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AGRICULTURAL BENEFITS AND ENVIRONMENTAL CHANGES RESULTING FROM THE USE OF DIGESTED SLUDGE ON FIELD CROPS

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The effects of digested sludge application on the chemical composition of soil, plant, and water samples from a large field lysimeter facility are discussed. Specific hygienic aspects of digested sludge were also investigated and, it was found that viruses are not likely to survive the heated anaerobic digester environment and, although digested sludge contains large populations of fecal coliform bacteria, these organisms die away rather rapidly during storage and after spreading on the soil. Results from green house and field studies indicate that several crop plants show favorable growth responses when fertilized with digested sludge, however, concentration levels of several chemical elements in soils are increased above native amounts and are also increased in plant tissues. As long as digested sludge application rates do not exceed those which will result in unacceptable concentration levels of NO_3-N in drainage or groundwaters, sludge of the quality employed in this study can be safely used to increase the production of good quality crops.

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ABSTRACT

The construction, equipage and operation of a large field lysimeter facility are described in detail. The effects of digested-sludge application on the chemical composition of soil, plant, and water samples from this lysimeter facility and another lysimeter facility established in 1939 are discussed. Specific hygienic aspects of digested sludge were also investigated and, it was found that viruses are not likely to survive the heated anaerobic digester environment and, although digested sludge contains large populations of fecal coliform bacteria, these organisms die away rather rapidly during storage and after spreading on the soil as a surficial application of sludge. Results from greenhouse and field studies indicate that several crop plants show favorable growth responses when fertilized with digested sludge. With increasingly greater sludge applications, concentration levels of several chemical elements in soils are correspondingly increased above native amounts and their levels are also increased in plant tissues. A phytotoxic condition with soybeans traceable to high salt concentrations was encountered in a greenhouse study; the condition was ameliorated by leaching. The first limiting factor in determining digested sludge application rates on crop lands is its N content. As long as digested sludge application rates do not exceed those which will result in unacceptable concentration levels of NO_3-N in drainage or ground waters, sludge of the quality employed in this study can be safely used to increase the production of good quality crops. If digested sludge is continuously applied on land at rates which supply N in amounts which greatly exceed the plants' capacity to utilize it, soluble P and some of the more soluble heavy metal constituents of sludge may eventually adversely affect the growth of crops or result in the accumulation of some chemical element in crop tissues at concentration levels that might pose a threat to animal or human health. Studies are in progress to determine the fate of several selected chemical elements added to cropped soils as constituents of the digested sludge which has been applied annually at various rates since 1968. The results from these several studies show that P, Cu, Ni, Zn, and Cd should be given special consideration with regard to monitoring their accumulation in soils, absorption by crops, and transport in percolating water when digested sludge is used as a soil amendment.

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SECTION I

CONCLUSIONS

The conclusions presented here are stated in general terms. Practically all of these have been previously presented in publications such as monthly and annual progress reports, scientific journal articles, conference proceedings, graduate student theses, etc. No attempt was made to compile conclusive statements from the reports for each individual study. Rather, it is left to the reader to inspect the summary presented for each study reported herein and to accept or reject the conclusive statements arrived at by an individual principal investigator.

1. A lysimeter facility consisting of 44 prisms of soil having provisions for collection of runoff and drainage water was constructed. In addition to the automatic collection capability, design permits measuring the hydrograph and identifying individual water samples with the hydrograph and with real time. Thus, water quality of discrete samples are associated with the characteristics of the hydrograph.

2. Frequent digested sludge applications by spray irrigation caused a reduction in corn and soybean yields as compared to fertilization with inorganic fertilizers and well water. The crops did not appear to be affected by sludge spray irrigations in any other way than a reduction in leaf size. The reduction in the size of leaves, and thus leaf area, was probably caused by frequent coating with sludge and was associated with a reduction in photosynthetic rate as the absorption of light by the leaf was decreased by solid particles of sludge adhering to their surfaces. It is doubtful that two or three applications of digested sludge by spray irrigation during a growing season would cause a significant decrease in crop yields. In the research described here sludge was spray irrigated as often as possible, which sometimes was as often as two or three times per week. Where digested sludge has been applied by ridge and furrow irrigation, crop yields have been comparable or greater than those obtained with inorganic fertilizers applied at rates estimated to be adequate for maximum yields.

3. Nitrogen contained in digested sludge is the first limiting factor

on rates of application for field crops. Our data indicate that about five to eight centimeters of sludge applied on the surface immediately after withdrawal from the digester would satisfy the annual nitrogen needs of non-leguminous crops without producing excessive nitrate nitrogen in percolated water. In the interest of higher loading rates to obtain the ameliorating effects of organic matter in the reclamation of surface-mined land, reduction of the nitrogen content of sludge would be desirable. However, where maximum utilization of nitrogen is the objective, digested sludge should be injected or immediately incorporated into the soil to reduce the losses of nitrogen by ammonia evaporation.

4. Digested sludge is an effective source of phosphorus for fertilization of crops. The phosphorus contained in digested sludge is readily available for crop plant uptake, as evident by the immediate and consistent increases in available phosphorus (as measured by Bray P_1 test) in soils after sludge has been applied and incorporated.

5. Relative to its nitrogen and phosphorus supplying capacity, digested sludge is not a good source of potassium for growing crops. Thus where digested sludge is utilized as a nitrogen and phosphorus fertilizer on soils having a demonstrated need for supplemental potassium fertilizer, the practice of supplying potassium from inorganic fertilizer sources should be continued.

6. Heavy metals are ubiquitous constituents of digested sludge and they occur to the greatest extent in the solid phase. After application to soil, the heavy metals for the most part remain in the layer of incorporation or plow layer. Thus, many agronomists are of the opinion that long time, continuous use or disposal of sludge on cropland will eventually lead to an accumulation of the more soluble or plant available trace elements in soils to a level where toxicity may occur to either the plants or to consumers in the subsequent food chain. In much the same way that macronutrients are increased in plant tissues following sludge applications, the absorption of all trace elements by plants are enhanced to some degree. The trace elements which have been increased in plant tissues by sludge application to the greatest extent are Zn, Cd, and Fe. However, phytotoxicity traceable to trace element or heavy metal toxic conditions has not been observed and neither have concentration levels of trace element in plant tissues reached proportions that constitute a health hazard to animals consuming all or any part of the plants fertilized with sludge. Nevertheless, since Cd is one of the elements which shows the greatest relative increase in plant tissues with sludge applications and is one which is considered to be a source of adverse physiological and pathological effects in animals, its circulation in food chains will continue to be closely scrutinized during the continuation of studies.

7. The rate of infiltration of digested-sludge liquid is low regardless

of whether the surface soil is of silt loam or sandy texture. Thus, on sloping land special precautions should be taken to control the distribution of sludge applied to the soil surface. After drying, digested sludge does not affect the infiltration of water into the soil surface. Shallow ponding of sludge in the furrow for even a few days does no apparent harm to plants. Where adequate drainage exists or is induced, salt accumulation in humid region soils is not expected to be a problem but may limit application rates in arid and semiarid regions.

8. Sludge organic residue decreases the bulk density of the soil. Grease contained in sludge has not proven to be a problem and is rapdily decomposed after application on or in soils. Organic carbon has accumulated in amended soils, but has presented no observable problem. On the contrary, applications of digested sludge has apparently improved the tilth in soils having naturally low organic matter contents.

9. Seed germination is inhibited if freshly digested sludge is incorporated in soil and the seed planted immediately. But if seeding is delayed for two to three days after an application of digested sludge, no inhibition is observed.

10. Properly digested sludge will produce no offensive odors or fly breeding problems after application to soil.

11. Applications of more than five to eight centimeters of freshly digested sludge per year to soil markedly increases the nitrate nitrogen content of leachate waters.

12. Nitrate nitrogen losses in leachate waters are positively correlated with sludge rates and discharge intensity of the leachate.

