

INVESTIGATION OF SOME FACTORS INFLUENCING
DENITRIFICATION IN A LABORATORY SOIL COLUMN
WITH A SURFACE LOADING OF LIVESTOCK WASTES

by 6408

CHARLES VERNON BOORAM JR.

B.S., Northeast Missouri State, 1966

B.S., Kansas State University, 1968

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A MASTER'S THESIS

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requirements for the degree



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INTRODUCTION

Livestock in the United States produce 2 billion tons of waste per year or an animal population equivalent to that of two billion people. This production is divided among 48.6 million cattle, 57 million hogs, 3 billion poultry, and 20.9 million sheep(27).

Loehr (16) in discussing water pollution control legislation pointed out that when waste disposal is on land, nitrogen levels may well be the limiting factor for the maximum amount of animal wastes that can be applied to the soil, since soluble nitrogen in the form of nitrates can percolate to the ground waters.

Keller and Smith (13) indicated that 30-50 percent of the rural water supply samples they studied in Missouri contained more than 5 mg. per liter of nitrate (USPHS standard is 45 milligrams per liter of nitrate). They reported that the main contaminating source was waste matter at the site of animal habitation. Smith (22) reported in 1965 that feedlots were contaminating properly constructed deep wells which were located close to feedlots. The high concentration of ammonia nitrogen in feedlot runoff and lagoon effluent contribute to the nitrate concentration in surface waters when the ammonia is oxidized. This contamination of water supplies is important. If these waters are consumed by small babies, death may result.

Stewart et al (25) in 1967 reported that nitrates were moving through the soil and into ground water supplies under both feedlots and irrigated fields in crops, excluding alfalfa.

It is evident that agriculture is polluting the environment. It

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is also evident that nitrates are contributing to the contamination of some ground water supplies. Miner (19) reported in 1966 that cattle feedlot runoff contained considerable amounts of ammonia nitrogen. This provides a potential source of nitrogen for nitrification and the formation of nitrates.

The nitrate ion is very mobile and is therefore readily transported through the soil profile. The soil would provide an excellent media to dispose of animal wastes if the bacteria could biologically reduce the nitrates to nitrogen gas. The problem is to determine the conditions under which denitrification will occur.

PURPOSE

The purpose of this research was to further develop equipment so that a nitrogen balance could be run on a laboratory soil system. The primary objective was to determine under what conditions denitrification would occur when a soil is provided with large amounts of nitrogen carrying organic matter.

REVIEW OF LITERATURE

Nitrogen removal by the process of microbial denitrification has been a topic of research for many years. Much research has been done to determine the various steps that occur as nitrates are transformed to nitrogen gas (molecular nitrogen). There is general agreement that denitrification will occur when anaerobic conditions exist. This concept holds true for soil systems and liquid waste treatment systems. There exists a school of thought which holds that in a permeable soil

it is impossible for denitrification to occur. This precludes the possibility that zones of anaerobiosis occur.

Since many literature reviews exist on the processes of mineralization, nitrification, and denitrification, only a summary of the processes will be included in this thesis. A few examples of denitrification in biological waste treatment systems will be included to show the similarity between the soil and biological waste treatment systems.

A small portion of the total nitrogen in the nitrogen cycle is present in an available form for plant life at any one time (7). Most of it is tied up in plants that use it for synthesis and animals that feed on the plants. Ninety-eight percent of the nitrogen in the soil occurs in the organic form. Inorganic nitrogen rarely exceeds 2 to 3 percent of the total soil nitrogen.

Mineralization of nitrogen from fresh organic residues or from the more stable humus fractions is dependent upon a number of environmental factors. Physical and chemical conditions of the soil habitat, such as moisture, pH, aeration, temperature, the total nitrogen status of the soil, and the inorganic nutrient supply, will govern the activities of the soil flora and the rate of mineralization.

The capacity of soil microorganisms to convert ammonia to nitrate is of utmost significance. This sequence of events is termed nitrification. Ammonia is oxidized to nitrite and the nitrite is oxidized to nitrate. The intermediate steps between these states are still somewhat in doubt.

Denitrification has been defined as the biological reduction of nitrate to nitrite to molecular nitrogen or the oxides of nitrogen.

The process results in the escape of nitrogen into the atmosphere. This mechanism has been known for nearly a century. Since that time, researchers have determined that when denitrification occurs there must be an energy source for the bacteria to utilize, and the oxygen concentration must be low.

Use of the soil as a disposal medium for wastes containing high concentrations of nitrogenous materials will require that the nitrogen which reaches the nitrate stage be converted to a form that will not rapidly move through the soil profile. The avenues of escape would be for nitrates to be converted to molecular nitrogen or the volatilization of nitrogen as ammonia.

Koelliker (15) reported an 80 percent reduction in total nitrogen concentration when swine finishing wastes were applied by sprinkler irrigation to plots of fescue grass. It was assumed that nitrogen was lost due to denitrification.

Gillham and Webber (11) conducted a study to determine the quantity of inorganic nitrogen reaching the groundwater from a particular barnyard, and to examine various factors affecting the extent of the resulting contaminated zone of groundwater. Several observation wells were installed to measure the water quality. The ground water was flowing beneath the barnyard, so it was possible to monitor the amount of inorganic nitrogen added by the barnyards.

Nitrogen gas is present in the atmosphere; therefore, the demonstration of molecular nitrogen release is difficult to show. Because of measurement difficulties, field experiments usually involve the measurement of nitrogen losses over a long period of time. The

preparation of a soil nitrogen balance requires that precise data be available on the quantity of nitrogen put into a system and that leaving the system via leaching and volatilization.

Allison (2) discussed several examples of lysimeter research conducted since 1910. Experiments were conducted at Ithaca, New York, for long periods of time. During a 15 year period, two vegetable crops were grown per year plus the growing of rye as a winter cover crop. Twenty-five percent of the nitrogen could not be accounted for in the nitrogen balance. Experiments at Geneva, New York, indicated losses of 17-36 percent of the available nitrogen to a rotation of timothy, barley, and wheat. It was concluded that deficits of nitrogen escaped to the atmosphere. Experiments conducted at Windsor, Connecticut, Knoxville, Tennessee, and Riverside, California, reported that 15 to 20 percent of the total nitrogen could not be accounted for.

Greenhouse experiments conducted using clay pots have the advantage over field lysimeters in that drainage losses are prevented. Both the lysimeter and the clay pot method rely upon the deficit of nitrogen to show that denitrification occurs. The unaccounted fraction presumably is lost by chemical and biological volatilization. Four methods of volatilization have been proposed and are listed as follows:

- a) nonbiological losses of ammonia;
- b) chemical decomposition of nitrite under acid conditions to yield nitrogen oxides;
- c) production of molecular nitrogen by the nonenzymatic reaction of nitrous acid with ammonia or the amino acids; and
- d) microbial denitrification leading to the liberation of the

molecular nitrogen and nitrous oxides.

The previous methods described all use the concept of chemistry by difference. P.J. Ross et al (20) developed a gas tight growth chamber to investigate gaseous nitrogen changes that occur in the soil-plant-atmosphere system. Atmospheric air was replaced with argon and oxygen (75% argon and 25% oxygen). Isotopes of nitrogen were used in the nitrogen added to the system. This allowed the tracing of the N^{15} isotopes when the gas was analyzed with the mass spectrometer. Plants grown in the sealed chamber were analyzed for total nitrogen, and then redistilled for isotope ratio analysis. This type of system allowed experiments of up to 30 days in length.

Stefanson and Greenland (23) conducted an experiment using a sealed soil-plant system. Atmospheric air was replaced with a mixture of argon and oxygen. This enabled the detection of small amounts of evolved nitrogen. The gaseous components were analyzed using a gas chromatograph.

George (10) designed a system to study denitrification in the laboratory. The system was designed so that water samples in the profile could be taken automatically. The gas analysis system was automated. He was unable to quantify the flow of gas passing through the system, but was able to show a loss of nitrogen from the soil-water-manure system.

Meek et al (17) analyzed the relation of dissolved oxygen, soluble carbon, and redox potential to the movement of nitrate in soil columns. Disappearance of nitrate was associated with decreases in redox potential, oxygen content of the soil solution, and oxygen concentration in the soil atmosphere. Denitrification occurred without submergence

if the redox potential was sufficiently low. The study demonstrated that periodic drying increased soluble carbon in the soil solution. This promotes denitrification.

Growth of the micro-organisms concerned in denitrification is not dependent upon the reduction of nitrate. Bacteria are active in many other transformations. Arable fields contain an abundance of denitrifying micro-organisms. Hence, the potential for volatilization is enormous, but the conditions must be suitable for a denitrifying type of metabolism for the bacteria (1).

As early as 1950 investigators began to consider the denitrification process as a means of removing nitrogen from sewage and industrial waste water. Table 1 gives possible sources.

TABLE 1 Nitrogen Removal Processes (21)

Biological	Chemical	Physical
Biological treatment	Ion exchange	Ammonia stripping
Anaerobic denitrification	Electrochemical treatment	Reverse osmosis
Algae harvesting	Electrodialysis	Distillation
		Land application

The existence of nitrates in a waste water is essential if the denitrification process is to work. Many schemes have been used to remove nitrates in waste treatment systems. The basic scheme is to aerate the liquid waste and put it through an anaerobic stage. The aeration and the bacteria cause nitrification. The anaerobic stage causes the nitrates to be converted to molecular nitrogen.

One of the problems in the operation of the denitrification process is in the control procedures necessary for maintaining continuous nitrification and denitrification.

Sherman and MacRae (24) studied the reduction of nitrate by adaptive cells of pseudomonas denitrificans in relation to the oxygen concentration in the surrounding fluid. No nitrate reduction occurred when oxygen was continually present at low concentrations. It appears that the competition between oxygen and nitrate for the donor electrons so favors the oxygen that nitrate reduction only occurs when the supply of oxygen is inadequate to meet the demand.

Bollag et al (8) isolated several reducing organisms from the soil. The organisms were compared under various environmental conditions. Various combinations of bacteria revealed that interactions occur among the microbes during growth in relation to their denitrifying activity.

Isolated cases have shown that denitrification can occur in waste treatment systems. Some research has been done with biological sewage treatment systems to remove nitrogen from the effluent. Young and Bungay (26) used 2 grams per liter of powdered iron at various pH levels to remove nitrates from sewage effluent. Ammonia was the principal end product in each case. Because of the low pH required, it is unlikely the process could be used on a large scale.

Pilot scale studies have shown that modification of the activated sludge process to include controlled zones of anaerobiosis, provides removal of 80 percent of the influent nitrogen. Barth et al (5) conducted a series of field surveys to determine whether deliberate

modification might increase nitrogen removal in municipal plants or whether the pilot plant data was atypical. Five plants were used for sample points in the study. Nitrogen balances were calculated for each of the plants. The inability to control process variables effectively makes more efficient nitrogen removal by modification of existing structures unlikely. The results demonstrated that denitrification was possible if oxidized nitrogen was present.

Johnson and Schroepfer (12) conducted a laboratory experiment on sewage effluent to investigate a biochemical, denitrification process for nitrogen removal. Appreciable nitrification is obtained in the activated sludge process. The dissolved oxygen concentration in the mixed liquor appears to be an important factor. Several batch denitrification tests were conducted to determine the rate of denitrification in an activated sludge unit. It was found that by adding raw waste to mixed liquor in the ratio of 1:5 the nitrate nitrogen was depleted in approximately 3 hours; while the ammonia nitrogen increased to 6 mg per l. The rate of denitrification was extremely low without the addition of raw waste.

Balkrishnan and Eckenfelder (6) observed that in an activated sludge process an oxygen concentration of 6 mg per l not only preserved but increased the nitrate concentration. There was good correlation between the rate of denitrification and the rate of total organic carbon removed. The factors important in denitrification are the presence of oxygen, amount and type of organic material, concentration of nitrate in the substrate, and solids concentration.

In conventional waste treatment processes, when nitrification is complete, little easily degradable organic carbon remains and organisms at the mixed liquor exit are not in active metabolism. Several process modifications have been attempted to supply organic carbon and stimulate the organisms by diverting a portion of some organic rich influent stream to mix with the nitrified process stream. Barth et al (4) combined the activated sludge process with the addition of methyl alcohol to force denitrification. The alcohol supplies a source of soluble carbon that the bacteria can readily oxidize.

Seidel and Crites (21) conducted a pilot plant study to test a denitrification system. They used an anaerobic filter with the injection of methanol at the entrance of the filter. They were able to remove 90 percent of the inorganic nitrogen with a detention time of 1.5 hours. The upflow anaerobic filter gave the best overall results.

Koelliker and Miner (15) conducted a study to determine the amount of denitrification in swine oxidation ditch wastes when a daily addition of raw manure feed was made. They concluded that the organic carbon in a well nitrified effluent from swine waste treatment is not sufficient as an organic carbon source for denitrifying bacteria. Nitrified swine wastes can be denitrified by adding raw swine manure to nitrified effluent at a feed rate of $BOD_5 = 3.26 \text{ NO}_3\text{-N}$.

Summary of Literature

The comparison of denitrification in a soil system and in a liquid waste treatment system would seem to show that many common factors exist. They are as follows:

- 1) Both require a source of soluble carbon;
- 2) Both rely upon a period of anaerobiosis;
- 3) Both require nitrification as a necessity for denitrification and;
- 4) Both are difficult to control process wise.

Experimental methods include lysimeter studies, clay pot studies in greenhouses, sealed soil columns (static system), and soil columns (dynamic system) with gas lysimetry. The sealed system and the soil column with gas lysimetry are unique. They allow the total quantification of nitrogen in the soil-water system. This allows the researcher to get away from the concept of chemistry by difference.

EQUIPMENT

The basic system used in this experiment was designed in 1969 by George (10). The driving force of the system is a tubing pump (See Figure 1). The controller for the motor allows variation of speed through a wide range. This allows considerable versatility for various environmental conditions.

The tubing pump, F, draws the atmosphere from the soil column, C. The air then passes through a calcium sulfate drying tube, G. This provides dry air for chromatographic, H, analysis. The gas then passes into an air displacement column, B.

