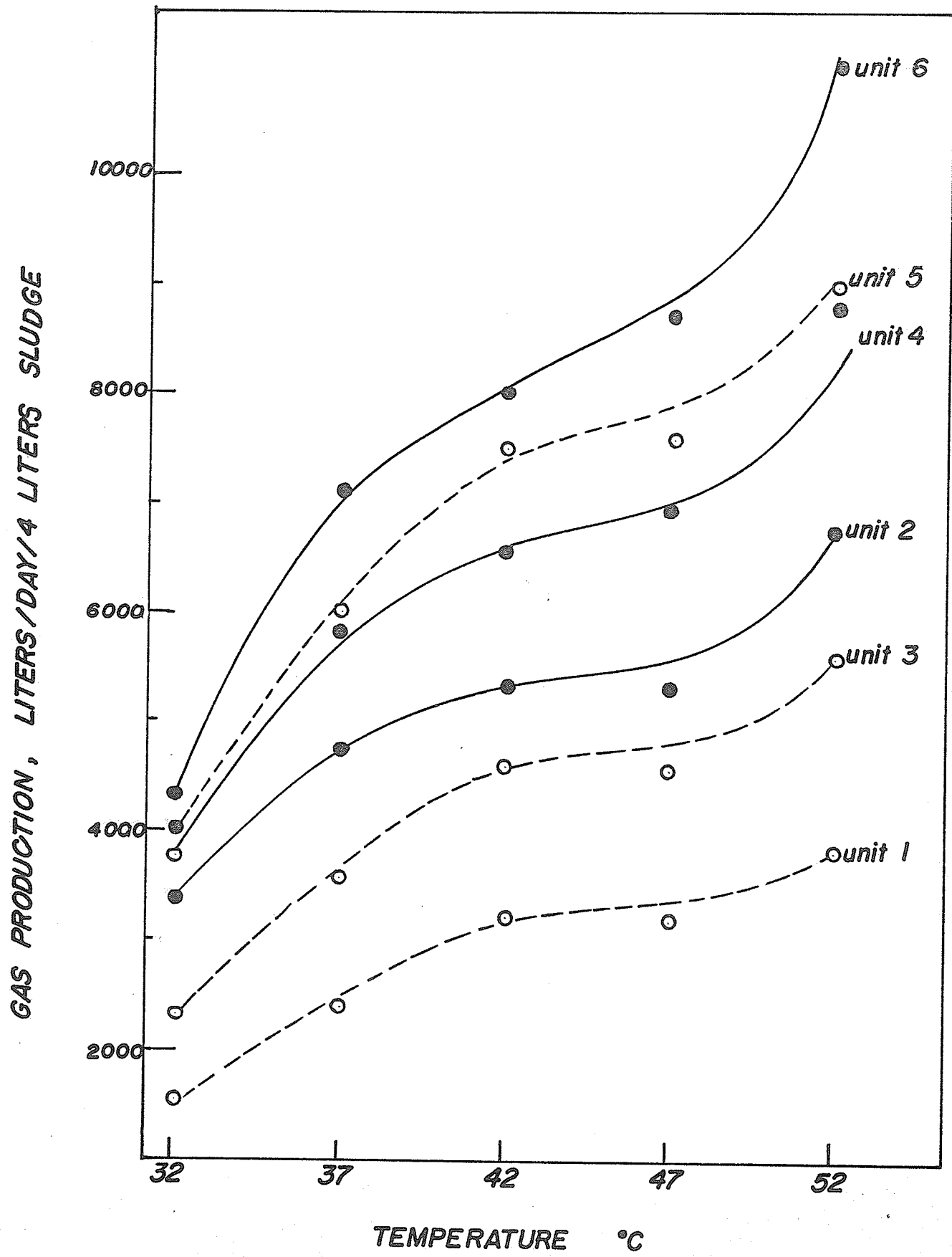