13. Ammonium nitrogen and phosphorus losses through leaching are not statistically related to sludge application rates or water discharge intensity.

14. Ammonium nitrogen losses in runoff water are negatively correlated with runoff intensity and sludge accumulation.

15. Phosphorus losses in runoff waters are positively correlated with sludge accumulations.

16. It is easy to advance arguments either to minimize or maximize the dangers of sludge irrigation of soils in respect to public health considerations. Known cases of digested sludge application over agricultural fields have been recorded for many years in several countries. Thousands of individuals in these waste treatment plants and sludge spreading fields have handled the material without succumbing to disease as a result of such operations. On the

other hand, the very fact that digested sludge harbors a large population of fecal coliforms renders it suspect as a potential vector of bacterial pathogens. Our studies have shown that the sludge fecal coliform population decreases markedly following application to the soil or upon aging after removal from the digester. Lagooning of digested sludge prior to application would serve the purpose of reducing the fecal coliform and, presumably, the pathogenic bacterial population. Pathogenic organisms will rapidly die away after application on the soil surface. They will move from the point of application for the most part only in an absorbed phase on eroded sediments. Therefore, the establishment of recommended erosion control, structures and practices on the land utilization site provides an additional public health protection factor.

17. Porcine enterovirus did not survive beyond the fourth day in a heated anaerobic digester. This suggests viral agents probably do not survive the 14-day anaerobic digestion cycle used in sewage sludge treatment. Further die away of viral agents would occur during the lagooning period and at the point of application on soil surfaces.

18. Storage of digested sludge in deep lagoons for the 2 to 3 month period, recommended as a safety factor to permit die away of pathogenic organisms, will not result in a significant change in the total nitrogen content of the material. Because over one-third of the total nitrogen contained in digested sludge is lost by ammonia evaporation following application on land surfaces, information regarding similar nitrogen losses during open storage of digested sludge was required for estimating nitrogen loading rates on land. It was determined that the surface area to sludge volume ratio is the most important factor controlling ammonia nitrogen volatilization. Thus, the removal of ammonium nitrogen as a constituent of the decanted effluent during lagooning operations is the major reason why lagooned sludges often contain lower total nitrogen contents as compared to sludges drawn directly from anaerobic digesters.

SECTION II

RECOMMENDATIONS

Based on the results of the several studies reported herein, recommendations are presented from the standpoint of utilizing the information for (1) implementing spreading operations of digested sludge on land (2) the need for further research relevant to environmental changes, including effects on plant composition, with long-term continuous use of digested sludge as a fertilizer or soil amendment.

Implementing Land Spreading Operations

The use of digested sludge as a source of nutrients for growing crops and as a soil amendment for reclaiming severely disturbed land is the most environmentally safe and economically sound solution to a growing solids waste handling problem. All other methods of sludge disposal pose a direct or potential air and/or water pollution hazard which may go undetected and when detected would be difficult to abate. When sludges from municipal wastewater treatment plants are burned, buried in land fills or dumped into oceans, control over the dispersion of chemical constituents into the environment is relinquished to a very marked degree. On the other hand, when digested sludge is used as a fertilizer and soil amendment on properly selected and prepared sites, environmental changes are observable in the nature of response of the vegetation, runoff and drainage water quality determinations, and the results from soil sample analyses. The cost and effort required for monitoring a digested sludge utilization, or land spreading operation to insure that adverse environmented impacts do not go undetected is rather small in comparison with the total benefits derived. If the environment is adversely altered, various practical soil, water and crop management practices can be applied to promptly remedy or abate the situation. Control over potential pollutants is retained where emphasis is on sludge utilization rather than its disposal.

Digested sludge can be utilized as a supplemental N fertilizer. However, when digested sludge is utilized to furnish all of the supplemental N needed to provide optimum fertility for nonleguminous crop plants, a large part of the P that would be concomitantly applied is wasted.

Inorganic P fertilizer materials are already in short supply. Regardless of whether or not the dire predictions for a chronic shortage of P fertilizer, as based on known reserves of high grade ore, proves to be correct, certainly P fertilizers will become increasingly more expensive as regulations to reduce environmental degradation effects associated with mining are enforced. Therefore, it will be in the interest of society as a whole if digested sludge is utilized at rates just sufficient to satisfy P fertilizer needs. If digested sludge is applied at rates just adequate to satisfy either supplemental N or P requirements of crop plants, the findings presented in this report do not provide grounds for objecting to its use.

On the contrary, when digested sludge is spread on lands at rates which supply N in quantities that greatly exceed the capacity of the vegetation to utilize it, precautionary measures are required to protect water supplies against the introduction of objectionable concentration levels of NO_3-N. In some land reclamation schemes it may be desirable to utilize sludge for its unusual buffering capacity and high organic matter content to ameliorate existing chemical and/or physical characteristics of soil or geological material that inhibit vegetative growth. Where digested sludge is used as a soil amendment to obtain these rapid ameliorating changes, annual loading rates will generally be considerably greater than those required just to satisfy requirements for N and P fertility. It is at the high loading rates required to improve the productivity of lands, as contrasted to fertility enhancement or maintenance rates, that many questions arise regarding the advisability of utilizing digested sludge.

Many questions center on the quality of waters from land areas amended with sludge. With regard to runoff water it is neither feasible nor desirable to hold all the water on the application area. It is however an essential requirement that soil erosion control structures and practices be established to insure that sediment yields from the area are maintained within tolerable limits. In this way acceptable runoff water quality is assured simply by keeping soil losses due to erosion within acceptable limits. With regard to drainage water it has already been mentioned that the foremost concern with high sludge application rates is the increase in concentration levels of NO_3-N to be expected. Where large annual applications of digested sludge are applied in land reclamation projects, stratagems should be instituted to limit NO_3-N content in drainage water. Stratagems may range from reducing the N content of the sludge to recovering drainage water for recycling or removal of the N by denitrification processes. The simplest and most economical method of reducing the amount of NO_3-N in drainage water is by adopting surface application methods, thereby allowing sludge to dry before it is incorporated into soil or geological material. Thus N losses are maximized by way of NH_3 evaporation. Systems for injecting or incorporating wet sludges in soil materials should be avoided or some fail-safe N management scheme instituted where the main objective is to apply large amounts of sludge to rejuvenate soil or to reclaim severely disturbed lands.

Some of the most vocal resistance to land spreading of digested sludges arises with regard to its trace element content, because of the potential threat to human health that may be posed by its utilization. From the results of the several studies reported here, it appears that if digested sludge is used at rates just sufficient to provide supplemental N and P fertility the likelihood of increasing concentrations of one or more trace elements to levels in foodstuffs that could cause untoward effects in man or livestock is a fairly remote possibility. When digested sludge is used as a soil amendment and applied at rates which greatly exceed that needed to optimize soil fertility, the possibility does exist that trace elements may be absorbed and transported into plant tissues in amounts which may be harmful to animals. Thus, where large annual applications of digested sludges are applied on land, concentration levels of Zn, Cu, Cd, and Ni in plant tissues should be monitored. These, especially Cu, Cd and Ni, are the trace elements which are harmful in animal feed stuffs at low concentration levels and/or accumulated at relatively high concentrations in the tissues of plants fertilized with sludge. The concentration levels of one or more of these four elements in plant tissues will signal the need for remedial action. However, critical concentration levels in plant tissue for any of the four metals has not yet been established and only scant attention has been given to interactions among these and other elements.

Research Needs

Further research should be directed toward assessing environmental changes resulting from digested sludge application rates comparable to those anticipated for use in land reclamation projects. Special emphasis should be placed on N management practices and toward determining the fate of trace elements where digested sludge is applied at rates required to rapidly amend soils and geological materials for increased crop production. The use of digested sludge to correct naturally occurring phytotoxic conditions in severely disturbed lands and to reduce the leaching of water polluting substances from mined and industrial waste disposal areas should receive further study.