A sensing device was necessary to determine the pressure changes in the air displacement column, B. A mercury manometer, D, was equipped with small wire electrodes to function as a switch. The mechanism described is for the output side of the system. As the tubing pump, F,

LEGEND FOR FIGURE 1

- A - Oil Reservoir
- B - Air Column
- C - Soil Column
- D - Mercury Manometers
- E - Sample Flasks
- F - Vacuum Pump
- G - Drying Tube
- H - Gas Chromatograph
- I - Solenoid Valve
- J - Relay
- K - 110 Volt Source
- L - 24 Volt Source

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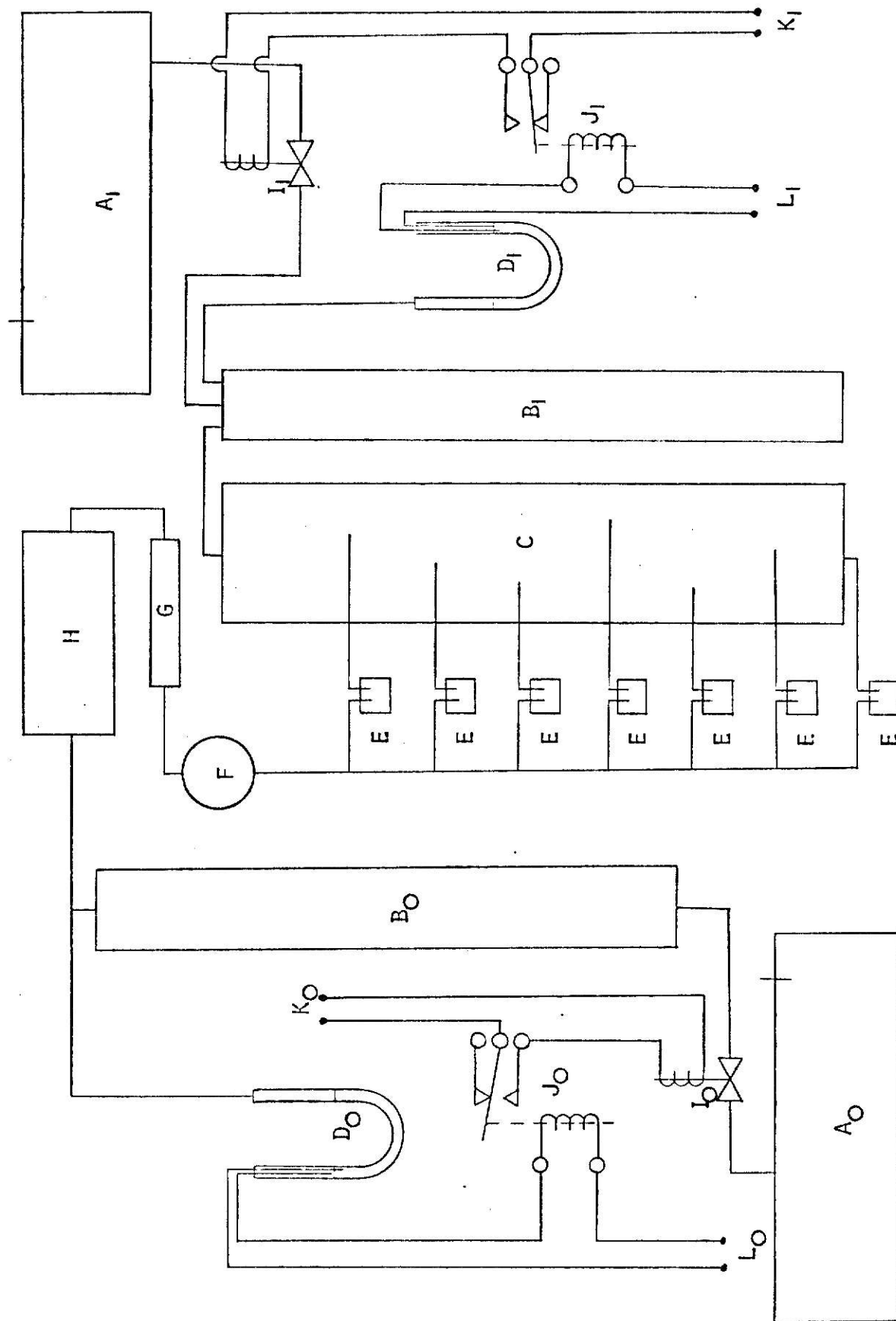


FIG. 1 FUNCTIONAL DIAGRAM OF EXPERIMENTAL SYSTEM

removed air from the soil profile, C, pressure was increased in the air column. This moved the mercury up to the wire contacts. The circuit through the mercury was made, energizing a normally closed relay and completing the circuit to the solenoid valve, I_o . The solenoid valve opened and allowed oil to flow out of the air column, B_o , until the pressure equalized and the mercury switch, D_o , broke contact. Making the contact through the mercury caused the solenoid valve, I_o , to close. The reverse process occurs on the input side of the system.

The flow to oil through the solenoid valve, I_o , and into storage tank, A_o , provided a method to drive the wet test meter. As air was displaced from the reservoir, A_o , it was forced into the wet test meter. This caused the hand on the meter to move and make contact with the brush contact. When contact was made, the control circuit (10) was activated. This activated an integral gas sampling valve on the gas chromatograph. Within the control circuit, a reset timer was activated which activated the recorder. This allowed the output gas to be sampled automatically.

A Fisher - Hamilton Model 29 Gas Partitioner with a 1 millivolt recorder was used to monitor the gas removed from the soil system. Samples were injected into the partitioner at two-one hundredths cubic feet intervals.

The Model 29 Gas Partitioner employs a dual column, dual detector chromatographic system to separate and measure carbon dioxide, nitrogen, methane, and carbon monoxide.

The first gas separation column was 6 feet long and 1/4 inch in

diameter. It was packed with di-2-ethyl-hexylsebacate on 60-80 mesh column pack. The second column was 6.5 feet long, 3/16 inch in diameter, and was packed with 42-60 mesh molecular sieve. As each gas passed one of the detecting thermistors, it produced an electrical signal which was amplified and recorded by an auxiliary recorder. Each separated component produces a peak on the recorded chromatogram. The height of the peak was proportional to the concentration of the compound that produced it; the time at which the peak appears identifies the compound.

A sample component was quantitatively determined by comparing its peak height with that of a standard. Before each weekly run was started, it was necessary to run a standard air sample.

The concentration of the unknown components was determined by using the following equation:

$$C(S) = \left[\frac{H(S)}{H(STD)} \right] C(STD)$$

$C(S)$ = Concentration of sample component percent by volume,

$C(STD)$ = Concentration of standard component percent by volume,

$H(S)$ = Peak height of sample component, and

$H(STD)$ = Peak height of standard component.

A plexiglass cylinder with an inside diameter of 8.5 inches and 72 inches long was used as the module for the soil. The ends of the cylinder were faced with a lathe to insure a good seal. The bottom plate was permanently glued to the end of the plexiglass cylinder. Studs were glued to the inside of the bottom of the cylinder so that a perforated metal plate could be used to support the soil. Fasteners

were fabricated to provide a method to secure the top lid of the column and make it possible to remove the top lid. The top lid was sealed with Formica Brand 140 Adhesive.

Air brought into the column was injected through ports in the top lid of the column. Weekly additions of distilled water were added through a port in the top lid of the column. The wall of the cylinder was fabricated so that samples of the water could be removed through samplers at specified points in the profile. Tensiometers were used in an attempt to monitor the soil moisture content.

An automatic control panel designed by Mensch and Reece (18) was used to control equipment that extracted water from the soil-water system. The controller operates on 24 volt D.C. Minute and second pulse generators in the counting system make it possible to measure time from one second to 99 minutes. The basic purpose of the control panel was to control the tubing pump and solenoid clamps that allow one point to be sampled at a time. The water samples were drawn from the soil column by a tubing pump. The suction line from each of the 13 collection flasks ran through a solenoid operated pinch clamp to a common suction line, which led through the tubing pump and the gas chromatograph. The automatic control panel made it possible to control the tubing pump and the solenoid clamps so that the suction lines to each sample bottle could be opened separately.

Erlenmeyer flasks, E, containing sulfuric acid were used to trap water samples. Electrodes with 110 volt potential between them were used to by pass a full sample bottle. The water level in the Erlenmeyer flasks makes contact with the electrodes, completing a relay

circuit, and the controller advances to the next step.

This system provides for water sampling, gas sampling and analysis, and gas quantification. Water samples are trapped in Erlenmeyer flasks. The chromatograph determines the concentration of carbon dioxide, oxygen, nitrogen, and methane. The air columns allow the volume of air to be measured as it moves into and out of the soil system. Photographs of the equipment are presented in Plate I and Plate II.

PROCEDURES

The soil used in the system was a loamy sand found in the Hunter's Island area south of Manhattan, Kansas. It was sieved through a coarse screen to remove large sticks and stones. The soil was weighed and placed in the plexiglass cylinder. Soil was added until the depth was approximately 60 inches.

The soil was compacted by allowing distilled water to flow up through the soil profile until the water level was above the top of the soil. The water was then released and the soil was allowed to settle for 5 days. The alternate wetting and drying was continued for 21 days. This allowed the soil to settle uniformly.

Soil cores were removed at 12 points in the soil profile, after the profile was settled, and fritted glass samplers were inserted. A composite of the cores removed from the column was analyzed for nitrogen forms.

A layer of air dry manure equivalent to 50 tons per acre (oven dry basis) was added to the soil system. The manure was covered with 4 inches of soil. The top was then placed on the column and sealed.

The control panel was set so that sample points 1 through 13 could

be sampled every two minutes. This sequence was used until the sample bottles contained enough liquid for proper laboratory analysis. A second sequence was used to extract air and water during the remainder of the test run. This sequence extracted a sample from point 13 every 24 minutes.

Water samples were trapped in Erlenmeyer flasks containing sulfuric acid. The acid stopped all biological action and trapped the ammonia.

The experiment was started on 25 August 1970 and was continued until 23 December 1970. Four and one-half liters of distilled water were added to the soil for the first run and three and one-half liters were added for each of the next 16 runs. This was equivalent to 3.75 inches per week.

After 17 test runs of 1 week duration, the soil was sampled at 4 inch intervals and analyzed for organic matter, pH, and nitrogen forms. The manure layer was separated and also analyzed for nitrogen forms.

Soil-Water Quality Analysis

Soil samples in this experiment were analyzed for pH, organic matter, ammonia nitrogen, nitrate nitrogen, and total nitrogen by the Kansas State Soil Testing Laboratory. The results are tabulated in the appendix.

Water samples were analyzed in the Sanitary Engineering Laboratory at Kansas State University. Nitrates were determined using the Brucine test (see Standard Methods), nitrites were determined using the Hach Kit, and ammonia was determined using the direct nesslerization technique (see Standard Methods). The water samples were also analyzed

for Chemical Oxygen Demand and Biological Oxygen Demand according to procedures outlined in Standard Methods (3).

ILLUSTRATIONS

EXPLANATION OF PLATE 1

- Fig. 1 Stand used to support soil column and mechanism to allow rotation of soil column for removal of soil from column
- Fig. 2 Empty soil column with sampling positions denoted by rubber stoppers

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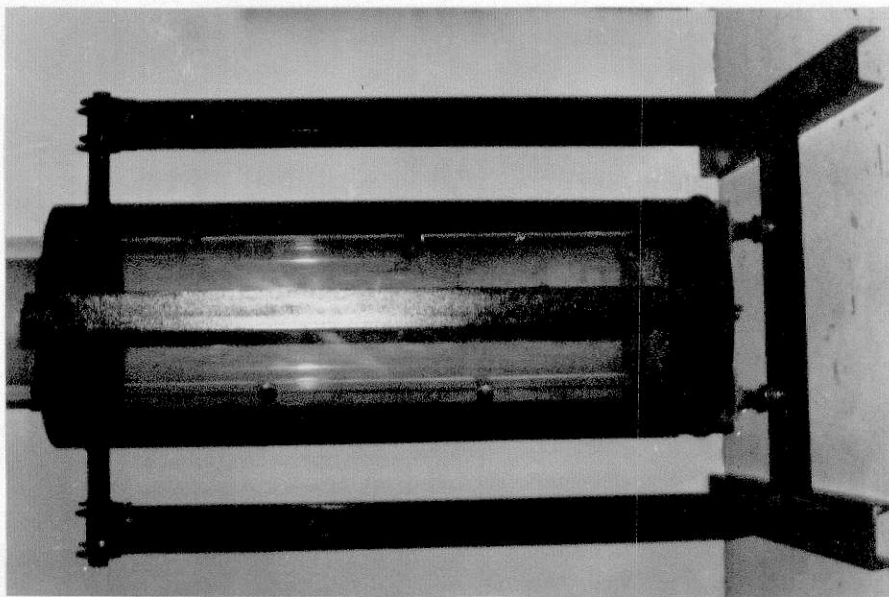


FIG. 1

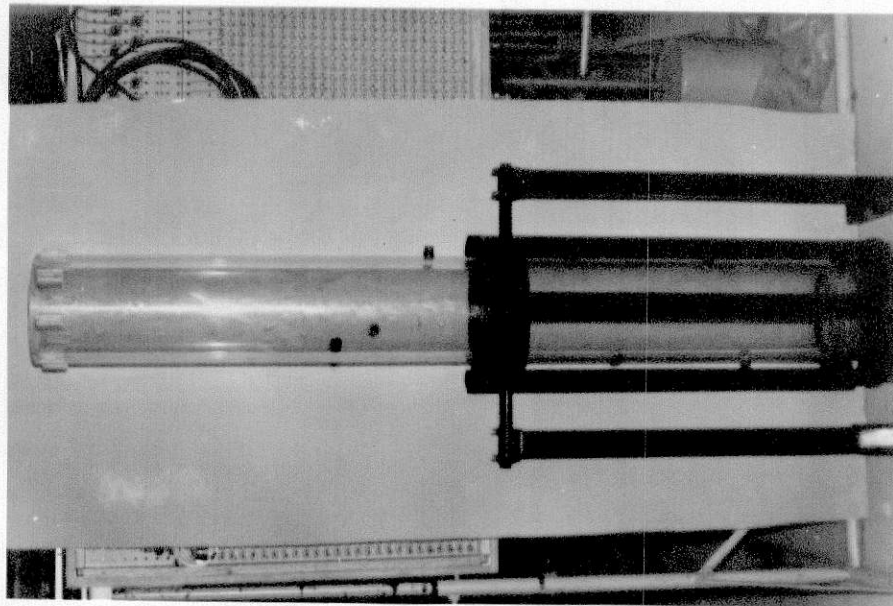


FIG. 2

EXPLANATION OF PLATE 2

Fig. 1 Gas chromatograph and recorder used for gas analysis

Fig. 2 Soil column with soil and manure in place with the
input air displacement column located to the right of
the soil column

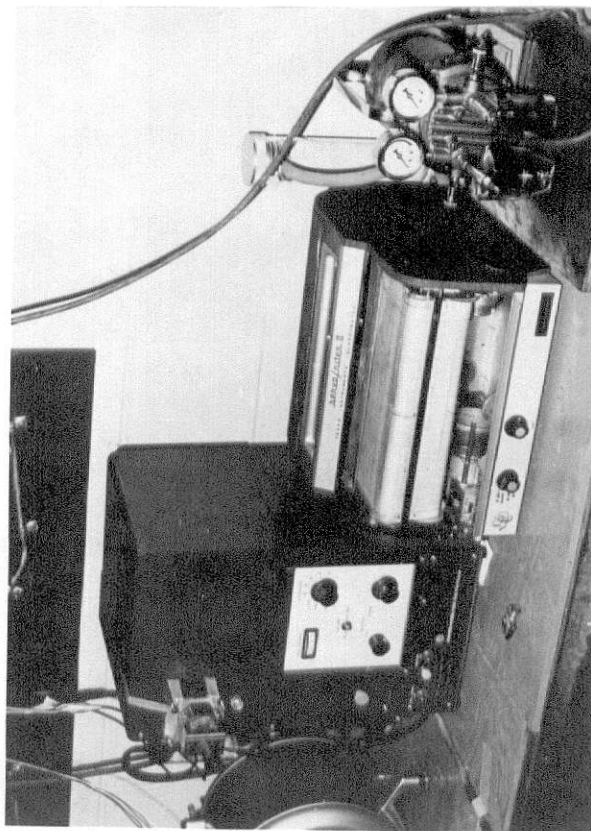


FIG. 1

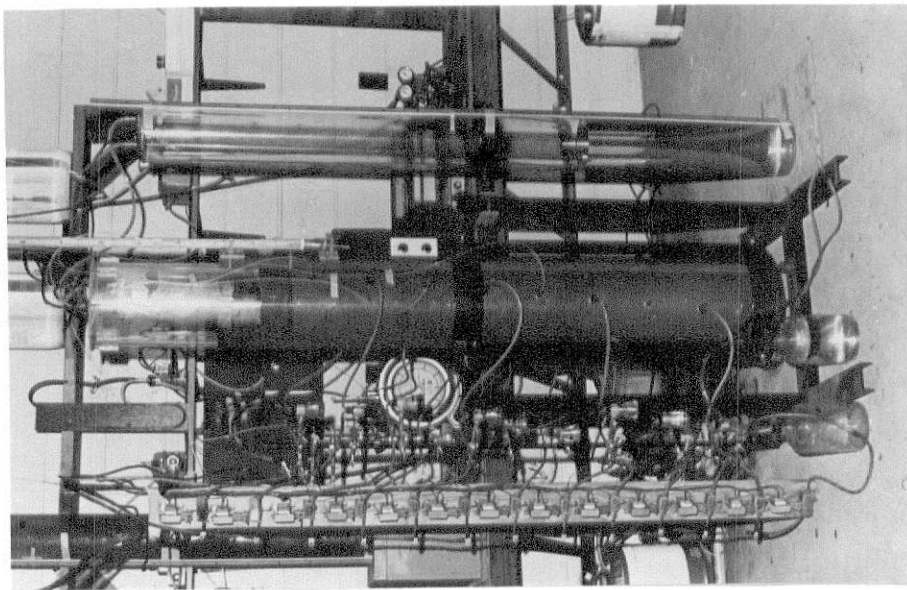


FIG. 2

RESULTS

The collection of data to evaluate denitrification in a laboratory soil column was continued for 17 weeks(Experiment 1). Graphs shown in Figures 2 through 22 explain some of the events that occurred in experiment 1. During this time various problems occurred. The major problem being that of malfunctioning solenoid valves. This allowed the oil in the output air displacement column to be drained into the storage reservoir. The water sampling system, the gas sampling system and the gas measurement system performed satisfactorily.