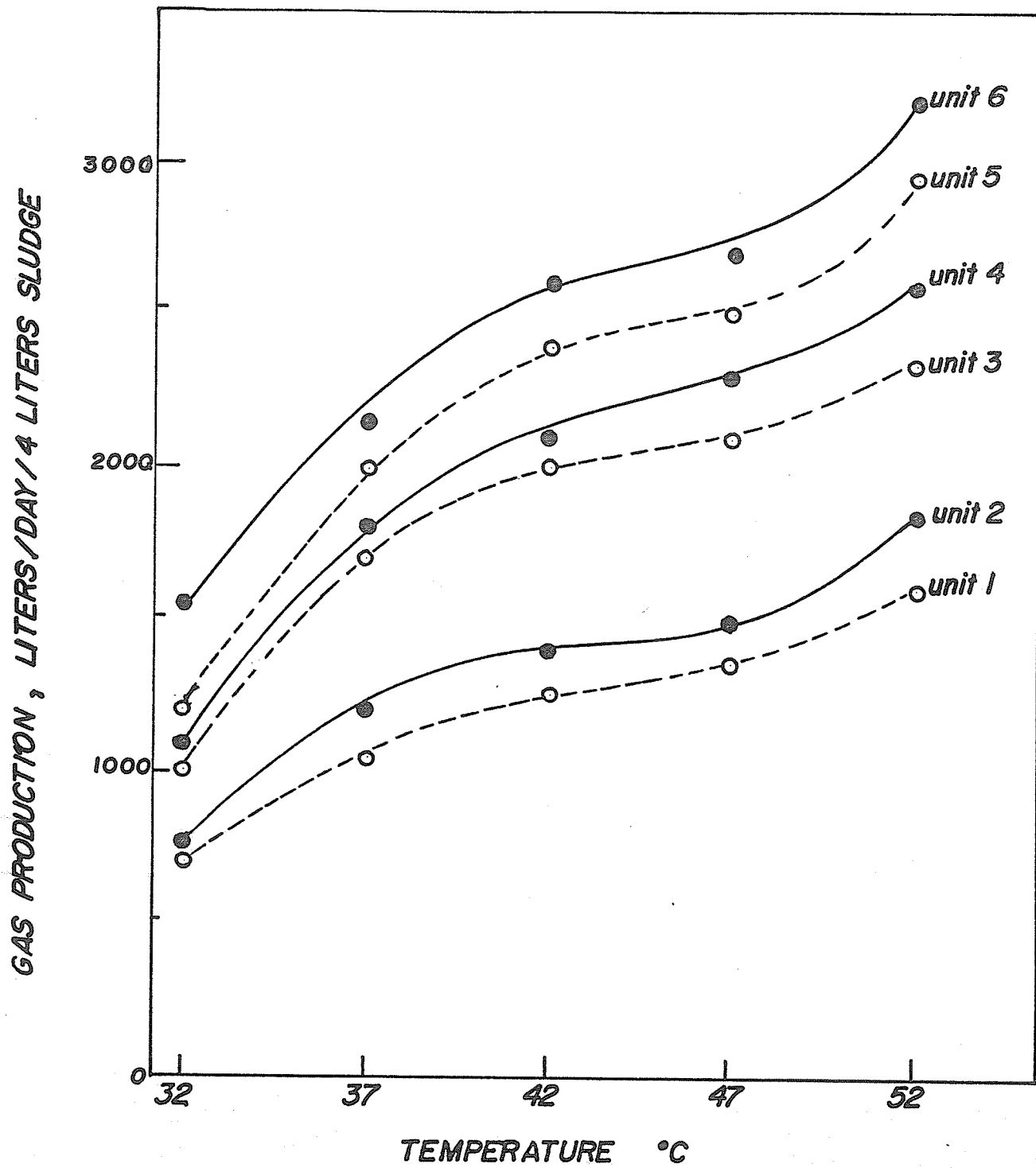