The coarse of changes in the organic fraction of sludge deserves study as does the nature of secular change in availability of the macroelements and microelements of physiological importance.

SECTION III

INTRODUCTION

Factors Contributing to Sludge Handling Problems

Sludge quantities increasing - The disposal of wastewater treatment plant residues is the most difficult and increasingly costly problem confronting major sanitary district staffs. For cities of over 50,000 population the average per capita suspended solids load at wastewater treatment plants was reported by Loehr (92) to be 0.114 kg per day on a dry-weight basis. Plants receiving large quantities of industrial waste may have average per capita loadings approaching twice the average value. For example, the Metropolitan Sanitary District of Greater Chicago has an average per capita loading of about 0.182 kg per day. It is expected that the average per capita loadings will increase because the installation of garbage grinders in homes will augment suspended solids by an average of 60 percent, (Am. Soc. of Civil Eng. Manual, 1959). In these regards it is pertinent to note that at the time Loehr (92) collected his data, only about 12 percent of the homes were equipped with garbage grinders.

At present the activated sludge treatment process is most frequently used for secondary treatment of wastewater. From the standpoint of suspended solids removal, the process when preceded and followed by sedimentation, is about 85 to 90 percent efficient. Eventually, as wastewater treatment facilities are upgraded to include tertiary treatment processes, the efficiency for suspended solids removal should be at least 98 percent. Therefore, it is likely that in the near future somewhere between 95 to 98 percent of the per capita loading reaching the wastewater treatment plant will be retained as fresh sludges.

Along with the increase in quantities of wastewater given tertiary treatment for improved removal of solids, higher priorities are also likely to be given to reducing phytoplankton nutrients to lower concentrations in effluent. The removal of nutrients will require the addition of chemicals such as the dosing of effluent with lime or alum to precipitate soluble phosphates. Added chemicals which cannot be economically regenerated for recycling will add materially to the solids handling problems. Assuming an average wastewater flow of 511 liters per capita per day (92), a chemical dosage of only 50 ppm will increase the per capita per day suspended solids in fresh sludge by 0.023 kg.

Considering the trend toward greater usage of garbage grinders, tertiary treatment processes, and chemicals for reducing nutrient concentrations in effluent, an average value of 0.159 kg per capita per day of solids as fresh sludge would appear to be a conservative estimate of production, at least for the larger advanced wastewater treatment plants. For each million population served by sewers, about 153 metric tons of fresh solids will be removed from about 511 million liters of wastewater requiring treatment each day. Therefore, municipal sludge handling problems will increase, even if our sewered population should remain static.

Kinds of sludges generated - The solids separated from wastewater during sewage treatment are a complex array of organic and inorganic residues. Upon reaching the wastewater treatment plant, about 60 percent of the suspended-solids load is removed by sedimentation. The solids portion removed by this sedimentation is called primary sludge. The solids not removed by the primary treatment sedimentation process are transferred to another tank as a constituent of the effluent where they are mixed with large quantities of aerobic microorganisms and large volumes of air. The microorganisms use the O in the air to convert part of the organic waste into carbon dioxide and water to obtain energy, while converting another large portion of the waste into new cells. Waste converted into new microbial cells and collected by sedimentation after removal from the aeration tank is called activated sludge. To maintain a microbe population in the growth phase a portion of the activated sludge is recycled to the aeration tank, but for the most part it is wasted and thus often referred to as waste-activated sludge. The primary sludge and the waste-activated sludge generated during secondary treatment when taken together, make up the fresh sludge discussed above.

In the United States many attempts to spread primary or raw sewage sludge on land have ended in failure. Waste-activated sludge has been successfully used as a fertilizer material only after heat drying and then at only light applications which could be thoroughly incorporated with soil. Such biologically unstable materials as primary and waste-activated sludge cannot be spread on land or lagooned because of odor and fly problems. In the older literature, waste-activated sludge is sometimes referred to as aerobically digested sludge. Waste-activated sludge is highly unstable with regard to further biological degradation and should not be referred to as a digested sludge. To stabilize waste-activated sludge sufficiently for land surface application by an aerobic process would require a detention time of about 20 days (72). Studies at the University of Wisconsin have demonstrated the adaptability of an aerobic digestion process to the stabilization of mixtures of raw and waste activated sludge (114). Aerobic digestion of primary sludge has been evaluated by Viraraghanan (161) for average climatic conditions in the vicinity of Madras, India.

In the older literature, discussions regarding sludges from Imhoff tanks are often confused with those concerned with sludges from heated anaerobic digesters; both simply referred to as anaerobic digested sludge by some authors. While some degree of anaerobic sludge stabilization is accomplished in Imhoff tanks, it may or may not be comparable to that accomplished in a heated anaerobic digester where environmental conditions are maintained near optimum for rapid biclogical degradation of organic sludge constituents. Lohmeyer (93) reviewed the literature pertaining to heated anaerobic digestion and presented recommendations for managing digesters to obtain the best overall results with the least difficulty. In a later literature review, Pohland (120) discussed anaerobic decomposition in terms of two phases. The first he designated as liquefaction and hydrolysis, the second, fermentation and gasification. A rather heterogeneous group of bacteria convert the proteins, carbohydrates, and lipids contained in the waste largely to fatty acids, carbon dioxide and ammonia nitrogen during the first stage. During the second stage strict obligate anaerobic bacteria convert the fatty acids produced during the first stage to methane and carbon dioxide. Toerien and Hattingh (153) reviewed the literature toward presenting the current state of knowledge about the microbiology and biochemistry of the anaerobic digestion process and to identify areas needing further research. They state that it seems probable that fungi and protozoa do not play significant roles in the degradation of organic matter during anaerobic digestion. Andrews (6) presented a dynamic model for the anaerobic digestion process, which has usefulness in predicting the results of changes made in the operation of digesters.

The above reports regarding aerobic and anaerobic digestion processes for raw (primary) and waste-activated sludges are sufficient to emphasize the attention that has been given to organic waste stabilization. Some of the reasons given for stabilization of sludges are that it promotes rapid dewatering, reduces the initial bulk of solids for more economical handling, destroys pathogenic organisms for health protection, and noxious odors are eliminated. Another most important reason for stabilization is the elimination of housefly infestations of stored waste. Apparently the housefly will readily breed in raw, waste activated or partially digested sludge, but not in a well digested sludge (57)(162)(170).

To overcome some of the objectionable characteristics of primary and waste activated sludge, the use of heated anaerobic digesters has proven to be both satisfactory and economical (98). Heated anaerobic digestion of sewage solids is used to accomplish two primary objectives. First, about 50 to 70 percent of the organic fraction of sludge solids is biologically converted to methane and carbon dioxide, reducing the amount of total solids that must be handled by about 40 percent. After digestion, the organic fraction of the remaining solids is sufficiently stabilized against further biological degradation so the material can be lagooned, dewatered on open drying beds, or applied on the surface of soils without causing noxious odors or providing a substrate for fly breeding. By anaerobic digestion the projected sludge handling problem may be reduced from 153 to 96 metric tons per day per million population.

Cost of sludge disposal - Cost for the incineration of sludge (includes wet-air oxidation, multiple-hearth, and fluidized-bed) ranges from 30 to 42 dollars per dry ton as reported by Burd (26) and from 50 to 57 dollars as reported by Bacon and Dalton (8). Because these estimates were made from data collected several years ago they are probably conservative. If the greater cost for minimizing air pollution and increased cost resulting from inflation are considered, the cost for incineration of sludge solids today is probably greater than 60 dollars per dry ton. Furthermore, incineration does not provide for a permanent solution to the solids handling problem. The ash accumulating from the oxidation of fresh sludges amounts to 30 to 35 percent of the original dry weight and presents some of the same disposal problems as those encountered with the original material.