The composite soil sample taken before the experiment started contained 7.47 ppm of nitrate nitrogen, 2.34 ppm of ammonium nitrogen and 0.0275 percent total nitrogen(organic-N + ammonium-N). Figure 3 shows a plot of the final $\text{NH}_4\text{-N}$ and $\text{NO}_3\text{-N}$ from the soil analysis. This indicates increased nitrification at the bottom of the column. The soil column contained 75.586 kilograms of oven dry soil. The soil contained 21.29 grams of nitrogen and the manure contained 6.86 grams of nitrogen for a total of 28.15 grams of nitrogen in the soil-manure-system(Table 2). The nitrate nitrogen was a very small contributor to the total mass of nitrogen in the system. A final analysis of the soil after 17 test runs indicated that the average nitrate nitrogen concentration was 5.31 ppm, the average ammonium nitrogen concentration was 5.82 ppm, and the average total nitrogen content was 0.0296 percent. The manure lost 5.29 grams of nitrogen and the soil gained 1.42 grams of nitrogen for a net loss of 3.87 grams of nitrogen. Approximately 79.6 percent of the nitrogen lost from the system was due to leaching with the remaining 20 percent due to denitrification. Considering the total amount of nitrogen in the soil and manure, 2.8 percent was lost due to denitrification and 10.94 percent was lost due to leaching for a total loss of 13.74 percent.

TABLE 2
NITROGEN BALANCE SHEET

Initial Nitrogen:

Soil	21.29 grams
Manure	6.86 grams
Total	<u>28.15</u> grams

Final Nitrogen:

Soil	22.71 grams
Manure	1.57 grams
Total	<u>24.28</u> grams

Nitrogen Removed in H₂O 3.08 grams

Nitrogen Removed in air (0.79 grams)

Change in soil nitrogen content = +1.42 grams

Loss of nitrogen from manure = 5.29 grams

Net loss from manure and soil = 3.87 grams

Percent leached from bottom of soil column= 79.6 percent

The ammonia concentration was initially high in the water samples immediately below the manure layer. This high concentration of ammonia moved down the soil profile. When the experiment was stopped after 17 weeks, the ammonia concentration was negligible in all water samples. The maximum ammonia concentration was 110.00 ppm, the maximum nitrate nitrogen concentration was 207.00 ppm and the maximum nitrite nitrogen concentration was 87.50ppm for the 17 test runs.

The nitrate nitrogen concentration was originally high at the upper level of the column, but decreased to negligible concentrations during run 5 and began increasing as new nitrates were formed. This is easily seen in the average nitrate and ammonia nitrogen concentrations in the water samples for the 17 test runs (Fig. 2 and Fig. 5). Figure 5 indicates that nitrification was occurring. The initial high nitrate concentration decreased to acceptable concentrations during run 3 (10 mg/l $\text{NO}_3\text{-N}$). During run 5 (Fig. 10) the average nitrate nitrogen concentration started increasing. The average ammonia concentration increased to a peak during run 3 (Fig. 8). This means that the microorganisms in the soil were transforming the organic nitrogen to ammonia and then after a short delay the ammonia was converted to nitrate and leached out of the soil column. As the mineralization of nitrogen decreased, the nitrification rapidly increased for a short period of time and then started decreasing. This is reasonable to expect because of the lack of ammonia to convert to nitrate.

This leads to an assessment of the nitrate and ammonia movement in the water samples during each test run. Data are presented in Figures 6-22. During run 1 (Fig. 6) the distribution shows no definite trend. This is attributed to the initial concentration of nitrate nitrogen

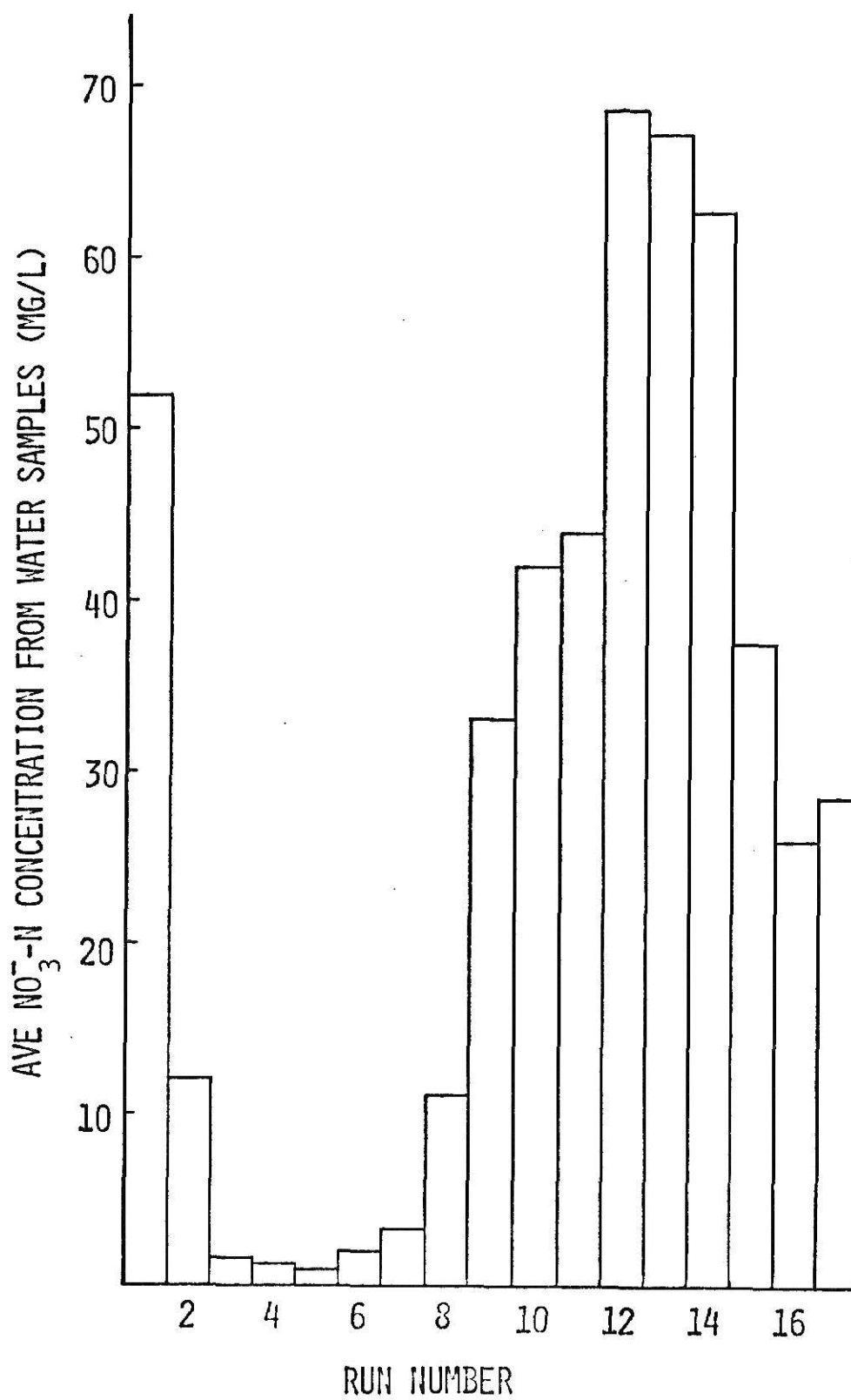


FIG. 2 AVERAGE NITRATE NITROGEN CONCENTRATION
IN WATER SAMPLES

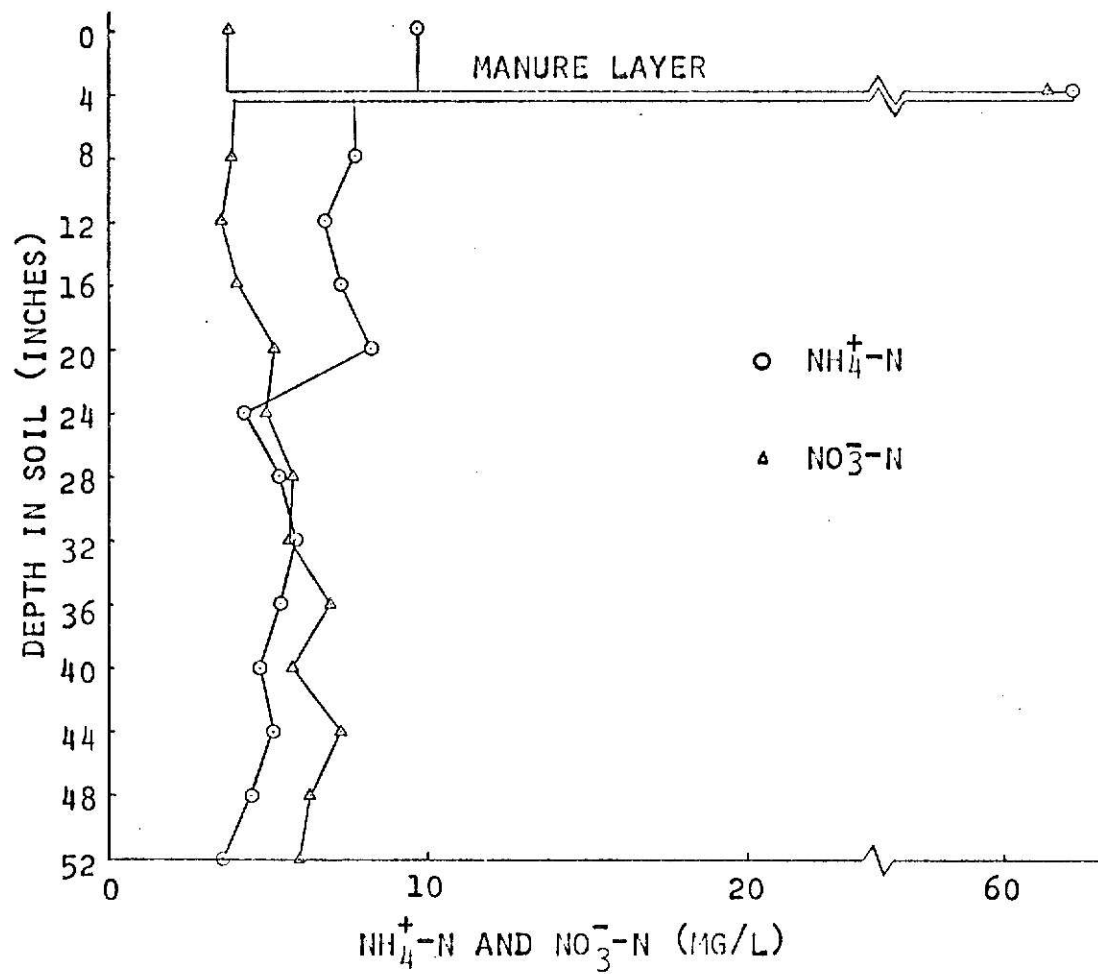


FIG. 3 AMMONIUM AND NITRATE
CONCENTRATIONS IN THE SOIL AT
THE END OF TESTING

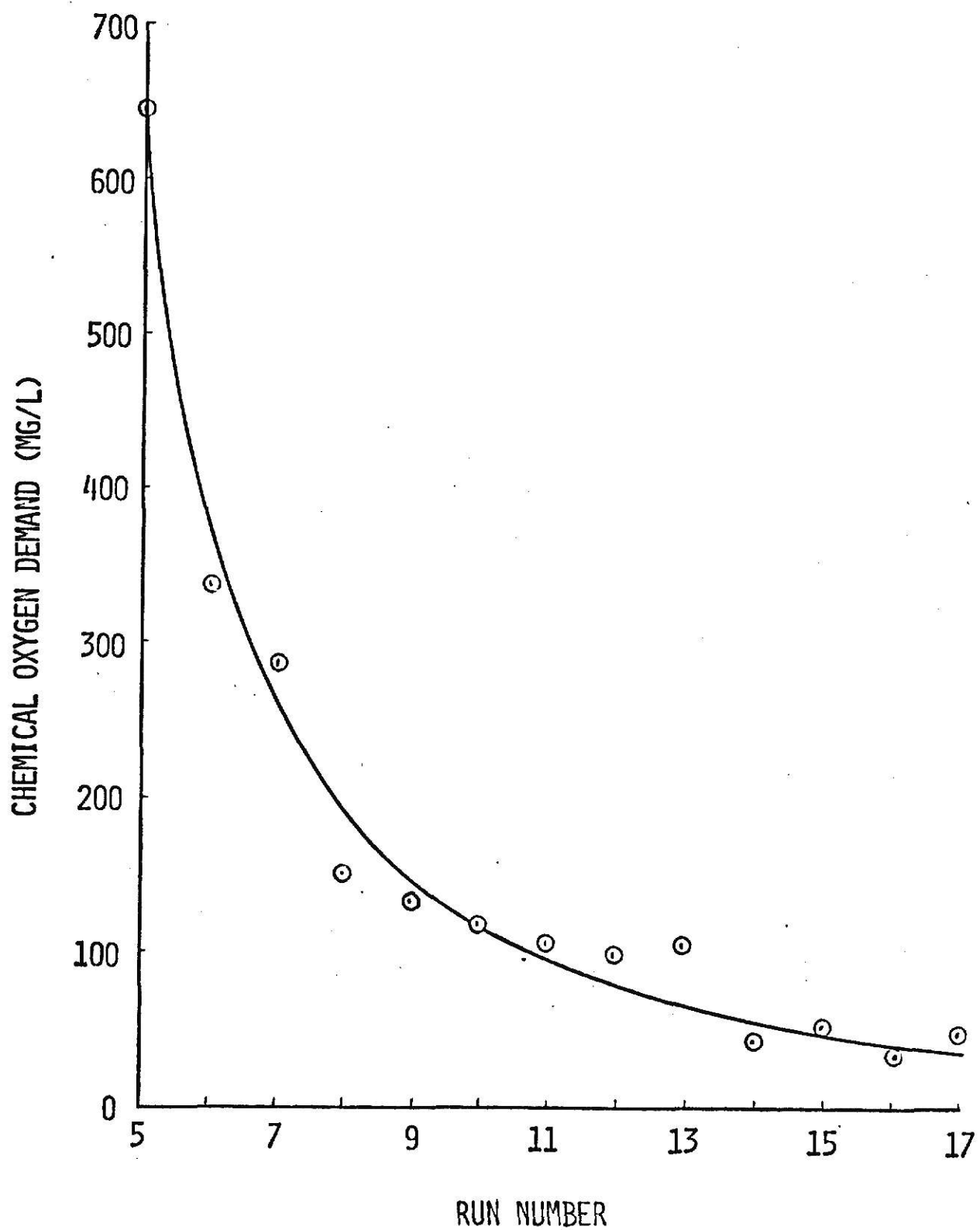


FIG. 4 AVERAGE CHEMICAL OXYGEN DEMAND OF
WATER SAMPLES PER TEST RUN

in the soil. During run 2 (Fig. 7) the nitrate concentration leveled out with most of the initial nitrates having moved out of the soil column (Fig. 7). The ammonia concentration during run 2 increased to 146 ppm at sample point 2. This was expected in the area directly below the manure layer. During runs 3, 4, and 5 the nitrate concentrations dropped to a constant level of 1-3 ppm for all the sampling points in the column. The ammonia concentration was at one of its highest levels during run 3 (196 ppm) and decreased to 106 ppm in run 5.