Fig. 11b. Gas production in litres/day/4 litre sludge as a function of temperature in dairy cattle manure digestion. Solid lines indicate 15-day retention time; dotted lines indicate 10-day retention time. Units 1 and 2 have 0.15 lb.VS/ft³/day loading rate. Units 3 and 4 have 0.20 lb.VS/ft³/day loading rate. Units 5 and 6 have 0.25 lb.VS/ft³/day loading rate.



one might expect a selection mechanism would occur to favor organisms that find the new temperature to be optimum.

From the results obtained, it was apparent that there was a retardation of methane fermentation at the upper limit of the mesophilic range. As previously indicated, the rate of methane fermentation increased steadily from 32 to 42°C. This may be due to the fact that 37°C is the optimum temperature for most members of the mesophilic group. However, at 47°C, there was retardation in volatile solids reduction, gas production and methane content. The reason for this is not clear, but it is thought that at this transition zone between mesophilic and thermophilic populations neither mesophiles nor thermophiles flourish and consequently digestion will be slow and unsatisfactory (43).

The sharp increase of microbial activity from 47°C to 52°C indicated that another population of bacteria, probably thermophilic, became established in the digestion process. There was a significant increase in gas production, volatile solids reduction and methane content (Fig. 2a,b, 9a, b and 11a, b). This is likely due to a more complete digestion of organic material at the high temperature (43).

ECONOMICS OF RECOVERY OF ENERGY FROM HOG AND DAIRY CATTLE WASTES

In hog waste digestion, the average gas yield per

day per pound of volatile solids fed ranged from 7.7 to 16.8 ft.³ (Fig. 10a). Analysis of the gas evolved showed a methane content of approximately 60 percent. The heating value of the gas was estimated to be 570 BTU/ft.³. On the basis of these data, and on averaged hog manure composition, the heating value from the daily waste can be calculated.

It has been reported that a hog of average weight produces approximately 1.1 pound dry manure per day (49) which is equivalent to 0.95 pound of volatile solids. Granting that the digester is operating at 37°C with 0.25 lb. VS /ft.³/day loading rate and 15 day retention time, the amount of gas produced would be 545 ml/gm VS destroyed/day, or 8.8 ft.³/lb. VS destroyed/day (Fig. 10a). At such yields, the heating value of the daily waste from a hog is calculated to be $8.8 \times 0.95 \times 570 = 4500$ BTU. This value agrees with that reported by Taiganides et al (11).

In the same way, the heating value of the daily waste from dairy cattle is estimated to be 31,000 BTU per animal.

Results indicate that the fertilizer value in terms of ammonia increases as a result of anaerobic digestion (Fig. 7a, b). The wastes are initially rich in organic nitrogen in the form of urine, amino acids and proteineous material. These nitrogeneous materials are degraded to ammonia by saprophytic bacteria during the fermentation. In addition to phosphates and trace elements, raw manure

contains a relatively high nitrogen content though in a form not directly available as a plant fertilizer. After mineralization and solubilization during the anaerobic digestion process, the nitrogen is converted largely to ammonium bicarbonate, a form more directly usable by plants. Its fertilizer value accordingly is enhanced. In Taiwan, it is reported that the digested manure is being utilized as fertilizer to produce Chlorella, a unicellular algae with a high nutritive value for animal feeding (10).

GENERAL CONCLUSION

GENERAL CONCLUSION

Anaerobic digestion of livestock waste is a complex phenomenon mainly because of the large differences in digestible material normally available and because of the heterogeneity of the bacterial population. As such, the picture of each individual biochemical mechanism is often obscured. As a result, quantitative chemical analyses relative to the intermediate and final products of digestion are routinely used as artificial criteria for the evaluation of digester conditions. All the chemical characteristics are directly or indirectly dependent on each other and no one of these characteristics alone can adequately represent the conditions of the digester.

Based on the results obtained, digesters operating at a temperature range of 37 to 42°C is recommended. The rate of volatile solid reduction and gas production at 42°C were, respectively, 30% and 200% greater than that at 32°C in hog waste digestion, Fig. 2a and 9a. Similar results were also observed in dairy cattle waste digestion, Fig. 2b and 9b. Although the quality of the gas in terms of methane content and the quality of effluent sludge was better at 52°C than at mesophilic temperatures in both wastes digestion, the amount of heat energy required to maintain the digester at that temperature is not considered economically feasible (40).

Results also indicated that anaerobic digesters for dairy cattle waste may be loaded at rates of 0.25 lb.VS/ft³/day with retention times as short as 10 days without indication of digester failure. However, anaerobic digesters for hog manure should not be loaded at rates greater than 0.25 lb.VS/ft³/day and should have a retention time between 15 to 18 days for good reduction of volatile solids and COD. Higher loading rates may result in accumulation of ammonia which is detrimental to the digestion process.

Statistics in June 1969 indicated that there were 1,019,000 cattle and 612,000 hogs in Manitoba (49) producing approximately 10⁷ pounds of wet manure daily. Estimation of recoverable energy from this renewable biomass through anaerobic digestion is shown in the data below:

	livestock population	Estimated heat energy from daily waste per head	Total energy	
			heat energy	gasoline* equivalent
Hog	612,000	4,500 BTU	2.75x10 ⁹ BTU	2.3x10 ⁴ Imp.Gal.
Cattle	1,019,000	31,000 BTU	3.16x10 ¹⁰ BTU	2.63x10 ⁵ Imp.Gal.
			3.43x10 ¹⁰ BTU	2.86x10 ⁵ Imp.Gal.