Waste-activated sludge has sometimes been heat dried and sold as a low-grade organic fertilizer. Dry, waste-activated sludge contains about 4 to 6 percent N, 3 to 7 percent P_2O_5 equivalent and 0.25 to 0.6 percent K_2O equivalent. Thus, from the standpoint of a fertilizer, the inconvenience and cost of supplying sufficient quantities of dried sludge to satisfy the nutrient requirement of most crops is too great to expect an increase in its marketability. Even before it was necessary to consider the installation of equipment to reduce air pollution Bacon and Dalton (8) reported that the net cost for disposing of 228 to 273 metric tons of sludge as a fertilizer material was 45 dollars per dry ton.

Burd (26) reported a cost of 50 dollars per dry ton for drying and applying sludge on land and 25 dollars per dry ton for the application of dewatered sludge on land. He also concluded that the cost for disposal of dewatered sludge in land fills was about 25 dollars per dry ton. Cost estimates for permanent lagooning of digested sludge range from 12 (26) to 49 (8) dollars per dry ton. A number of variables determine the actual cost of land disposal schemes, but the major variables are the initial cost of land and distances sludge must be transported from the wastewater treatment facility to the disposal site. Whether sludges are applied on or near the soil surface, dumped in landfills or held in lagoons, all are aesthetically unacceptable because, if for no other reason, the land is condemned to a singularly low degree of usage.

In the last few years a great deal of attention has been given to the old idea of utilizing digested sludges as a source of nutrients to grow crops and as a soil amendment to ameliorate physical conditions in severely disturbed lands that are inimical to the establishment and growth of plants. It is not envisioned that disposal by utilization can be carried out without cost to the sanitary district. On the other hand, contrary to strict land disposal schemes, it is envisioned that the solids will be utilized in such a manner that land usage is either not changed or in the case of land reclamation the number of alternative land uses is increased. In 1968, members of Harza Engineering Company estimated the cost for pumping digested sludge containing 3 to 5 percent solids a distance of about 80 km and distribution it on land in amounts just sufficient to supply the nitrogen needs of nonleguminous plants. On the bases of a 6 percent interest rate and amortization of all construction costs over 50 years, and including maintenance and operation of the sludge distribution equipment, they estimated the cost for sludge disposal by agricultural utilization to be 22.30 dollars per dry ton. Wirts (169) estimated the cost for pumping digested sludge to be 15 to 22 cents per metric ton km. He pointed out that cost depends on tonnage pumped and suggested that a connected population of 2 million people is an economical starting point for considering pumping distances of 80 to 160 km. At the present time, sludge is being transported from the Metropolitan Sanitary District of Chicago wastewater treatment plants to an agricultural utilization site 160 miles downstate by a unit train. The unit train contains 30 tank cars, each having a 76,000 liters capacity. By another contract, sludge is being barged 290 km from Chicago to a land reclamation site. While transportation costs vary with the solids content of the digested sludge they have generally ranged from 30 to 35 dollars per dry ton during these short-period (3-year) rail- and barge-haul contracts. With a continuous or sustained operation, transportation cost by rail and barge could be considerably reduced. On a sustained operational basis it does not appear unreasonable to consider transportation distances of 320 km from large municipal waste treatment facilities when contrasted to cost for alternative methods of sludge disposal.

Land requirements - If all municipal waste waters generated in the continental United States were given secondary treatment and the resulting solids stabilized for utilization as a fertilizer and soil amendment, about 9.1 to 10.9 metric tons of solids would be available each year. The utilization of the solids in amounts just sufficient to meet the needs of nonleguminous crops for supplemental N would require an annual application of about 22.5 to 33.7 t/ha. Thus, not more than 0.4 million hectares of land would be required at any one time to utilize the total continental United States production of sludge solids. Only enough sludge solids would be available to treat slightly more than 0.2 percent of the 188 million hectares of cropland or slightly less than 0.06 percent of the total 771 million hectares contained in the continental United States. However, because of its potential as a source of sorely needed stable organic matter, municipal sludge exhibits its greatest value as a resource when used as an amendment for the reclamation of surface-mined lands. Since over 0.2 million hectares of land strip-mined for coal prior to 1964 already exist in various states of devastation, while another 0.2 million hectares have been or will be stripped during the 20-year period from 1964 to 1984, there is no scarcity of land which needs the nutrients and organic matter supplied in sludge. About 30 percent of the country's population is within economical sludge pumping distances to land that has been strip-mined for coal in Illinois, Indiana, Kentucky, Ohio, West Virginia, and Pennsylvania.

Those who express concern about the contamination of soils with constituents of municipal sludges probably are not aware of the relatively small amount of land needed. Confusion often exists between land requirements for sewage effluent disposal or renovation and that needed for solids utilization.

Criteria for selection of sludge utilization sites - In utilizing digested sludge as a soil amendment and fertilizer, the following criteria for site selection are recommended. (a) The site should be located where utilization of the sludge offers maximum benefits to the local agricultural economy, consistent with reasonable costs to the particular sanitary district. The local populace must be able to weigh the benefits to be realized from the sludge utilization program against the assumed or real stigma attached to an area that becomes the receptor of waste from a large municipality. People living in areas devastated by surface mining activities readily recognize the benefits to be realized by utilization of digested sludge to reclaim land. (b) To ensure that sludge applications are made under uniformly controlled conditions, the land must be susceptible to purchase or long-term lease by the sanitary district. (c) To minimize sludge distribution cost, all lands in the site should be contiguous, at least to the extent that the disturbance to existing residents is minimal. Surface-mined lands offer the best possibilities for obtaining large, contiguous acreage. There is little or no disturbance of existing residents, because this occurred during the stripping process. It is envisioned that much of the land will be repopulated with farm operators as the land is reclaimed to a high state of productivity. (d) Soil depths should not be less than 1.83 meters to permeable bedrock. Water tables should be capable of being maintained to average depths of at least 1.83 meters from the soil surface. Such minimum soil depths, with good management practices, will provide protection from ground water pollution. (e) Land slopes should not be so steep as to prohibit the establishment of water management and erosion-control structures at a reasonable cost. Slopes up to 18 percent may be acceptable where "push-up" terraces with permanently vegetated or sodded back slopes can be established. Unconsolidated geological materials must be sufficiently deep to bedrock in the borrow area so that after terrace construction a minimum 1.83-meter depth to bedrock is maintained.

Environmental Benefits and Public Health Protection

Chemical and physical - In Table 1 some average concentration values are presented for several chemical elements found in digested sludge from the Calumet and Stickney wastewater treatment plants of the Metropolitan Sanitary District of Greater Chicago. Sludges from both of these treatment plants have been used in the research conducted (since 1967) by members of the Agronomy Department, University of Illinois.

Table 1. Composition of anaerobically digested sewage sludges from MSD of Chicago, Calumet and Stickney treatment plants. Samples obtained during 1971 (Calumet late in year).

	Means (Wet Weight)	
Element	Calumet	Stickney
Cd ppm	3.0	14.0
Mn "	8.0	18.0
Ni "	3.0	15.0
Zn "	83.0	223.0
Cu "	16.0	67.0
Cr "	26.0	194.0
Fe "	726.0	2100.0
Pb "	16.0	75.0
Hg "	0.063	0.275
Na "	98.0	131.0
P "	757.0	1141.0
Ca "	963.0	1289.0
Mg "	180.0	484.0
K "	195.0	390.0
N %	0.09	0.156
% Solid	2.05	4.36
% Volatile	58.0	48.0

Anaerobically digested sludge, as it comes from digesters, contains 3 to 5 percent solids as finely divided and dispersed particles. It looks like crude oil and has an odor which many people describe as earthy or tarry. It can be easily transferred by pipes using ordinary pumping techniques and equipment. When applied to cropland at the rate of 5 cm/ha, it will supply all of the major essential nutrients, including: 224 to 392 kg/ha of NH_4-N; about the same amount of organic N, some of which will be slowly released in a form available to crops; 280 to 504 kg of P, of which about 80 percent is in organic matter; and 45 to 90 kg of K. Sulfur will also be supplied in amounts adequate for crops. The amounts of Ca and Mg supplied will exceed the average annual losses of these elements by leaching in humid regions.