The nitrate nitrogen began increasing during run 6 (Fig.11) with the maximum concentration being 9.5 ppm. Sample points 8-13 showed no increase in nitrate nitrogen. The ammonia concentration was highest in the first 7 sample points. The maximum nitrate nitrogen concentration moved down to sample point 4 during run 10 (Fig.15) and the ammonia moved down to sample point 6. During the first 10 runs the ammonia moved ahead of the nitrate nitrogen. During run 11 (Fig.16) the maximum nitrate concentration moved ahead of the ammonia nitrogen. This same trend was exhibited in run 12, with the downward movement of both the ammonia and the nitrate nitrogen. The ammonia concentration in run 13 moved slightly ahead of the nitrate nitrogen. During run 13 the maximum ammonia concentration was at sample point 11 with the maximum nitrate nitrogen at sample point 10.

The maximum nitrate nitrogen and ammonia nitrogen moved to sample point 12 during run 14 (Fig.19). During run 15 (Fig.20) the major quantity of nitrate nitrogen moved out of the soil column. The ammonia decreased to negligible concentration in the first 11 sample points. The

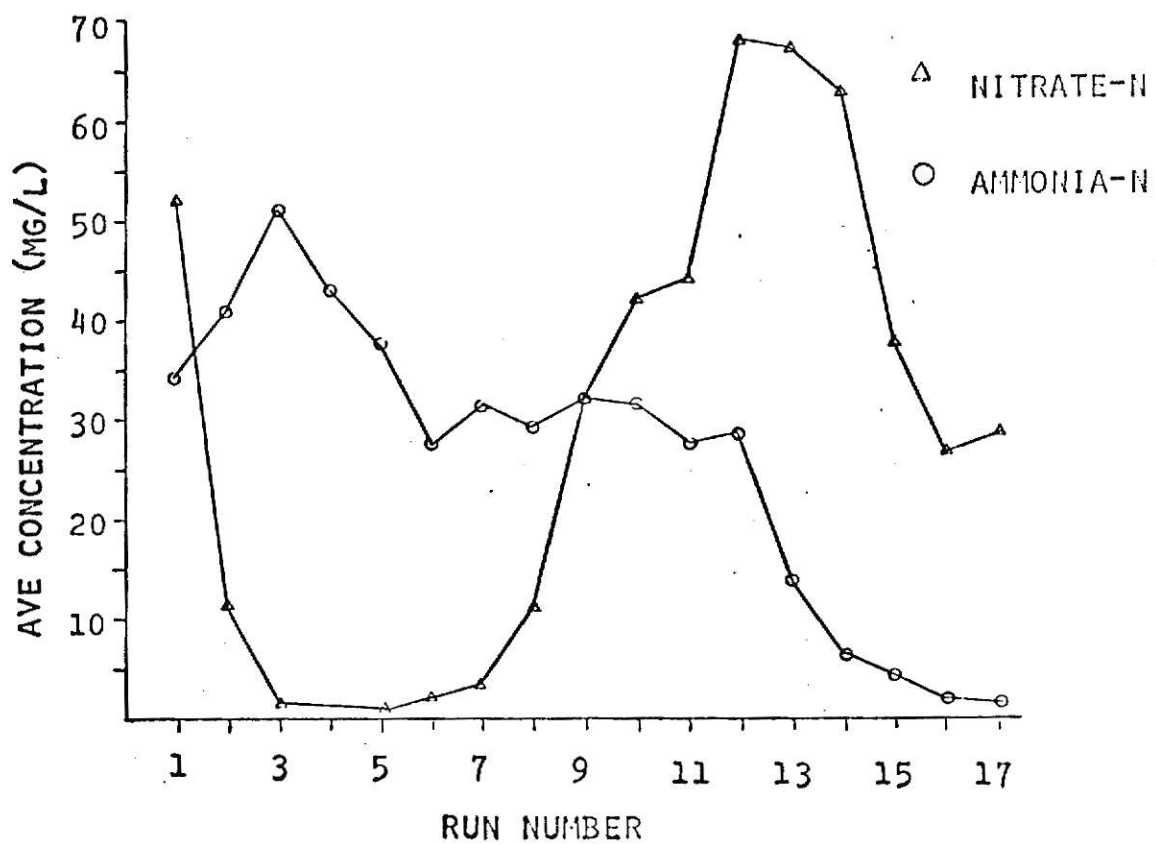


FIG. 5 AVERAGE NITRATE - N AND AVERAGE AMMONIA - N CONCENTRATION IN WATER SAMPLES FOR TEST RUNS 1 - 17.

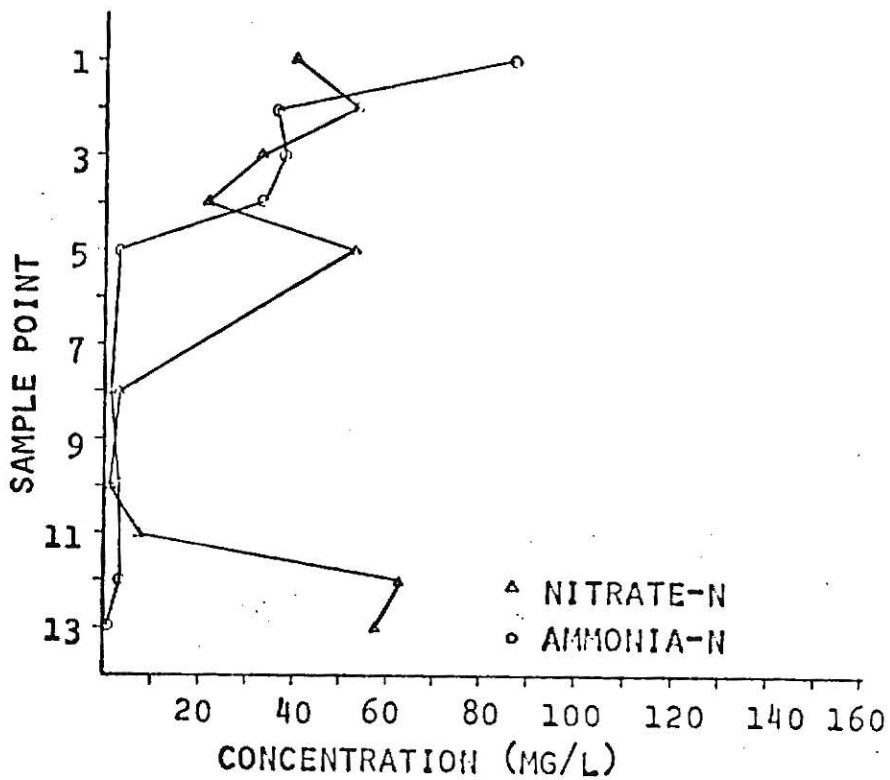


FIG. 6 NITRATE - N AND AMMONIA - N CONCENTRATIONS IN WATER SAMPLES FOR TEST RUN 1.

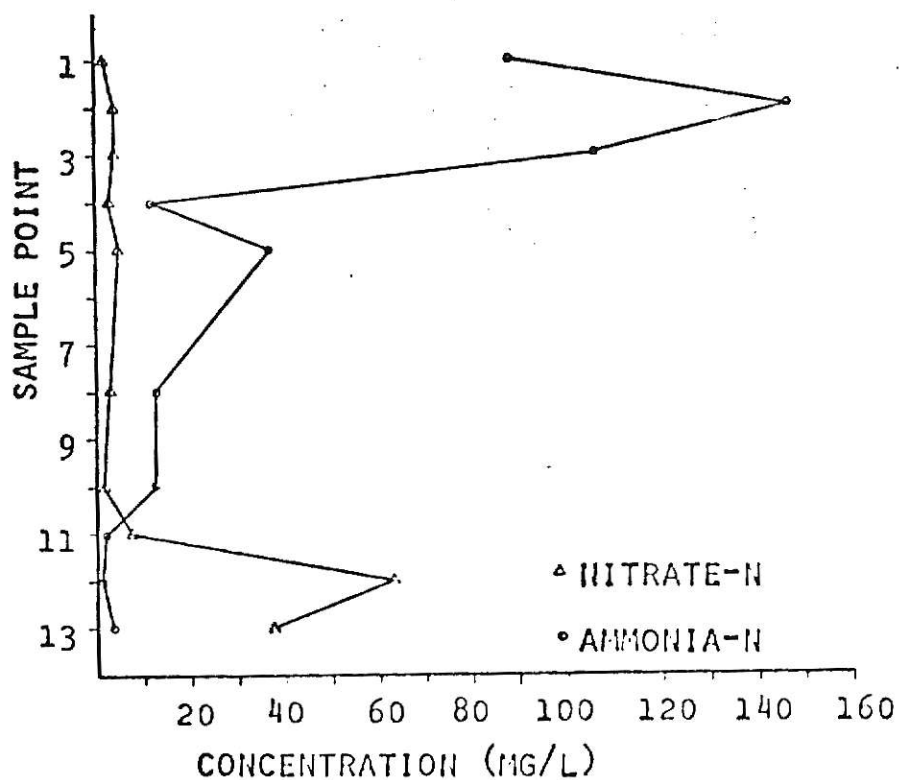


FIG. 7 NITRATE - N AND AMMONIA - N CONCENTRATIONS IN SAMPLES FOR TEST RUN 2.

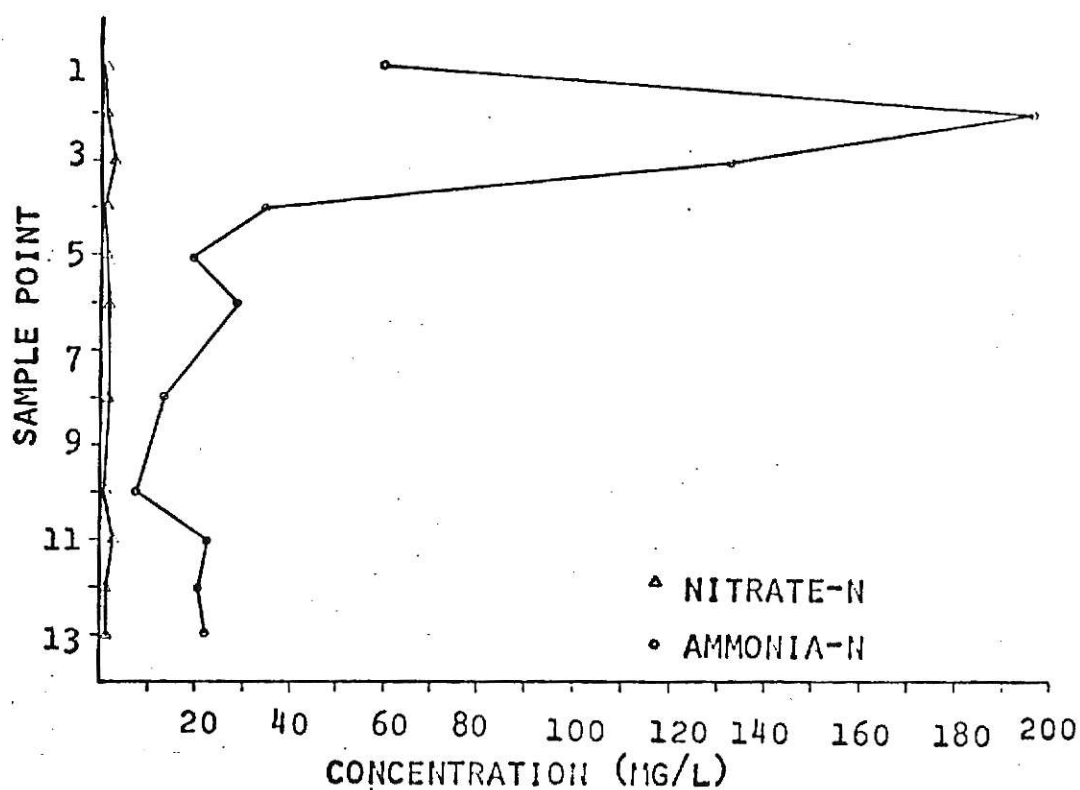


FIG. 8 NITRATE - N AND AMMONIA - N CONCENTRATIONS IN WATER SAMPLES FOR TEST RUN 3.

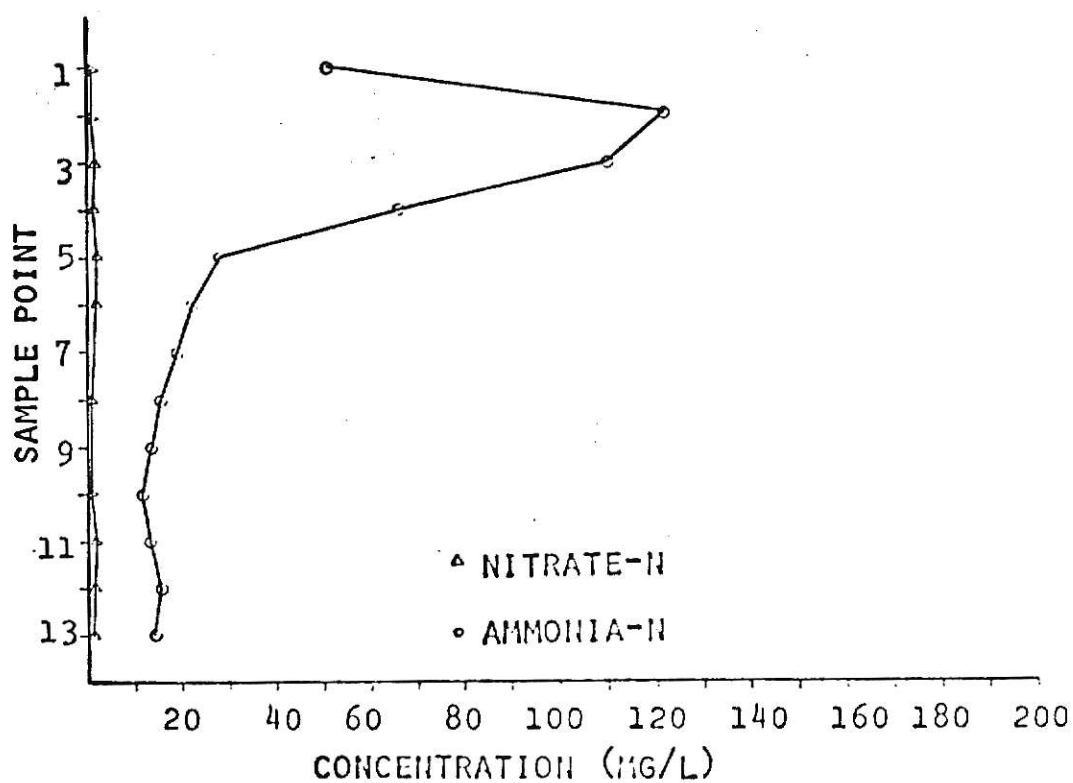


FIG. 9 NITRATE - N AND AMMONIA - N CONCENTRATIONS IN WATER SAMPLES FOR TEST RUN 4.