*Calculation is based on the assumption that one imperial gallon of gasoline on complete combustion yields 120,000 BTU of heat energy (50).

Approximately 3.34×10^{10} BTU of heat energy in the form of methane gas is theoretically recoverable from the daily waste of hog and cattle in Manitoba. This is a tremendous amount of energy and together with equally large amount of energy from poultry wastes, represent a significant source of energy.

Anaerobic digestion applied in the treatment of municipal and domestic sewage has been practiced for many years. Anaerobic treatment of animal wastes has been studied in recent years. Applications with significant gas recovery are apparently successful in Taiwan, India and parts of Europe. In Taiwan, it has been confirmed by personal communication that 6000 units are operating successfully. This development has been the result of a program sponsored by the Joint Chinese-American Commission on Rural Re-construction. It is predictable from the present estimation of experimental result that a significant amount of recoverable energy from the daily livestock waste in Manitoba would make it feasible for future large scale operation of anaerobic digesters for the treatment of livestock wastes. Anaerobic digestion not only provides an effective means of reducing the pollution potential of these very large quantities of livestock wastes produced daily, but also provides a possible alternative in lieu of fossil fuels in the forthcoming years.

REFERENCES

REFERENCES

1. Starr, C. (1971). Energy and power. Scientific American. 225: 3, 37.
2. Miner, J.R. (1971). Farm animal waste management. U.S. Northern Central Publication, 206 Special Report 67, May.
3. Carre, I. (1947). Farm manure gas. Chem. et. Indust. 30: 147
4. Brother McCabe, J. and W.W. Eckenfelder, Jr. (1957). In "Anaerobic Digestion and Solid-liquid Separation". Reinhold Publishing Corp., New York, N.Y.
5. Nicholson, A., agent at Pyrford Court Estate, Pyrford near Woking, Surrey, England. Personal communication.
6. Schmidt, F. (1951) Development of the Bihugas Plant Allerhopits, capacity and efficiency. Deutshe Bihugas Gmbh, Verden/Aller.
7. Schmidt, F. and W. Eggersgluss (1954). Production of high power gas from agricultural wastes or other organic matter. Pap. World Pwr. Conf., Rio de Janeiro.
8. Grindrod, B. (1971). The Marvelous Chicken-powered Motor Car: The Mother Earth News No. 10, July, 1971.
9. Singh, R.B. (1971). Bio-Gas Plant, Generating Methane from Organic Wastes. Rashtra Bhashha Press, Publisher, Raja ki Mandi AGRA-2, India.

10. Chinese-American Joint Commission on Rural Reconstruction (1970). 'The animal methane-chlorella cycle'. Taipei, Taiwan.
11. Taiganides, E.P., E.R. Buamann, H.P. Johnson and T.E. Hazen (1962). Anaerobic digestion of hog wastes. J. Agr. Eng. Res. 70: 327.
12. McKinney, R.E. (1962). In 'Microbiology for Sanitary Engineering'. McGraw-Hill, Publisher, New York, N.Y.
13. Buswell, A.M. and S.L. Neave (1930). Laboratory studies of sludge digestion. Illinois St. Water Surv. Bull. 30:1.
14. Barker, H.A. (1936a). On the biochemistry of the methane fermentation. Arch. Mikrobiol. 7: 1404.
15. McCarty, P.L., J.S. Jeris and W. Murdoch (1963). Individual Volatile acids in anaerobic treatment. J. Water Pollut. Control Fed. 35:1501.
16. O'Shaughnessy, F.R., (1914). The utilization of the phenomena of putrefaction with special reference to the treatment and disposal of sewage sludge. J. Chem. Soc. Ind. 33: 3.
17. Hutchkiss, M. (1924). Studies on the biology of sewage disposal. A survey of the bacteriological flora of a sewage treatment plant. J. Bacteriol. 9:437.
18. Ruchhoft, C.C., J.G. Keller and G.P. Edwards (1930). Studies of the bacterial population during sludge disposal. J. Bacteriol. 19: 269.