High application rates of digested sludge on cropland can cause obvious NO_3 problems. To determine maximum sludge-loading rates on soils, total and soluble nitrogen contents must be known. The soluble N in anaerobically digested sludge is in the NH_4-N form, but under proper soil aerobic and temperature conditions it is rapidly converted to mobile NO_3-N.

Thus, the loading rate of sludge on cropland is limited by the amount of soluble N plus an annual mineralization of the organic N supplied by sludge applications. If loading rates are based on the amount of N furnished to meet crop needs and losses by volatilization, soluble P applications will also be at low enough levels that P will not present a eutrophication threat to water supplies. When sludge-loading rates are based on safe N application rates, the capacity of most soils other than sands to inactivate P by adsorption and conversion to sparingly soluble precipitates or compounds is great enough to maintain P levels in drainage water to less than one ppm.

When the main objective is land reclamation, sludge-loading rates may be considerably greater because disturbed lands generally have small or nonexistent organic N reservoirs. The amelioratory effect of organic matter on the physical properties of soil materials may make it desirable to increase sludge-loading rates on marginal or severely disturbed lands above those recommended for productive agricultural lands. However, as the highly stabilized sludge organic matter accumulates in soils with succeeding applications, the slow mineralization of organic N must be taken into account to prevent losses of NO_3-N to water supplies, within or adjacent to the treated areas.

Many toxic and nontoxic organic waste materials occurring as constituents of sludge arise as discharges from industrial processes, such as the chemical production of textiles, plastics, pharmaceuticals, detergents, and pesticides. After a period of microbial acclimation, some organic toxic substances, such as phenols and formaldehyde, can be almost completely removed from wastewater by biological treatment, even though at sufficiently high concentrations they are bactericidal (74). Others, which are nonbiodegradable under aerobic conditions, may be removed from effluent with or by absorption on sludge sediments and later biologically degraded during anaerobic digestion of the solids. Of all the organic materials, polychlorinated biphenyls (PCB's) have been of greatest concern to those involved with municipal waste utilization. Many sludges contain 1 to 4 ppm or more and, like other chlorinated hydrocarbons, PCB's are only very slowly degraded by microorganisms. Where we have applied 105 metric tons of digested sludge a small increased concentration of PCB's was found in the soil, but they were not taken-up in detectable concentrations in soybean and corn plant tissues. Since bacteria are the first group of soil microorganisms to be decreased by abnormally high concentrations of chlorinated hydrocarbons, we have made total counts from soil samples collected from plots which have been treated with up to 124 metric tons of sludge over a period of 4 years. Total bacteria populations were found to be higher in soils treated with sludge. The positive correlation between total bacteria and amounts of applied sludge was highly significant. It appears that sludge applications have modified the soil environment in a manner that favors the maintenance of a highly active population of bacteria resulting in a greater rate of pesticide degradation than might be expected in soils not treated with sludge.

Heated anaerobically digested sludge is outstanding in its ability to increase the humus content of soils. For example, in 1941 a study was initiated at the Rothamsted Experiment Station in England to compare the effects of four types of organic manures with inorganic N fertilizers on market-garden crops (100). The organic manures were farmyard manure, digested sewage sludge, a compost of straw and sewage sludge. Each of the organic manures was applied at the rate of 33.8 to 67.5 t/ha per year. After nine years, N in the top 22.9 cm of soil was 0.088 percent where inorganic N, the familiar fertilizer source, had been applied as compared to a value of 0.089 for control plots. At application rates of 33.8 to 67.5 t/ha per year of digested sludge, the N content in the soil surface was 0.176 percent and 0.247 percent, respectively. These data indicate that the amounts of N in the sewage-sludge plots increased about three times as much as in the corresponding plots treated with farmyard manure and compost made from straw and farmyard manure. The surface soil in sewage-sludge treated plots contained about 50 percent more N than plots treated with equivalent amounts of compost made from straw and sewage sludge. Following the first nine years, treatments between 1951 and 1960 with digested sewage sludge produced only a slight increase in soil N percentages. However, N contents remained at a considerably higher level in sewage-sludge-treated plots than were obtained with either farmyard manure or compost.

In 1960, Jansson (75) investigated some specific properties of the humus fraction of fresh cow dung, well-rotted farmyard manure, and digested sewage sludge. He found that the size of the lignin-like complex in farmyard manure and digested sludge was somewhere between that from fresh plant residues and that developed from soil humus, but fresh cow dung was similar to fresh plant residues. Jansson stated that "the oxidation rate of the farmyard manure and the sludge is similar to that of the humus of an acid podzol" (acid forest soil).

More recently we have found that 306 t/ha of anaerobically digested sludge incrementally applied during four years on Blount silt loam soil increased its organic carbon content from 1.2 to 2.4 percent in the surface 15.2 cm.

Lunt (96) reported that digested sludge had a very favorable effect on several soil properties. He reported a moderate increase of 3 to 23 percent in moisture holding capacity, non-capillary porosity, and cation exchange capacity following the incorporation of digested sludge into soils. Furthermore, he found an increase in soil aggregation ranging from 25 to 600 percent which could be attributed to digested-sludge additions.

The results of the studies described above indicate that a sizable proportion of the organic material produced in a 15-day anaerobic digestion process has properties very close to that of natural soil organic matter or humus. Digested sludge is one of the few materials that can be used to effect a rapid increase in the humus content of soil. It is the only substance with these properties that is available in quantity.

To reestablish soil organic matter contents in severely disturbed or eroded lands to levels equivalent to those characteristic of productive soils will take many years under normal cultural practices. For example, in nature the time necessary to build up soil organic matter profiles to a point of equilibrium with their environment has been estimated to be not less than 200 nor more than 1,000 years from studies conducted on soil profiles in Columbia and California (78). Considering the importance of soil organic matter as a storehouse of slowly available plant nutrients, a source of cation exchange capacity, and a promoter of stable soil structure, two centuries is too long to wait for natural processes to build up the soil organic matter levels in unproductive lands while we seek ways to dispose of a material which can be used to effect a beneficial, immediate change.

Some waste treatment plant sludges contain higher concentrations of Cr, Zn, Cu, Pb, Ni, Hg, and Cd than are found in typical agricultural soils. Berrow and Webber (17) reported the results from analyses of 42 sewage sludges collected from rural and industrialized city wastewater treatment plants in England and Wales. On a dry matter basis they found the sludges contained consistently greater concentrations of Ag, Bi, Cu, Pb, Sn, and Zn than are present in typical agricultural soils. In a small number of sludges, B, Co, Mo, Cr, and Ni were present in sludges at greater concentrations than found in typical soils. They correctly point out that the amount of trace elements present in soluble or available form is more important in relation to uptake by plants than is the total content. Thus, they assessed the solubility of several trace elements by extracting with 2.5 percent acetic acid. In Table 2 their extractability data and some of ours are presented by decreasing solubilities of several elements in soils. These data and others from our field studies confirm our earlier opinions that we must be mainly concerned with first six elements presented in Table 2 when municipal waste are utilized as a fertilizer.

On the basis of total and extractable concentrations of trace elements in sludges, Berrow and Webber speculate that where sludges are used over a period of several years to fertilize crops some of the accumulating trace elements may give rise to toxicity problems in plants. From the results of chemical analyses of samples collected from soils contaminated with trace elements by air pollution and the use of municipal compost and sludges, Purves (123) speculates that a "general enhancement of the level of potentially toxic trace elements in plants grown in urban areas could lead to deleterious effects both on the plants and on the health of those eating them." During five years of research using digested sludge we have not yet created trace element toxicities in various feed grain and forage crops nor have levels of any element increased in plant tissues to the extent that they would present a hazard to animals consuming the produce. Furthermore, LeRiche (87) analyzed soils and crops from a market garden experiment at Woburn, England where 1278 t/ha of sludge had been applied between 1942 and 1961. While there was an increase in the uptake of some elements by vegetable crops grown on the sludge-treated plots, as can be seen in Table 3 from the average values of his reported results, he reported that there was no evidence that crop yields were affected.