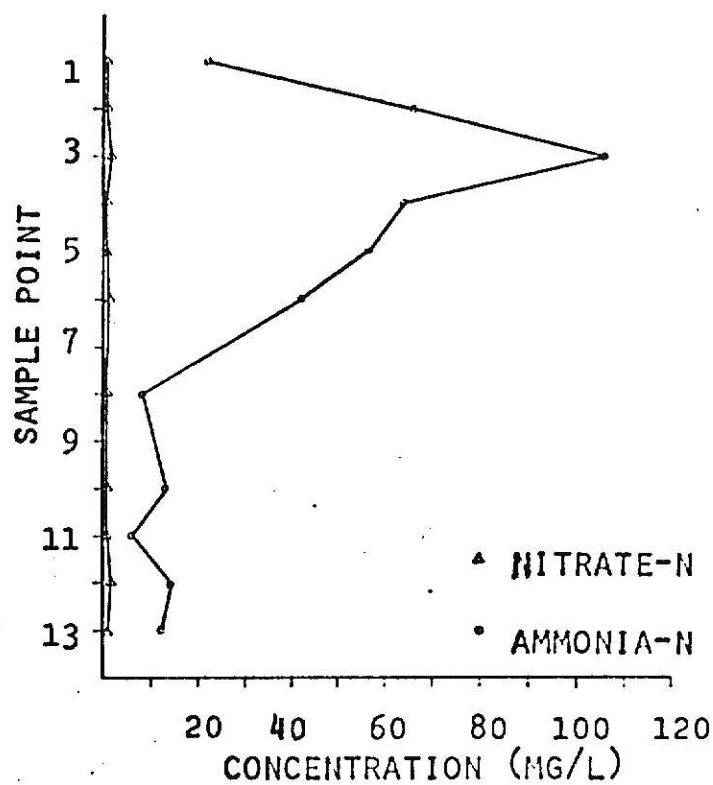


FIG. 10 NITRATE - N AND AMMONIA - N CONCENTRATIONS IN WATER SAMPLES FOR TEST RUN 5.

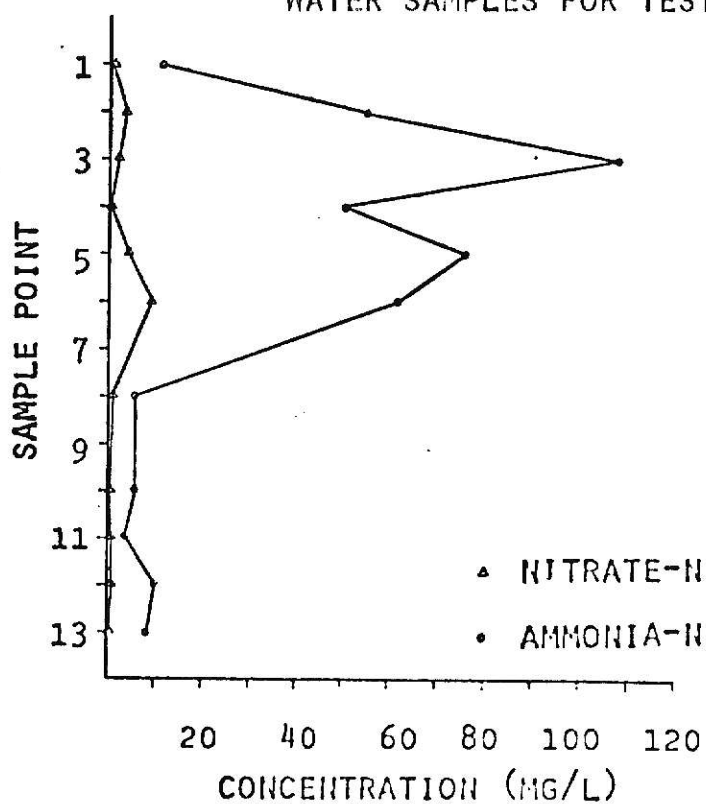


FIG. 11 NITRATE - N AND AMMONIA - N CONCENTRATIONS IN WATER SAMPLES FOR TEST 6.

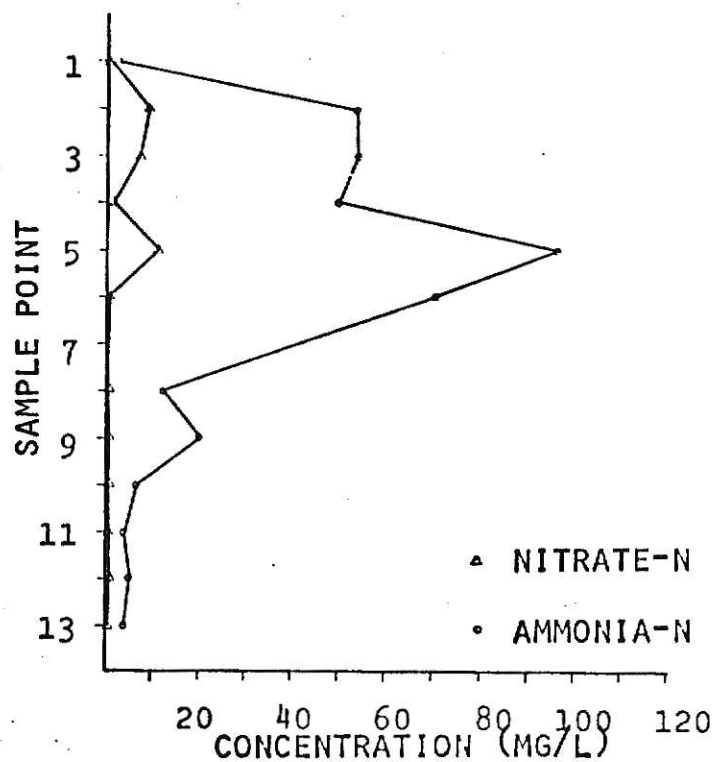


FIG. 12 NITRATE - N AND AMMONIA - N CONCENTRATIONS IN WATER SAMPLES FOR TEST RUN 7.

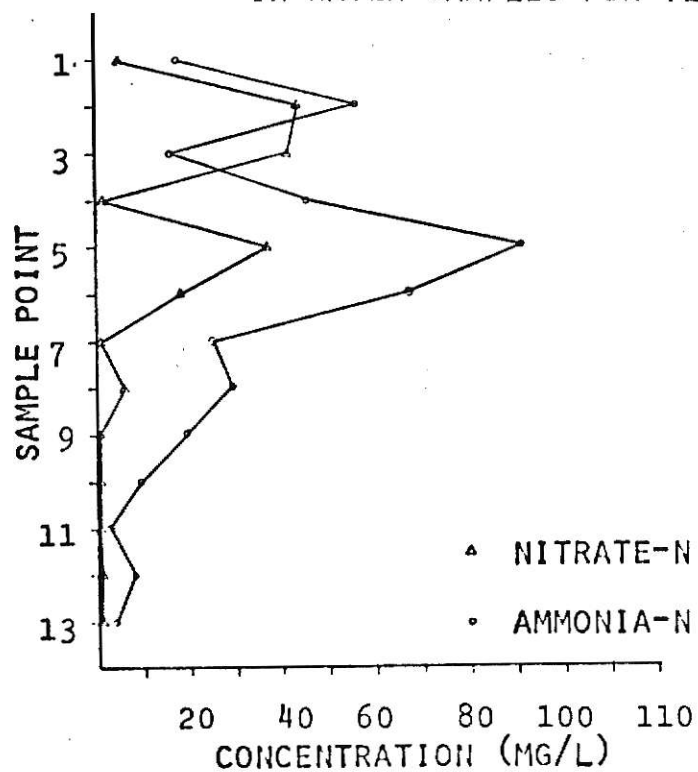


FIG. 13 NITRATE - N AND AMMONIA - N CONCENTRATION IN WATER SAMPLES FOR TEST RUN 8.

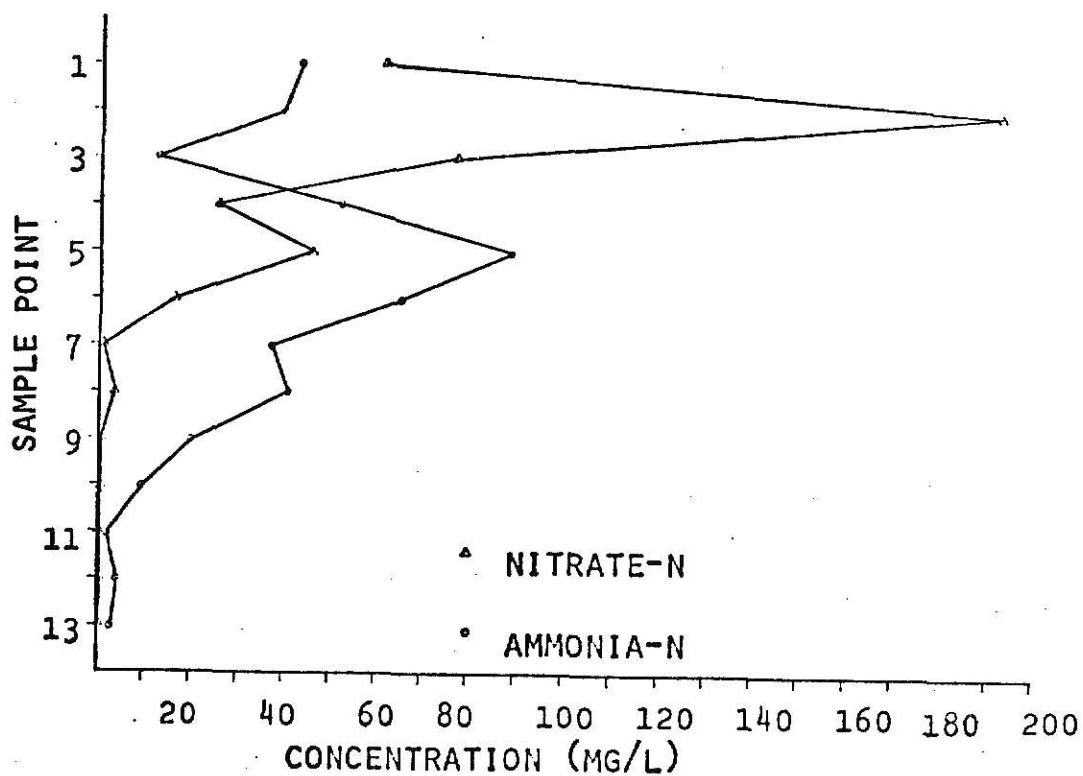


FIG. 14 NITRATE - N AND AMMONIA - N CONCENTRATIONS IN WATER SAMPLES FOR TEST RUN 9.

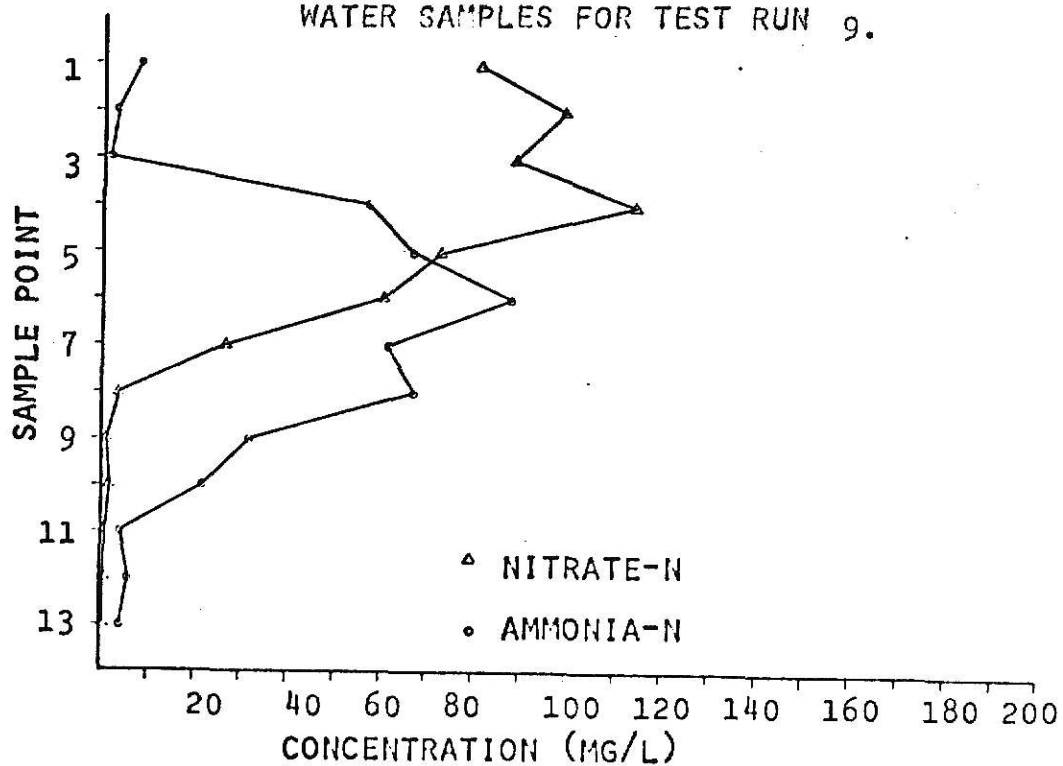


FIG. 15 NITRATE - N AND AMMONIA - N CONCENTRATIONS IN WATER SAMPLES FOR TEST RUN 10.

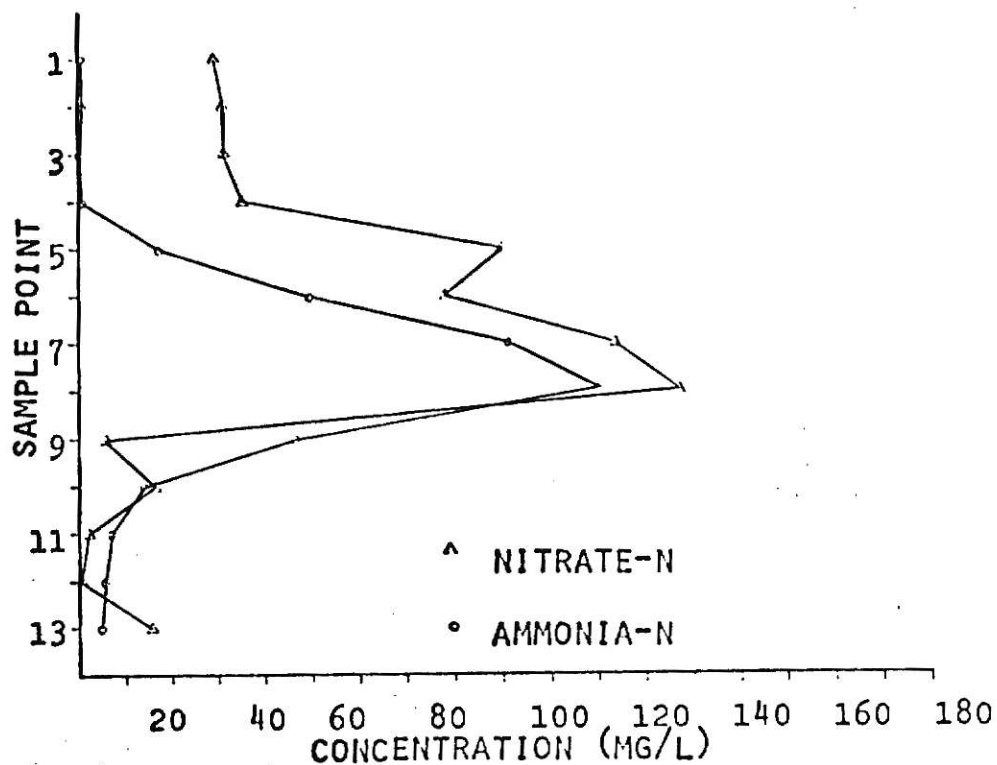


FIG. 16 NITRATE - N AND AMMONIA - N CONCENTRATIONS IN WATER SAMPLES FOR TEST RUN 11.