19. Hungate, R.E. (1950). The anaerobic mesophilic cellulolytic bacteria. *Bact. Rev.* 24:353.
20. Barker, H.A. (1940). Antonie von Leeuwenhoek. *J. Microbiol.Serol.* 6: 201.
21. Cookson, J.T. and N.C. Burbank (1965). Isolation of anaerobic and facultative bacteria present in the digestion process. *J. Water Pollut. Control Fed.* 37: 822.
22. Barker, H.A. (1936). Studies upon the methane-producing bacteria. *Arch. Mikro.* 7: 420.
23. Stephenson, M. and L.H. Stickland (1933). Hydrogenase, III. The bacterial formation of methane by the reduction of one-carbon compounds by molecular hydrogen. *Archs. Biochem.* 27: 1517.
24. Stadtman, T.C. and H.A. Barker (1951). Studies on the methane formation, X, a new formate-decomposing bacterium, Methanococcus vanniellii. *J. Bacteriol.* 62:269.
25. Barker, H.A. (1956). In "Bacterial Fermentation". John Wiley & Sons, Inc., New York, N.Y.
26. Schnellen, C.G.T.P. (1947). Onderzrekingen over dee Methaangisting. Dessertation Technische Hoogeschool, Delft.
27. Laigret, J. (1945). Methane fermentation by Bacillus perfringens. *Acad. Sci., Paris.* 221: 359.
28. McCarty, P.E. (1964). " The methane fermentation" in "Principles and applications of Aquatic Microbiology." John Wiley and Sons, Inc., New York.
29. McCarty, P.L. (1967). "Kinetics of waste assimilation in

- anaerobic treatment." In " Industrial Microbiology." American Inst. of Biological Sciences, Washington, D.C. 7: 144.
30. McCarty, P.L. and R.E. McKinney (1961) Volatile acid toxicity in anaerobic digestion. J. Water Pollut. Control Fed. 33: 9, 978.
31. Hobson P.N. Personal Communication.
32. Standard Methods for the Examination of Water and Waste Water. 13th ed. American Public Health Association, Inc., New York, N.Y.
33. Albertson, O.E. (1961). Ammonium nitrogen and the anaerobic environment. J. Water Pollut. Control Fed. 12:3. 359.
34. Kugelman, I.J. and P.L. McCarty (1965). Cation toxicity and stimulation in anaerobic waste treatment. J. Water Pollut. Control Fed. 37: 1, 97.
35. McCarty P.L. and R.E. McKinney (1961). Salt toxicity in anaerobic digestion, J. Water Pollut. control Fed. 33: 4, 399.
36. Taiganides, E.P. and R.K. White (1969). The menance of noxious gases in animal units. Trans. ASAE. 12: 3, 359.
37. Dague, R.R., R.L. Hopkins and R.W. Tonn (1970). Digestion fundamentals applied to digester recovery- two case studies. J. Water Pollut. Control Fed. 42:9, 1666.
38. Gramms, L.C., L.B. Polkowske and S.A. Witzel (1969). Anaerobic digestion of farm animal wastes. Presented at ASAE annual meeting, Purdue University, Indiana.

39. Maly J. and H. Fadrus (1970). Influence of temperature on anaerobic digestion. J. Water Pollut. Control, Fed. 43: 4, 555.
40. Heukelekian, H. (1939). Further studies on thermophilic digestion of sludge solids. Sew. Works Jour. 2: 219.
41. Goleuke, C.G. (1958). Temperature effects on anaerobic digestion of raw sewage sludge. Sew. Ind. Wastes. 30: 1225.
42. Malina, J.F. (1964). Thermal effects on completely mixed anaerobic digestion. Water and Sew. Works. Jan.
43. Rowe, D.R. (1971). Anaerobic sludge digestion, mesophilic-thermophilic. Water and Sew. Works. March.
44. Anaerobic sludge digestion--manual of practice, No. 16 J. Water Pollut. Control Fed. 38: 10, 1683.
45. Dague, R.R. (1968). Application of digestion theory to digester control. J. Water Pollut. Control Fed. 40: 12, 2021.
46. Mueller, L.E. , E. Hindin, J.V. Lunsford and G.H. Dunstan (1959). Some characteristics of anaerobic sludge digestion--I. Effect of loading. Sew. Ind. Wastes. 31: 669.
47. Pohland F.G. (1962). Laboratory investigation on the thermological aspects and volatile acids, alkalinity behavior during anaerobic sludge digestion. Ph.D. Thesis, Purdue University.

48. Toerien, D.F. and W.H.J. Hattingh (1969). Anaerobic digestion; the microbiology of anaerobic digestion. *Water Res.* 3: 385.
49. Hudek E.P. (1970). Waste management problems in the primary agricultural industry. Seminar on waste management, Univ. of Manitoba, Sept. 30.
50. Kirkpatrick, P.C. (1963). *Perry's Chemical Engineer's Handbook*. 4th ed. McGraw-Hill, Publisher, New York, N.Y.