Table 2. Average concentrations of trace elements extracted by 2.5 percent acetic acid from 42 sludges collected in England and Wales (17) and percent of total amount present that was extractable.

Element	Mean extractable as ppm d.w.	Mean % soluble of total content
*Cd	144	65
Mn	300	56
Ni	190	46
Zn	1540	44
Co	8.8	32
**B	10	25
Cu	96	6.9
V	3	4.8
Cr	22	3.1
Fe	650	2.8
Pb	20	2.8
Mo	0.12	1.9
Sn	0.58	0.5

* Unpublished 0.1 N HCl data

** Hot water extractable

The behavior of such trace elements in soils and their uptake by crop plants are influenced by several factors. One of these is soil pH. Most heavy metal toxicities in terrestrial plants have been associated with pH values of less than five. Liming soils can, to a large extent, control the uptake of many trace elements.

Practices which promote better soil aeration, such as drainage and structure development may lead to decreased solubilities of some trace elements. According to Jenne (77) oxides of iron and manganese act as "sinks" for heavy metals and the extractability or leachability of the metals is determined by the Eh (reduction-oxidation potential) and pH of the system. Keeping the Fe and Mn hydrous oxides in soils and sediments in the form of thin coatings on silicate minerals instead of discrete crystalline minerals permits a chemical activity in far greater proportion than would be expected on the basis of their concentrations

Table 3. Availability of trace elements and their uptake by vegetable crops growing on a soil treated with 568 tons/acre of sewage sludge (LeRiche, 1968, Harpenden, England).

				Parts per million (dry matter)						
				Co	Cr	Cu	Mo	Ni	Pb	Zn
0.05N HOAc Extractable	Sewage 1958			--	4.5	18	--	51	3.5	750
	Sewage 1959			--	2.5	22	--	49	3.0	850
	Soil 1959		Treated	--	2.8	20	--	17.5	5.0	395
			Untreated	--	0.5	5.0	--	4.3	1.2	87.5
Total Contents	Leeks 1960		Treated	0.16	0.54	15.0	1.10	6.95	1.60	135
			Untreated	0.18	0.71	5.75	0.50	2.0	1.15	46
Total Contents	Globe Beets 1960	Top	Treated	<0.1	1.0	10.0	0.7	16.5	2.6	510
			Untreated	<0.1	0.9	9.0	0.5	3.2	2.4	219
		Root	Treated	<0.1	0.8	18.0	0.3	13.0	1.6	250
			Untreated	<0.1	0.3	11.0	0.1	1.7	0.9	130
Total Contents	Potatoes 1961	Top	Treated	0.35	3.00	8.3	0.98	5.25	2.60	270
			Untreated	0.38	1.70	4.3	0.38	1.70	2.80	90
		Root	Treated	0.02	0.03	9.5	0.28	0.58	0.19	28
			Untreated	0.03	0.09	9.5	0.40	0.25	0.25	30

Table 3 (cont). Availability of trace elements and their uptake by vegetable crops growing on a soil treated with 568 tons/acre of sewage sludge (LeRiche, 1968, Harpenden, England)

				Co	Cr	Cu	Mo	Ni	Pb	Zn
				Parts per million (dry matter)						
0.5N HOAc Extractable	Soil 1967		Treated	--	2.6	58	--	8.1	4.2	275
			Untreated	--	0.9	14.5	--	3.4	1.6	84
Total Contents	Carrots 1967	Top	Treated	<0.08	0.88	9.9	0.85	3.00	1.7	99
			Untreated	0.06	0.41	8.2	0.58	1.14	1.09	48
		Root	Treated	<0.05	0.07	4.6	0.12	2.30	0.07	42
			Untreated	<0.05	0.03	6.3	0.13	1.45	0.06	34

alone. As the solubilities of iron and manganese compounds are increased by reducing conditions, the heavy metals originally adsorbed on the surfaces of their oxides are displaced by hydrogen and the metals become more mobile in soils.

Some heavy metals may form inert and insoluble compounds with clays and organic compounds. Thus, many trace elements are less available to growing plants than the total concentrations of these elements would indicate.

When grown on the same soils, tissues from different crop species, and even different varieties of the same species, differ markedly in concentrations of nutrient and pollutant elements. The selection of crops or even varieties of a particular crop species thus affords a control over the entrance of undesirable amounts of trace elements into food chains. With regard to selection, Gabelman (50) says, "The ease of discovery of these genetic differences within species has been surprising. We have been too conservative in assessing this potential."

Perhaps we have not observed trace element toxicities in plants by the use of stabilized sludge because it may contribute toward establishing a better balance of nutrient availability and uptake by crop plants. We have learned from greenhouse and other studies that there are many synergistic and antagonistic interactions between various ionic metal species in sludge and soils affecting the absorption of chemical elements by plant roots and their translocation within plants. As we learn more about interaction effects, we may be able to decrease abnormal uptake of one trace element from soils by supplying another to the soil or crop.

Clearly if or when a trace element problem does occur as a result of utilizing municipal sludges as a fertilizer and/or soil amendment, there are management practices available which can be introduced to alleviate the situation. Except perhaps in coarse sandy textured soils, the heavy metals will move very little with percolating water. Thus, most of the trace elements will remain at the point of application unless they are transported away in an adsorbed phase on eroded sediments. By establishing erosion control structures and practices, complete control can be maintained over all elements applied on land as a constituent of sludge except some anion and anion forming species such as nitrate, sulfate, chloride, borate, etc. At any rate, those chemical elements which present the greatest potential hazard to animals will be retained in place and can be managed if the need develops. To a large extent the opportunity to manage trace elements is lost once they are disposed of in water environments like the ocean or in air by incineration, or by storing ash residues in landfills where they can be leached by percolating water.

Biological - Although no incidence of disease is known to have been traced to the use of digested sludge as a fertilizer or soil amendment, it is still one of the greatest sources of concern for many. From a rather extensive literature survey it appears that most of the intestinal pathogenic bacteria are either destroyed or their populations are reduced to very low levels by heated anaerobic digestion of sewage solids. Results from several studies indicate that the pathogenic organisms of tubercle bacillus, *Taenia saginata*, *Ascaris lumbricoides*, and hookworm are not destroyed as rapidly in a heated anaerobic digester as are the commonly used pathogenic indicator organisms, *Escherichia coli* or fecal coliforms.

One of the most crucial questions which could not be answered from a search of the literature was that of the fate of viruses during the anaerobic digestion of sewage solids. Even if viruses were not recovered from digested sludge, one could not be sure that they were not present in an adsorbed phase on the solids. To answer the question regarding the survival of viruses in the heated anaerobic digester environment we initiated some laboratory studies using a swine enterovirus (ECPO-1) which has bio-physical properties similar to human enteric viruses. After gas production had stabilized in six laboratory scale digesters fed with a mixture of primary and waste-activated sludge, they were inoculated with 10^5 plaque forming units of the swine virus. After inoculation, 20 ml of fluid were periodically withdrawn from the digesters and mixed with milk and fed to germ free piglets. The feces from the piglets were then collected and assayed for the viable virus. The viruses were not found in the feces of piglets fed sludge material which had been inoculated and digested for a period of time of five days or longer (108). It thus appears that a 14-or 15-day heated anaerobic digestion period would provide a considerable margin of safety with regard to the destruction of viruses.

As Berg (16) suggested, perhaps the simplest method for reducing viruses and other pathogen organisms in sewage is by long storage of the material. From laboratory studies, Berg (16), determined the time in days required for a 99.9 percent reduction in the number of viruses and bacteria by storage at different temperatures. The die-away data presented in his Table 5 are exhibited here as Table 4. On the basis of these and other data, it appears that an additional margin of safety against pathogenic contamination of the environment could be achieved by holding digested sludge in reservoirs for a minimum period of two months before it is applied on land.