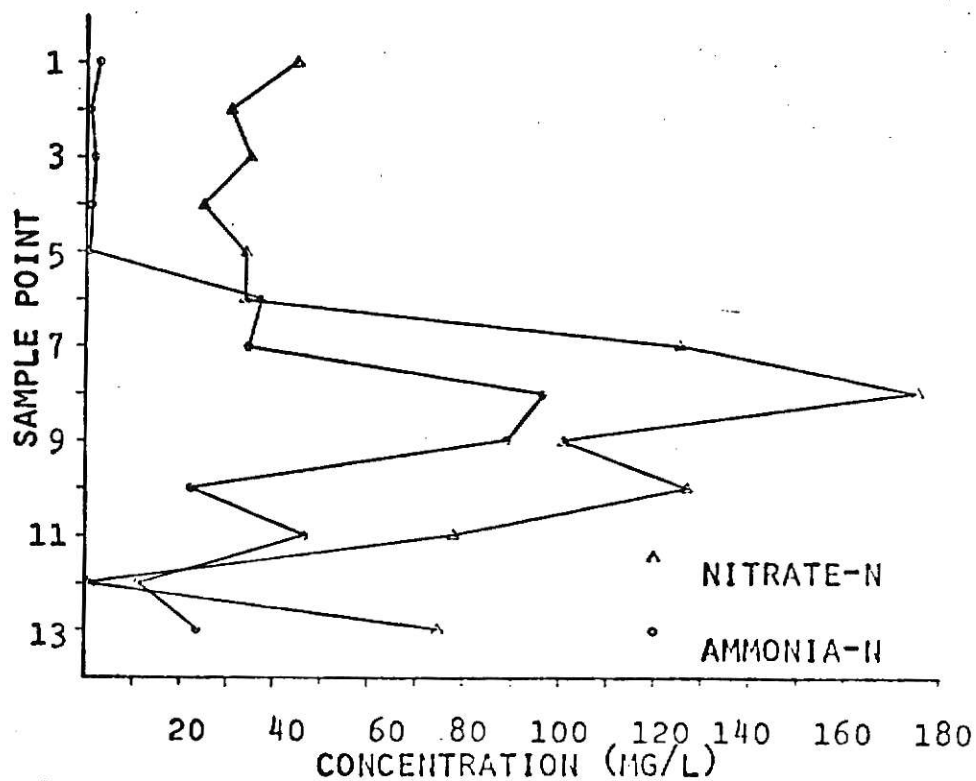


FIG. 17 NITRATE - N AND AMMONIA - N CONCENTRATIONS IN WATER SAMPLES FOR TEST RUN 12.

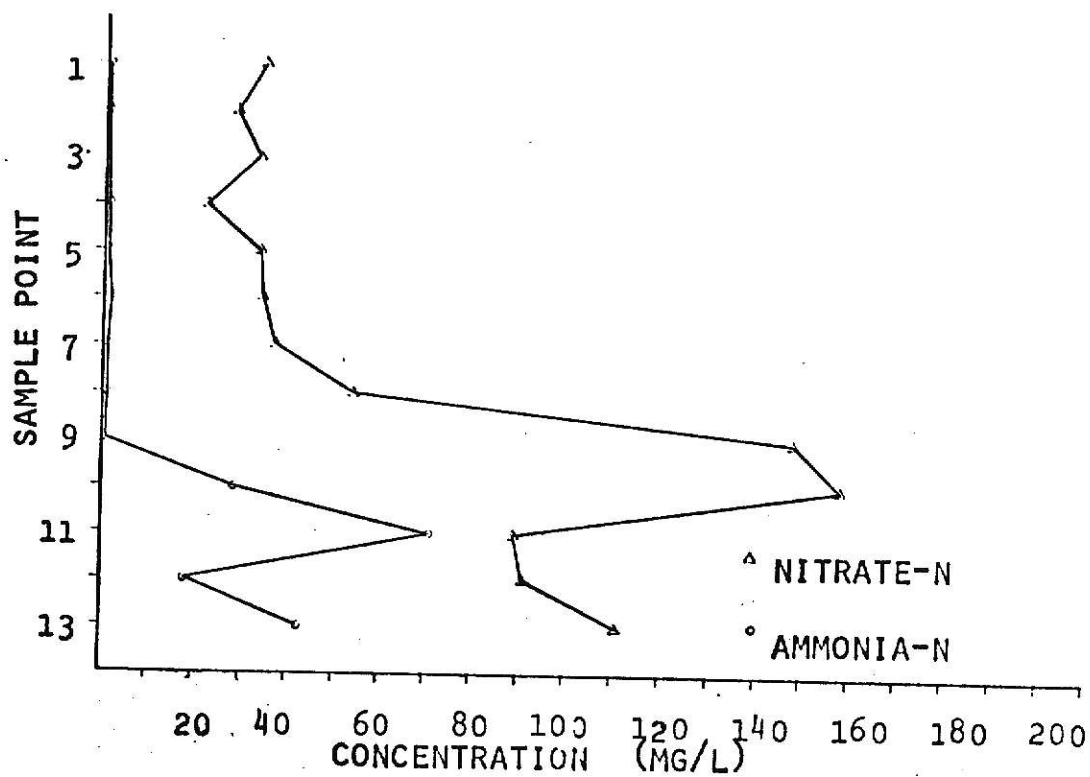


FIG. 18 NITRATE - N AND AMMONIA - N CONCENTRATIONS IN WATER SAMPLES FOR TEST RUN 13.

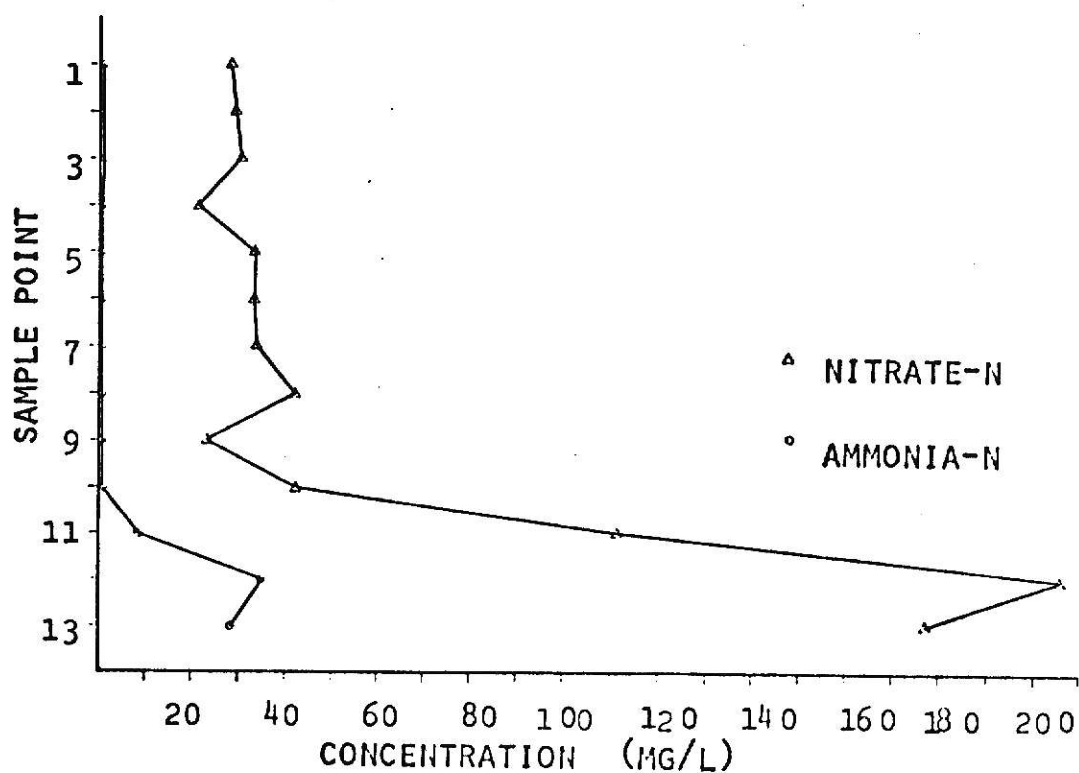


FIG. 19 NITRATE - N AND AMMONIA - N CONCENTRATIONS IN WATER SAMPLES FOR TEST RUN 14.

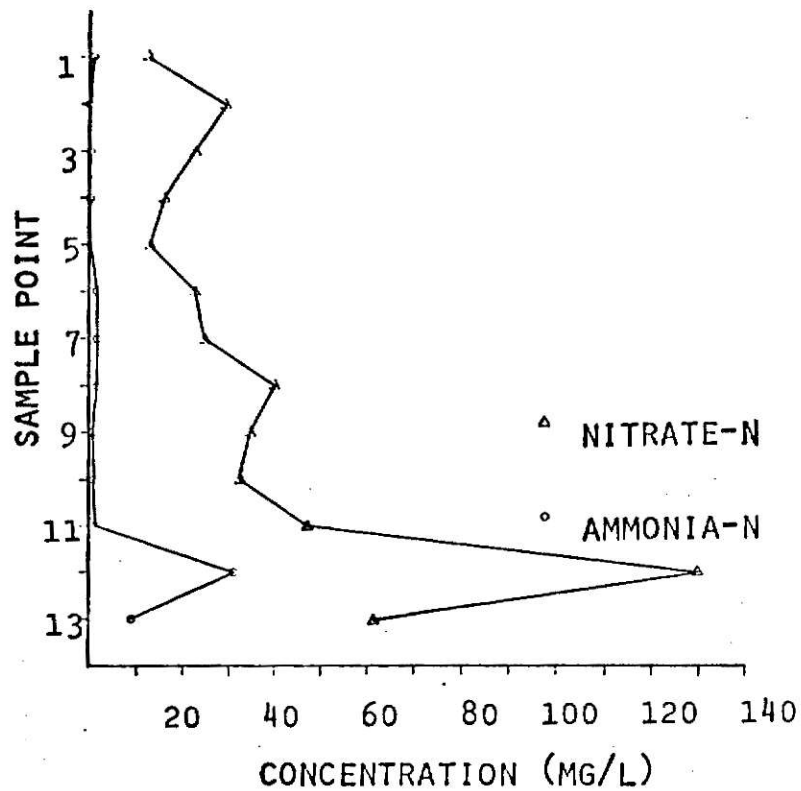


FIG. 20 NITRATE - N AND AMMONIA - N CONCENTRATIONS IN WATER SAMPLES FOR TEST RUN 15.

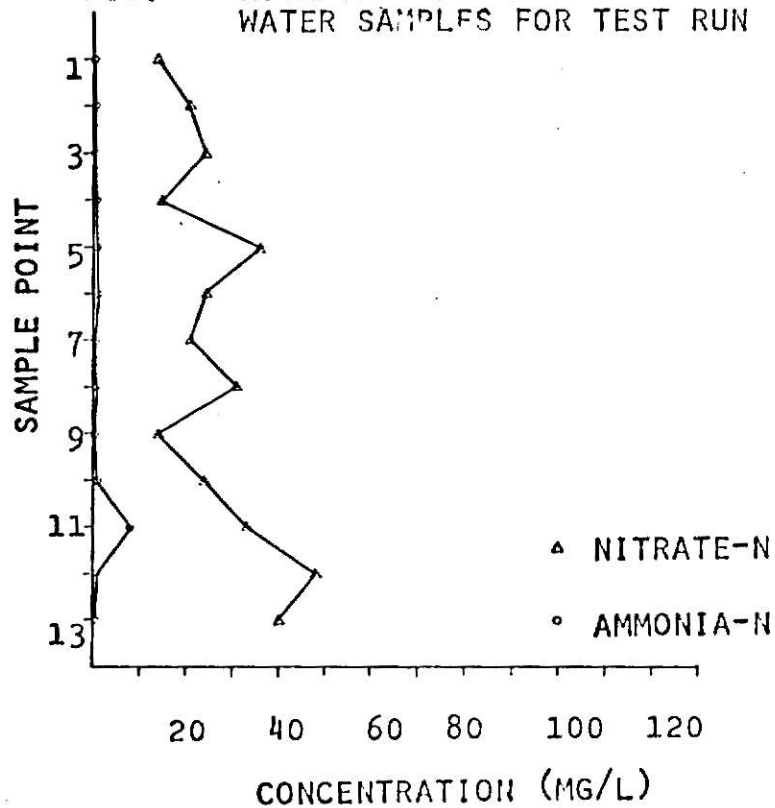


FIG. 21 NITRATE - N AND AMMONIA - N CONCENTRATIONS IN WATER SAMPLES FOR TEST RUN 16.

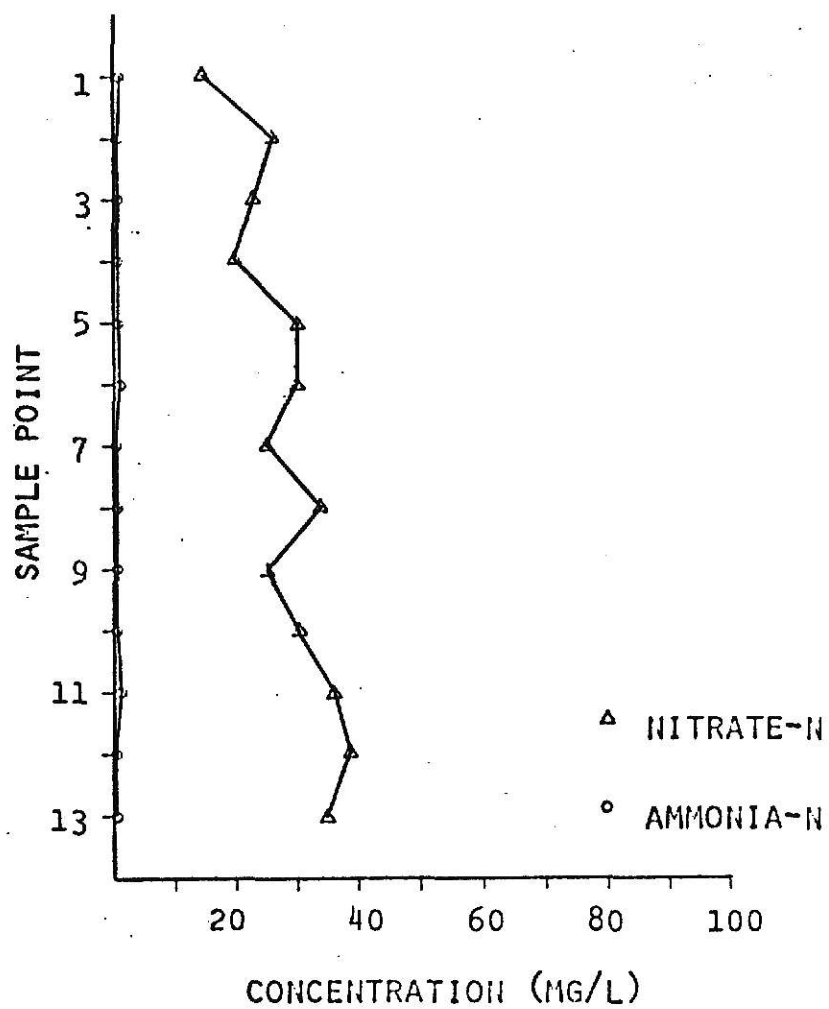


FIG. 22 NITRATE - N AND AMMONIA - N
CONCENTRATIONS IN WATER SAMPLES
FOR TEST RUN 17.

nitrate concentration decreased little in runs 16 (Fig.21) and 17 (Fig. 22). The concentrations leveled out so that the nitrate nitrogen plot was linear with respect to the sample points. The ammonia concentration dropped to an average concentration of less than 1 ppm during the last run (17).