After sludge is applied on the soil surface, die-away of many pathogenic organisms will occur rapidly as seen from the data in Table 5. The rapidity with which fecal coliform die-away occurs after digested sludge is applied on soil surfaces can be discerned in Table 5. Furthermore, it has generally been concluded that wastewaters percolating through unsaturated soil materials are purged of pathogenic organisms within the 1.5 m depth (30). If this is true for wastewater applications, one would

Table 4. Effects of storage: Laboratory study demonstrating days required for 99.9% reduction of viruses and bacteria in sewage (16).

	No. of Days Temperature ° C		
Organism	4°	20°	28°
Poliovirus 1	110	23	17
Echovirus 7	130	41	28
Echovirus 12	60	32	20
Coxsackievirus A9	12	--	6
Aerobacter aerogenes	56	21	10
Escherichia coli	48	20	12
Streptococcus faecalis	48	26	14

Table 5. Disappearance of fecal coliforms in a sludge cake covering a soil surface (unpublished data, Agron. Dept., Univ. of Illinois).

Days after sludge application	No. of fecal coliforms per gm sludge cake (dry weight)
1	3,680,000
2	655,000
3	590,000
5	45,000
7	30,000
12	700

expect it to be applicable in the case of sludge utilization. Since sludge solids are rapidly filtered and clog the surface of soils, the rate of water infiltration during sludge applications is exceedingly low in comparison to that from wastewater applications. Therefore, frequent applications of sludge can be made only when the evapotranspiration potential is relatively high. That is to say, sludge is most likely to be applied on agricultural lands during the late spring, summer, and early fall seasons when evapotranspirational potentials generally exceed actual soil moisture losses. For the most part, sludge will be applied when ambient temperatures favor a rapid-die-away of bacterial and viral pathogenic organisms.

Like many of the potential chemical water pollutants, lateral movement of pathogenic organisms which might survive the digestion and storage period, can occur only if excessive soil erosion processes are permitted to operate on the sludge utilization site. Thus, pathogenic pollution of surface waters can be avoided by the same structural and management practices recommended for the conservation of soil and water.

In 1966, after an extensive study of various alternative methods for handling the daily production of 819 metric tons of sludge, staff members of the Metropolitan Sanitary District of Greater Chicago concluded, on the basis of the information available, that the most economical and environmentally satisfactory method of sludge disposal was to utilize it as a fertilizer and soil amendment. Toward obtaining further information with regard to the utilization of sewage sludges, members of the Agronomy Department, University of Illinois, were requested to submit a research proposal to MSD of Chicago. The proposed research was to be broad enough to include all aspects of sludge utilization in which there were insufficient information to predict environmental changes resulting from a large scale operation. With the paucity of pertinent information available in the literature when the proposal was submitted to MSD of Chicago in early 1967, many questions needed to be answered before a large scale operation could be initiated. Foremost among the several aspects needing further study was that of excessive trace element accumulations in soils with continuous applications of digested sludge. At least up to the time our project was initiated, agronomists had been largely concerned with difficiencies of trace elements in soils. Very little attention had been given to determining the effect on plants of unusually high rates of application of essential trace elements on soils, and even less with regard to nonessential trace elements.

Very little information was available in the literature to assess the relationship between sludge utilization and the potential for increasing incidents of diseases caused by pathogenic organisms.

Changes in the physical, chemical and biological properties of soils and their runoff and drainage effluents with continuous annual applications of sludges could not be predicted from results reported in the literature.

To determine environmental changes resulting from the use of sludge as a fertilizer and to obtain predictive information with regard to the capacity of soil and crop systems to assimilate various constituents of sludge, the proposed studies were centered on the establishment of a large field lysimeter facility. From the lysimeter facility plant, soil and water samples would be collected from three different soil types planted to different crops fertilized with annual applications of sludge at various loading rates.

After the cooperative project between the University of Illinois, the Metropolitan Sanitary District of Greater Chicago, and U.S.H.E.W. Office of Solid Waste Management was funded in April 1967, one of the first major tasks was to plan and design the field lysimeter facility and attendant equipment. Plans and specifications for the field research facility, which included a series of lysimeters and an associated instrument house, were prepared and submitted with the requisition to the University Purchasing Division on June 6, 1967. Construction of the instrument house was completed on September 15, 1967, and the lysimeters were completed on June 18, 1968. Thus, it was not possible to harvest a crop from the lysimeter facility until the 1969 growing season.

SECTION IV

FIELD LYSIMETER STUDIES

Northeast Agronomy Research Center Lysimeter Facility

General description - The field lysimeter facility was constructed on a small isolated watershed on the Northeast Agronomy Research Center where the original soil type was Blount silt loam. The soil is underlain by glacial till of very low permeability from about 76.2 cm below the soil surface to a depth of about 12.2 m. An overall plan view of the field research facility is presented in Figure 1. The facility consists of 44 lysimeter plots, each 15.2 m long and 3.1 m wide. The 44 plots are equally divided into 2 blocks, one each on the north and south side of the instrument house. Each block contains 12 plots of the original Blount silt loam; five plots of simulated Elliott silt loam, and five plots of simulated Plainfield sand. Since the Elliott silt loam soil is a prairie equivalent of the forested Blount silt loam, the simulation of a prairie soil was made by removing the Blount silt loam surface to a depth of 30 cm and replacing it with the surface of Elliott silt loam. Plainfield sand was simulated by excavating all of the original material within the boundaries of a plot to a depth of 1.5 m and then filling the pit with Plainfield sand.

A trenching machine was used to excavate a trench around the perimeter of each plot to a depth of 1.8 m. A continuous curtain of nylon reinforced 0.02 cm black plastic film was usspended from 20.3 cm below the soil surface to a depth of 1.8 m in the trench with the ends overlapped a minimum of 1.2 m. This moisture barrier was secured by spikes to the inside wall and the trench was backfilled with soil removed during excavation. A single line of 10.2 cm diameter clay tile was installed at a bottom depth of 86.4 cm through the longitudinal center of each of the Blount and Elliott silt-loam plots. Construction of the sand plots was somewhat different in that the line of clay tile was installed at a depth of 15 m and the walls lined with the plastic moisture barrier before the pit was filled to ground level with Plainfield sand.

To convey drainage water from the plot end nearest the instrument house, a 10.2 cm diameter rigid plastic (PVC) drain tube extends from each plot to within 61 cm of the basement and 76.2 cm above the basement floor of the instrument house. The plastic tube is attached to the clay tile by an adapter just inside of the plastic moisture barrier. A mastic material was used to achieve a waterproof seal where the plastic tube passed through the moisture barrier.

A second 10.2-cm PVC tube to convey runoff water extends from 61 cm inside and 168 cm above the basement floor to within 15.2 cm of the moisture barrier at each of the plot center ends nearest the instrument house. The end of the PVC tube at the end of a plot is located 45.7 cm below the soil surface and connected to a 90 degree elbow positioned in an upward direction to receive the thimble of the runoff water collection trough.

Each of the 88 plastic tubes for drainage and surface runoff water conveyance is placed on the maximum uniform grade obtainable from a plot to the basement of the instrument house. Grade is not less than 0.5 percent for tubes serving any plot.

The runoff water collection troughs are of a design similar to those used in soil erosion studies, except they are fabricated from fiber glass. Fiber glass was chosen as the construction material to avoid the introduction of heavy metals in the runoff water and soils. For the same reason, fiber glass strips 25.4-cm wide are used to completely enclose the plots along and above the moisture barrier discussed above. With the fiber glass strips installed in the soil to a depth of 15.2 cm the collection at the down slope end of all the runoff water from a plot is insured while excluding all water from outside the plot.