This analysis of the water samples has indicated that the ammonia moved ahead of the nitrate nitrogen until run 11. During runs 12-17 the ammonia and nitrate nitrogen moved through and out of the soil column at about the same rate. The ammonia continually decreased as the runs continued.

The COD analysis of the water samples showed no decrease with (See Table 12) depth as Koelliker (14) described in his field study of the renovation of liquid swine wastes. The COD data taken with respect to depth was very erratic. The average decrease in COD per run (Fig.4) was expected because organic matter was being removed and none was being added.

Analysis of the gas data indicated that the gas concentration measurements were in error. The analysis indicated an excess of about 9 grams of nitrogen. The volume of gas being measured was large compared to the change in nitrogen concentration in the air. A small error in the nitrogen gas concentration measurement results in a large error when it is considered that only 0.79 grams of nitrogen were unaccounted for in the soil, manure, and leachate.

This leads us to the next step in this research. The atmosphere contains 78 percent nitrogen and 21 percent oxygen by volume. The literature indicates that a synthetic atmosphere should be used to replace the normal atmosphere.

EXPERIMENT 2

A trial run was made to evaluate the use of the Fisher/Hamilton Model 29 Gas Partitioner with a synthetic atmosphere of helium and oxygen (80% He, 20% O₂). The same equipment was used in the experiment as in the first experiment. The soil used in experiment 1 was removed from the soil column, sieved, dried and placed back into the soil column for experiment 2. Water samples were analyzed for nitrate, nitrite and ammonia as described in the procedures for experiment 1.

The initial composite soil sample had a nitrate concentration of 8.5 ppm, an ammonium concentration of 16.1 ppm and a total nitrogen content of 0.0298 percent. The final soil analysis showed that the average nitrate concentration was 1.83 ppm, the ammonium concentration was 15.9 ppm and the average total nitrogen content was 0.0356 percent (TABLE 14).

The chemical oxygen demand increased to 8344 ppm during run 3 (TABLE 13). The biological oxygen demand for run 3 was roughly 1200 ppm. The literature indicates that the COD-BOD ratio is in the range of 6 to 15. The ratio from run 3 was 6.9, which is within values given in the literature.

The soil initially contained 0.55 grams of nitrate nitrogen and 19.28 grams of total nitrogen (organic-N + ammonium-N) for a total of 19.83 grams of nitrogen. The manure initially contained 0.05 grams of nitrate nitrogen and 6.81 grams of total nitrogen (organic-N + ammonium-N) for a total of 6.86 grams. This gives a total for the manure and soil of 26.69 grams of nitrogen.

In the final soil analysis, the soil contained 0.128 grams of nitrate nitrogen and 23.03 grams of total nitrogen(organic-N + ammonium-N). The manure contained 2.80 grams of nitrogen in the final analysis. This gives a total of 25.95 grams of nitrogen. The difference between the initial and final content of the soil and manure was 0.74 grams. This means that the rest of the nitrogen was lost due to denitrification, ammonia volatilization, or leaching.

An analysis of the water showed that 0.210 grams of nitrate nitrogen, 0.176 grams of ammonia nitrogen, and 0.008 grams of nitrite nitrogen were lost due to leaching (0.400 grams total). Approximately one-half of the nitrogen was lost due to volatilization and denitrification (46 percent). The remaining 54 percent was leached out of the soil column.

During the second experiment the chromatograph was not able to completely separate the components in the gas that was injected into it. There was also a very large difference in the input and output volumes from the soil column. It is possible that a leak developed because the peak heights from the chromatograph during run 3 looked like those of atmospheric air.

CONCLUSIONS

The objectives set out for in experiment 1 were not reached. A nitrogen balance was not possible. The gas measurement system provided a satisfactory balance of input and output gases for experiment 1, but not experiment 2.

A very small amount of the nitrogen in the system was lost due to

denitrification (0.79 grams - experiment 1). The system was held at aerobic conditions for the 17 test runs. This was interpreted to mean that no anaerobic pockets existed. The chromatograph used for the analysis of the gases did not have the necessary accuracy to correctly measure the small changes in nitrogen concentration. The soil system in the first experiment was held at near aerobic conditions for the entire experiment. This was why the amount of denitrification was low as compared to systems having an anaerobic period and then an aerobic period.

The movement of the ammonia is very much like that described by George (10). The nitrates followed the ammonia through the first 10 runs. During the remaining 7 runs the nitrate and ammonia moved together with the nitrate nitrogen at times moving ahead of the ammonia nitrogen.

During the second experiment the system was held at anaerobic conditions. The soil-manure system lost 0.34 grams of nitrogen due to denitrification or ammonia volatilization.

It has been demonstrated that the gas chromatograph is not capable of accurately separating the gases involved so that a nitrogen balance could be attained. The next step would be to use a synthetic atmosphere (He , O_2) and analyze the output gas on a mass spectrometer.

SUGGESTIONS FOR FURTHER STUDY

The equipment used in this research has shown that it can be used

to quantify the movement of nitrogen in the soil. It can also be used to measure the volume of input and output of gas from a soil system.

The gas chromatograph should be removed from the circuit. The gas sample should then be taken as a composite from the air displacement column and the sample analyzed with a more sophisticated gas chromatograph or a mass spectrometer. This is needed to show that the system is capable of providing a total nitrogen balance.

The next step would be to set up a series of 3 columns with 3 loading rates of manure with one application rate of water or to hold the manure loading rate constant and vary the water application rate. The present equipment with modifications could be used. The available equipment would also allow the water table to be varied. The various rates of manure application and water application would help to make predictions concerning movement of nitrate into the ground water. This data could be correlated with field studies involving various manure application rates.

It has been shown in research with liquid waste treatment systems that soluble energy sources for the bacteria usually are the limiting factor in denitrification. If this is true, it is possible that bacteria denitrifying animal wastes in the soil are limited in available energy. The rate of oxygen uptake would be an indicator of the energy available. Several methods exist for determining the oxygen uptake rate. They include continuous monitoring of dissolved oxygen uptake with a dissolved oxygen probe, the Hach BOD manometer, or the use of a Warburg manometer.

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APPENDIX I

TABLE 3

Soil Analysis Data

Sample depth in soil column (inches)	pH	OM, %	Total N %	NH ₄ ⁺ - N ppm	NO ₃ ⁻ - N ppm
Manure sample before testing	---	5.0	1.66	3354.9	120.8
Composite soil sample before testing	7.6	0.8	0.0275	2.34	7.47
Samples after 17 test runs					
0-4	7.6	0.90	0.020	9.6	3.7
4	6.9	4.40	0.378	60.7	58.4
4-8	7.8	0.70	0.031	6.7	3.9
8-12	7.9	0.60	0.029	5.8	3.6
12-16	8.0	0.65	0.033	6.3	4.0
16-20	8.0	0.60	0.035	8.2	5.2
20-24	8.0	0.50	0.028	4.3	4.9
24-28	7.9	0.50	0.036	5.4	5.7
28-32	8.0	0.85	0.031	5.8	5.7
32-36	8.0	0.55	0.027	5.4	7.0
36-40	8.1	0.55	0.033	4.8	5.8
40-44	8.0	0.90	0.034	5.2	7.3
48-52	8.0	0.70	0.026	4.5	6.3

TABLE 4
Ammonia Concentrations
(mg/l - N)

Run#	Sampling Positions													Total	Ave
	1	2	3	4	5	6	7	8	9-	10	11	12	13		
1	87.00	36.00	38.00	33.00	3.20	---	---	1.40	---	2.80	---	3.40	0.60	429.4	42.94
2	88.00	146.00	106.00	11.00	37.00	---	---	13.00	---	13.00	2.20	1.40	3.40	446.0	40.55
3	60.00	196.00	132.00	35.00	20.00	29.00	---	13.50	---	8.00	22.50	21.00	22.50	559.5	50.86
4	51.00	122.00	110.00	66.70	28.00	22.00	---	15.50	---	11.50	13.00	16.50	14.30	470.5	42.77
5	22.00	66.00	106.00	64.00	57.00	42.00	---	8.00	---	13.00	5.90	14.50	12.50	410.9	37.35
6	11.00	55.00	108.00	50.00	76.00	61.50	---	5.50	---	5.50	3.50	10.00	8.50	394.5	35.86
7	2.00	54.00	54.00	50.00	96.00	70.00	---	12.00	20.00	6.50	3.25	5.00	3.25	376.0	31.33
8	17.50	56.50	16.00	45.50	91.00	67.00	25.00	23.00	19.00	9.00	2.50	3.50	3.25	378.75	29.13
9	43.00	39.00	12.00	52.00	88.50	65.00	37.00	40.50	20.00	9.50	1.75	3.75	2.50	414.5	31.88
10	7.50	2.50	1.00	52.00	67.00	88.50	61.00	67.00	31.00	10.50	3.75	5.50	4.25	401.5	30.88
11	0.50	1.00	0.50	17.00	49.00	17.00	91.00	110.00	47.00	14.50	7.00	6.50	5.00	349.5	26.88

12	3.00	1.00	1.50	1.50	0.75	37.50	34.50	97.00	89.00	22.00	40.50	11.00	24.00	363.25	27.94
13	0.75	0.50	0.75	0.50	1.00	1.50	0.75	0.50	0.25	29.00	71.00	17.50	43.00	167.00	12.84
14	0.75	0.50	0.75	0.50	0.75	0.25	0.75		0.25		8.75	35.00	28.00	75.75	5.83
15	0.50	0.25				1.50	2.00	1.50	1.00	0.50	1.00	31.00	9.00	48.25	3.71
16	0.60	0.30	0.50	0.20	---	1.20	0.70	0.50	0.50	0.60	0.90	8.00	0.20	14.20	1.18
17	1.40	0.90	0.65	0.50	---	0.90	0.40	0.50	0.80	0.65	1.30	0.20	0.05	8.25	0.69

Total

396.5 777.45 687.65 462.99 583.20 536.85 252.60 409.40 228.80 156.55 188.80 193.75 433.30 5307.75

Average

23.32 45.73 40.45 27.23 38.88 35.79 25.26 24.08 20.80 9.21 11.80 11.39 22.81

TABLE 5
Ammonia Nitrogen Totals
(mg - N)

Sampling Positions														
Run#	1	2	3	4	5	6	7	8	9	10	11	12	13	Total
1	3.220	1.050	1.120	1.220	0.038	—	—	0.032	—	0.077	—	0.137	2.163	9.357
2	1.810	7.370	2.490	0.237	0.648	—	—	0.234	—	0.358	0.118	0.071	10.570	23.906
3	1.500	9.210	4.220	1.120	0.580	0.261	—	0.499	—	0.268	0.765	0.724	81.900	101.047
4	1.370	5.120	2.640	2.000	0.896	0.286	—	0.341	—	0.345	0.283	0.577	48.048	61.906
5	0.528	2.440	2.300	1.152	1.311	0.546	—	0.256	—	0.286	0.123	0.551	44.000	53.493
6	0.330	1.700	2.910	1.050	2.500	1.292	—	0.171	—	0.099	0.059	0.300	28.940	39.351
7	0.013	1.080	1.458	1.700	2.976	1.575	—	0.522	0.212	0.156	0.085	0.197	11.586	21.560
8	0.123	0.904	0.368	1.638	3.003	1.943	0.725	0.989	0.513	0.279	0.065	0.113	10.950	21.613
9	1.118	0.760	0.630	1.716	2.610	2.015	1.184	1.944	0.800	0.199	0.027	0.156	8.50	21.579
10	0.266	0.066	0.038	2.230	2.881	3.186	3.141	3.417	1.240	0.347	0.053	0.287	13.855	31.007
11	0.016	0.020	0.024	0.023	0.595	0.833	3.185	3.960	0.802	0.493	0.189	0.338	17.070	27.548

12	0.089	0.022	0.051	0.058	0.029	0.956	1.156	4.365	1.157	0.693	1.863	1.550	78.240	90.229
13	0.017	0.011	0.023	0.016	0.038	0.035	0.021	0.023	0.004	1.044	2.340	0.805	143.190	147.567
14	0.021	0.011	0.018	0.015	0.020	0.005	0.007		0.003		0.296	1.663	94.360	96.410
15	0.013	0.005				0.033	0.056	0.060	0.007	0.010	0.033	1.457	30.420	32.081
16	0.014	0.009	0.010	0.010	---	0.028	0.027	0.021	0.009	0.020	0.043	0.408	0.680	1.274
17	0.034	0.027	0.014	0.014	---	0.017	0.018	0.021	0.007	0.018	0.068	0.011	0.170	0.417

Total

10.482	29.805	18.314	14.192	18.125	13.011	9.520	16.805	4.754	4.692	6.410	9.345	624.932		
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TABLE 6

Nitrate Concentrations
(mg/l - N)

Sampling Positions

Run#	1	2	3	4	5	6	7	8	9	10	11	12	13	Total	Ave
1	40.00	53.50	33.13	21.00	53.00	—	—	120.00	—	118.00	—	21.00	58.00	521.73	52.17
2	2.15	4.45	4.10	2.80	5.20	—	—	3.00	—	1.40	7.40	63.50	37.00	134.40	12.21
3	1.00	1.25	2.95	0.85	1.30	1.80	—	1.80	—	1.10	2.75	1.35	1.25	17.40	1.58
4	0.65	0.85	1.85	1.10	1.90	1.75	—	0.95	—	0.90	1.20	1.50	1.15	13.80	1.25
5	0.80	0.90	1.60	0.65	0.85	1.10	—	0.65	—	0.75	0.85	1.40	0.90	10.45	0.95
6	0.55	3.40	1.83	0.30	4.55	9.50	—	1.20.	—	0.65	0.45	0.90	0.55	23.88	2.17
7	0.45	9.15	7.65	1.45	16.25	0.96	—	0.45	0.60	0.65	0.60	0.80	0.45	39.46	3.29
8	5.00	44.00	41.50	1.25	37.50	18.75	0.70	6.20	0.45	0.45	0.45	0.70	0.35	157.30	12.10
9	60.75	192.50	77.63	24.90	45.90	21.00	0.70	3.20	0.65	0.30	0.40	0.45	0.10	428.48	32.96
10	80.60	98.80	88.40	114.40	71.50	60.00	25.65	2.70	1.05	1.45	0.35	0.30	0.10	545.30	41.95
11	29.03	31.05	31.05	35.77	89.70	77.90	113.57	127.05	5.40	16.20	1.95	0.45	15.40	574.52	44.19

12	45.10	31.02	35.20	25.30	34.10	34.10	126.10	175.5	101.40	127.80	78.00	0.10	75.40	889.42	68.42
13	34.10	28.05	33.00	22.00	33.55	34.10	36.30	54.23	117.90	158.10	89.25	91.00	111.60	873.18	67.17
14	28.05	29.15	30.25	20.90	33.55	33.00	33.55	41.80	22.55	42.90	111.60	207.00	177.48	811.78	62.45
15	12.60	29.10	22.80	16.20	13.00	22.80	24.90	39.60	34.50	32.10	46.75	130.00	61.05	485.40	37.34
16	14.30	20.90	23.65	15.40	36.30	24.20	20.90	30.80	14.30	23.65	33.00	48.40	39.60	345.40	26.57
17	14.85	26.95	23.10	19.80	29.70	29.70	25.30	34.10	25.30	30.25	36.30	38.50	35.20	369.05	28.39

Total

369.98	605.02	459.69	324.07	507.85	370.66	407.67	643.23	354.10	556.65	411.30	607.65	623.08
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Average

21.76	35.59	27.04	19.06	29.87	24.71	40.77	37.83	32.19	32.74	25.71	35.74	32.79
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TABLE 7

Nitrate Nitrogen Totals
(mg-N)

Sampling Positions

Run#	1	2	3	4	5	6	7	8	9	10	11	12	13	Total
1	1.180	1.520	0.974	0.777	0.636	---	---	2.700	---	3.250	---	0.875	236.030	248.263
2	0.014	0.224	0.096	0.060	0.910	---	---	0.054	---	0.039	0.053	3.238	115.070	119.815
3	0.025	0.058	0.094	0.027	0.038	0.016	---	0.067	---	0.037	0.093	0.047	9.100	9.603
4	0.019	0.038	0.014	0.033	0.062	0.023	---	0.028	---	0.270	0.030	0.053	3.864	4.464
5	0.019	0.033	0.037	0.012	0.020	0.014	---	0.021	---	0.017	0.018	0.053	3.168	3.412
6	0.017	0.105	0.049	0.006	0.150	0.200	---	0.037	---	0.012	0.008	0.027	1.873	2.484
7	0.003	0.102	0.207	0.049	0.264	0.021	---	0.013	0.007	0.016	0.014	0.032	1.610	2.338
8	0.035	0.704	0.954	0.045	1.237	0.544	0.020	0.267	0.012	0.014	0.012	0.025	1.180	5.049
9	1.579	3.754	4.107	0.822	1.358	0.651	0.022	0.154	0.026	0.006	0.006	0.019	0.340	12.844
10	2.861	2.618	3.359	4.919	3.075	2.160	1.326	0.138	0.042	0.005	0.005	0.016	0.326	20.894
11	0.899	0.621	1.490	1.645	3.139	1.324	3.975	4.573	0.092	0.052	0.052	0.023	52.591	70.975

12	1.331	0.667	1.197	0.974	1.330	0.870	4.224	7.898	1.318	4.026	3.588	0.020	245.804	273.247
13	0.750	0.617	0.990	0.704	1.275	0.784	1.016	2.495	2.071	5.692	2.945	4.186	371.628	395.153
14	0.785	0.612	0.726	0.627	0.872	0.677	0.939	2.132	0.293	1.287	4.129	9.833	598.108	621.020
15	0.315	0.524	0.570	0.680	0.169	0.502	0.689	1.584	0.242	0.642	1.543	6.110	206.340	219.910
16	0.349	0.606	0.473	0.385	0.109	0.557	0.794	1.263	0.257	0.804	1.584	2.468	134.640	144.283
17	0.356	0.809	0.508	0.475	0.089	0.546	1.164	1.398	0.229	0.847	1.924	2.110	119.328	129.782

Total

10.861	13.612	15.875	12.240	14.733	8.889	14.169	24.822	4.588	17.558	16.005	29.135	2101.046
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TABLE 8

Nitrite Nitrogen Concentration
(mg/l - N)

Sampling Positions

Run#	1	2	3	4	5	6	7	8	9	10	11	12	13	Total	Ave
1				0.08		---	---	---	---		---		0.18	0.51	0.051
2			0.38			---	---	---	---	0.10	0.10		0.18	1.01	0.092
3							---		---						
4							---		---						
5							---		---						
6							---		---						
7							---								
8															
9		1.00	0.63											1.63	0.125
10	1.00			0.90	0.85	0.25		0.35						3.35	0.258
11					0.08		0.25	0.35						0.68	0.052

12									0.18						0.15	0.33	0.025
13	0.18	0.10	0.13	0.20	0.05	0.10	0.85	56.61	58.28	87.50	10.00	0.30	214.30	16.48			
14										0.25	0.69	1.13	2.07	0.159			
15											0.23	0.13	0.36	0.028			
16											0.20		0.20	0.015			
17											0.08		0.08	0.006			

Total	1.0	1.175	1.11	1.11	1.13	0.30	0.35	1.73	56.61	58.38	87.85	11.20	2.57				
Average	0.059	0.069	0.065	0.065	0.066	0.020	0.023	0.102	3.774	3.434	5.49	0.659	0.135				

TABLE 9

Nitrite Nitrogen Totals
(mg - N)

Sampling Positions

Run#	1	2	3	4	5	6	7	8	9	10	11	12	13	Total
1				0.003		---	---		---		---		0.718	0.722
2			0.009			---	---		---	0.028	0.005		0.544	0.588
3							---		---					
4							---		---					
5							---		---					
6							---		---					
7							---							
8														
9		0.019	0.033											0.052
10	0.036			0.039	0.037	0.009		0.018						0.139
11					0.003		0.009	0.013					0.512	0.537

12									0.008							0.408	0.416
13	0.004	0.003	0.004	0.008	0.001	0.003	0.039	0.792	2.098	2.888	0.460	0.999	7.299				
14										0.009	0.033	3.790	3.832				
15											0.011	0.423	0.434				
16											0.010		0.010				
17											0.004		0.004				
<hr/>																	
Total	0.036	0.023	0.045	0.046	0.041	0.010	0.012	0.078	0.792	2.126	2.902	0.518	7.397				

TABLE 10

Total Concentration of Nitrogen
(mg/l - N)

Sampling Positions

Run#	1	2	3	4	5	6	7	8	9	10	11	12	13	Total	Ave
1	127.0	89.50	71.13	54.08	56.20	---	---	121.40	---	120.80	---	24.40	58.78	723.29	72.33
2	90.15	150.45	110.48	13.80	42.20	---	---	16.00	---	14.50	9.70	64.90	40.58	552.76	52.28
3	61.00	197.25	134.95	33.85	21.30	30.80	---	15.30	---	9.10	23.25	22.35	23.75	572.90	52.08
4	51.65	122.85	111.85	66.80	29.90	23.75	---	16.45	---	12.40	14.20	18.00	15.45	483.30	43.94
5	22.80	66.90	107.60	64.65	57.85	43.10	---	8.65	---	13.75	6.75	15.90	13.40	421.35	38.30
6	11.55	58.40	109.83	50.30	80.55	71.00	---	6.70	---	6.15	3.95	10.90	9.05	418.38	38.03
7	2.45	63.15	61.65	51.45	112.25	70.96	---	12.45	20.60	7.15	3.85	5.80	3.70	415.46	34.62
8	22.50	100.50	57.50	46.75	128.50	85.75	25.70	29.20	19.45	9.45	2.95	4.20	3.60	536.05	41.23
9	103.75	232.50	90.26	76.90	134.40	86.00	37.70	43.70	20.65	9.80	2.15	4.20	2.60	844.61	64.97
10	89.10	101.30	89.40	167.30	139.35	148.75	86.65	70.05	32.40	11.95	4.10	5.80	4.35	950.50	73.12
11	29.53	32.05	31.55	36.27	106.78	126.90	204.82	237.40	52.75	30.70	8.95	6.95	20.40	925.05	71.16

12	48.10	32.02	36.70	26.80	34.85	71.60	160.60	272.68	190.58	149.80	118.50	11.40	99.55	1253.18	96.40
13	34.85	28.73	33.85	22.63	34.75	35.65	37.15	55.55	149.00	245.38	247.75	119.50	154.90	1199.69	92.28
14	28.80	29.65	31.00	21.40	34.30	33.25	33.80	41.80	22.80	42.90	120.60	242.69	206.61	889.60	68.43
15	13.10	29.35	22.80	16.20	13.00	24.30	26.90	41.10	35.50	32.60	47.75	161.13	70.18	533.91	41.07
16	14.90	21.20	24.15	16.60	36.30	25.40	21.60	31.30	14.80	24.25	33.90	56.60	39.80	360.80	27.70
17	16.25	27.85	23.75	20.30	29.70	30.60	25.70	34.60	26.10	30.90	37.60	38.78	35.25	377.38	29.03

Total

767.48	1383.65	1148.45	786.08	1092.18	907.81	660.62	1054.33	584.63	771.58	685.95	813.50	801.95
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Average

45.15	81.39	67.56	46.24	64.25	60.52	66.06	62.02	53.15	45.40	42.87	47.85	47.17
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TABLE 11

Nitrogen Totals
(mg - N)

Sampling Positions

Run#	1	2	3	4	5	6	7	8	9	10	11	12	13	Total	Ave
1	4.700	2.570	2.094	2.000	0.674	---	---	2.732	---	3.327	---	1.012	239.211	258.32	28.702
2	1.854	7.594	2.595	0.297	1.558	---	---	0.288	---	0.425	0.177	3.310	126.180	144.282	14.428
3	1.525	9.268	4.314	1.147	0.618	0.277	---	0.566	---	0.305	0.859	0.771	91.000	110.650	10.059
4	1.389	5.158	7.842	2.033	0.958	0.309	---	0.369	---	0.345	0.313	0.630	51.912	71.258	6.478
5	0.547	2.473	2.337	1.164	1.331	0.560	---	0.277	---	0.303	0.141	0.604	47.168	56.905	5.173
6	0.347	1.805	2.959	1.056	2.650	1.492	---	0.208	---	0.111	0.067	0.327	30.813	41.835	3.803
7	0.016	1.182	1.665	1.749	3.240	1.596	---	0.535	0.219	0.172	0.099	0.229	13.196	23.898	1.992
8	0.158	1.608	1.322	1.683	4.240	2.487	0.745	1.256	0.525	0.293	0.077	0.138	12.130	26.662	2.051
9	2.697	4.533	4.770	2.538	3.968	2.666	1.206	2.098	0.826	0.205	0.033	0.175	8.840	34.555	2.658
10	3.163	2.684	3.397	7.188	5.993	5.355	4.467	3.573	1.282	0.396	0.058	0.303	14.181	52.040	4.003
11	0.915	0.641	1.514	1.668	3.737	2.157	7.169	8.533	0.894	1.043	0.242	0.361	70.173	99.047	7.619

12	1.420	0.689	1.248	1.032	1.359	1.826	5.380	12.263	2.475	4.179	5.451	1.570	324.452	363.884	27.991
13	0.767	0.632	1.016	0.724	1.321	0.820	1.040	2.557	2.867	8.834	8.173	5.451	515.817	550.019	42.309
14	0.806	0.623	0.744	0.642	0.892	0.682	0.946	2.132	0.296	1.287	4.434	11.529	696.258	721.270	55.482
15	0.328	0.529	0.570	0.680	0.169	0.535	0.745	1.644	0.249	0.652	1.576	7.578	237.183	242.411	19.416
16	0.357	0.615	0.483	0.390	0.109	0.585	0.821	1.284	0.266	0.824	1.627	2.886	135.320	145.567	11.197
17	0.696	0.836	0.522	0.487	0.089	0.563	1.182	1.419	0.235	0.865	1.992	2.125	119.498	130.509	10.039

Total

21.685	43.440	39.347	26.478	32.906	21.910	23.701	41.734	10.134	24.106	25.319	38.999	2733.2	3082.9
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Average

1.276	2.555	2.315	1.558	1.936	1.461	2.370	2.455	0.921	1.418	1.502	2.294	160.78	3.082
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TABLE 12

Chemical Oxygen Demand

(mg/l)

Sampling Positions

Run#	1	2	3	4	5	6	7	8	9	10	11	12	13	Average
5	722	612	267	486	141	408	—	596	—	1247	510	—	1470	646
6	533	209	116	170	170	193	—	301	—	402	232	696	696	338
7	345	172	196	242	157	274	—	235	666	274	251	290	329	286
8	323	227	60	165	110	55	235	86	212	125	94	125	149	151
9	—	—	48	84	120	60	232	96	188	188	—	140	148	130
10	177	151	52	86	69	65	155	129	129	151	216	99	99	121
11	102	90	43	66	82	128	117	148	184	121	74	97	94	103
12	102	94	143	86	60	136	71	105	—	75	68	90	60	99
13	36	44	52	52	29	67	67	52	156	265	273	94	155	103
14	38	57	27	38	—	34	46	—	—	—	—	—	—	40
15	91	125	18	19	29	43	62	38	—	62	34	29	50	53
16	65	55	92	—	—	18	5	23	18	23	14	90	50	34
17	23	27	47	47	—	47	39	23	97	39	51	70	46	48

APPENDIX II

TABLE 13

SOIL ANALYSIS DATA
Experiment 2

Sample depth in soil column (inches)	Total N %	$\text{NH}_4^+ - \text{N}$ ppm	$\text{NO}_3 - \text{N}$ ppm
Manure sample before testing	1.66	3351.9	120.8
Composite soil sample before testing	0.0298	16.1	8.5
Samples after 3 test runs			
0	0.0399	5.6	1.4
4	0.682	195.5	15.5
13	0.045	30.1	3.1
28	0.031	23.4	1.1
45	0.027	4.5	1.7

TABLE 14
Nitrogen Balance for Experiment 2

Initial Nitrogen

Soil	19.83	grams
Manure	6.86	
Sum	<u>26.69</u>	

Final Nitrogen

Soil	23.15
Manure	2.80
Sum	<u>25.95</u>

Nitrogen lost from system = 0.74 grams

Nitrogen lost due to leaching = 0.40 grams

Nitrogen lost in gas = ? (0.34 grams)

Percent lost to leaching = 54 percent

INVESTIGATION OF SOME FACTORS INFLUENCING
DENITRIFICATION IN A LABORATORY SOIL COLUMN
WITH A SURFACE LOADING OF LIVESTOCK WASTES

by

CHARLES VERNON BOORAM, JR.

B.S., Northeast Missouri State, 1966
B.S., Kansas State University, 1968

AN ABSTRACT OF A MASTER'S THESIS

submitted in partial fulfillment of the

requirements for the degree

MASTER OF SCIENCE

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KANSAS STATE UNIVERSITY
Manhattan, Kansas

1971

ABSTRACT

A study to determine the factors affecting denitrification in a laboratory soil column was conducted. Loamy sand was selected as the soil for the study. A layer of air dry manure was placed in the soil four inches below the soil surface, at the rate of 50 tons per acre.

Previous research at Kansas State University indicated that it was possible for denitrification to occur when periods of anaerobiosis occurred. With this in mind, the first experiment was conducted at aerobic conditions for 17 weeks. A second experiment was conducted at anaerobic conditions for 3 weeks. Water was added at the rate of 3.75 inches per week for both experiments.

During Run 6 of Experiment 1 the nitrate concentration in the water samples started increasing. This continued until Run 12. The nitrate concentration steadily decreased during the remainder of Experiment 1. The ammonia concentration moved down the column ahead of the nitrate nitrogen during the first 10 runs of Experiment 1. The ammonia and nitrate nitrogen moved at about the same rate during the last 7 runs.

The soil gained in nitrogen concentration during Experiment 1 and the manure lost about 75 percent of the organic and ammonia nitrogen. A majority of the nitrogen lost from the system was due to leaching. Denitrification or volatilization accounted for about 20 percent of the nitrogen lost from the system.

An air displacement column was used to measure the intermittent gas flow. This provided a method to measure the input and output gases

from the soil column, but provided little help in the gas analysis, because the Fischer/Hamilton Model 29 Gas Partitioner provided poor quantitative results. Analysis of the data provided an excess of 9 grams of nitrogen for the nitrogen balance.

The second part of the experiment consisted of purging the soil column of atmospheric air and replacing it with 80 percent helium and 20 percent oxygen. It was hypothesized that the chromatograph would be able to detect the change in nitrogen concentration when the input gas contained 80 percent helium and 20 percent oxygen rather than 78 percent nitrogen and 21 percent oxygen. During Run 3 of Experiment 2, a leak developed in the system. The nitrogen concentration jumped to about 80 percent which would indicate that atmospheric air was entering the system. There was a higher rate of nitrogen loss from the soil column in Experiment 2.

Two critical points have been considered. The first being when continuous aerobic conditions exist and the second when the column is maintained at continuous anaerobic conditions. It appears that more useful information could be determined (a total nitrogen balance) if a better type of gas analyzing equipment were used. The best method of gas analysis would be the mass spectrometer.

This system does provide a method to determine the rate of nitrate movement through a soil profile. The rates of application of water to the soil system in these two experiments have been high. The variation of water application rates would give some insight into the rate of nitrate movement into the ground water when correlated with field studies. This data could then be interpolated to other types of soil to give some meaningful information on nitrate movement in soil systems.