On all of the lysimeter plots, the lowest point is the end toward the instrument house. Thus, runoff water flows toward the instrument house from both the north and south blocks of lysimeter plots.

The instrument house was constructed in a natural depression of the slightly greater than 0.8 ha watershed. The 16.7 x 3.65 x 2.44 m eave-height frame building was constructed on a concrete first floor over a poured steel reinforced 20.3-cm thick walled basement, which stands 2.9 m high above the footings. Ten 10.2 cm diameter bell and spigot floor drains were installed in the basement floor to conduct unwanted water discharged from the plot drainage tubes to a 20.3 cm diameter tile installed below the center of the basement floor.

Heating is provided by 2 wall-mounted thermostatically controlled 220 volt electric heaters at each end of the first floor of the instrument house. One end of the first floor of the building was later partitioned, to provide a totally air-conditioned room to protect instrumentation circuitry from wide variations of temperature and humidity.

The 20.3-cm diameter clay tile line used to drain excess water from inside the basement of the instrument house and the basement footing tiles is connected to a surface inlet located in the natural drainage way of the small water shed 46.5 m west of the instrument house. Behind the surface inlet, a small earthern dam was constructed to insure the capture of all runoff water from areas outside of the lysimeter plots. Thus, all water from the research area is disposed of through the 20.3-cm diameter tile line that conducts water through a 5680 liter septic tank and finally to a sand and gravel filter field. All water from the research area is filtered through 76.2 cm of sand and gravel before it is discharged to a small stream that flows intermittently.

To provide water for the instrument house and for irrigation of lysimeter check or control plots, a well was drilled to a depth of 61 m. However, since a sustained flow of only 22.7 liter of water per minute was obtained from the well, two used 37,900 liter capacity railroad tank car containers were buried near the well to store water for irrigation. Two other used plastic-lined 30,320 liter capacity railroad tank car containers were buried end to end with the water tanks to provide storage for digested sludge. The two water tanks were connected with 7.6 cm diameter metal pipe and a 227 liter per minute capacity pump was mounted on the end of one water tank. One two-stage vertical turbine pump with a capacity of 1,500 liter per minute was mounted on the ends of each of the separate sludge tanks. Both the water and sludge pumps develop heads of about 54.9 m. All pumps, motors, and exposed plumbing were enclosed in an insulated, propane-heated pump house. Metal pipe of 7.6-cm diameter was used for all plumbing inside the pump house. By use of check and gate valves, the plumbing from the pumps was installed in such a manner that the main irrigation line can be used to supply plots with either water or sludge. Also, sludge can be circulated in the same storage tank or be pumped from one sludge storage tank to the other for mixing.

PVC pipe (7.6-cm diameter) was used for the main irrigation line which was installed at a minimum depth of 45.7 cm below the soil surface. As may be seen from Figure 1, the main irrigation line was extended from the pump house through the east-west center border of the north block of plots, to the west side of the instrument house and then returned to the pump house through the center east-west border of the south block of plots. The irrigation system was so designed that a large return flow of sludge could be maintained to keep solids in suspension in the storage tank and also prevent settling of solids in the irrigation pipe. It may also be noted from Figure 1 that a "T" joint was installed in the main irrigation line west of the instrument house by which means the line was extended to the surface inlet discussed above. The main irrigation pipeline was laid on a uniform grade of approximately 0.5 percent from 6.1 m west of the pump house to the surface inlet so that the line could drain when the gate valve at the surface inlet was opened. Plumbing inside the pump house is so arranged that, after irrigation of plots receiving sludge treatments, the gate valve at the surface inlet may be opened and the north and south portions of the irrigation line may be alternately flushed and cleaned with water. The irrigation line must be flushed with water each time before irrigating check plots with water.

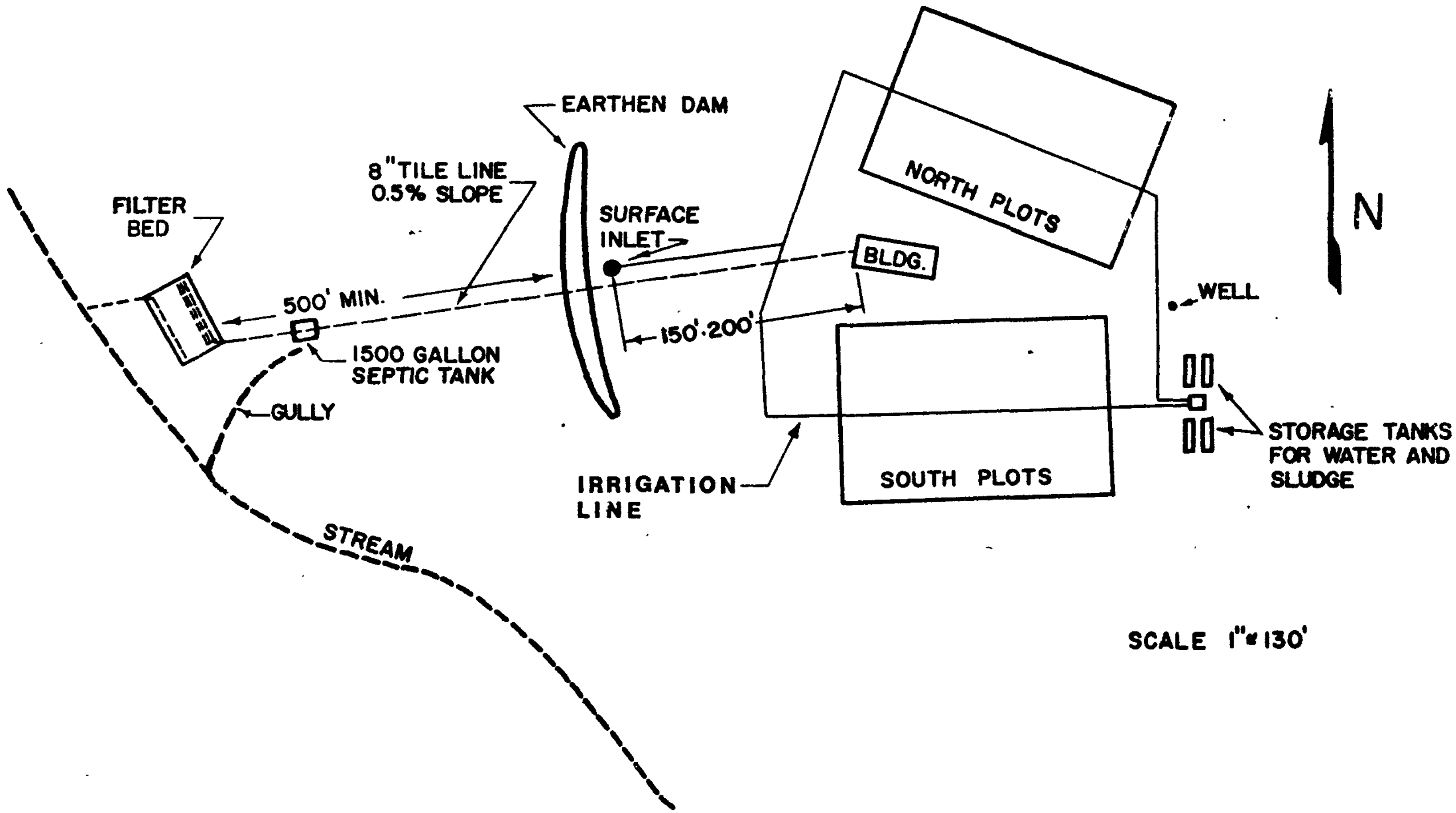


Figure 1. Schematic layout of field lysimeter research facility at the Northeast Agronomy Research Center, Elwood, Illinois (in. x 2.54 = cm; ft x 0.305 = m; gal. x 3.78 = 1).

Risers were installed in the main irrigation line through the north and south block of plots so that one riser, by means of a valve and key, could supply either water or sludge to irrigate any one of four plots.

Although irrigation equipment is commercially available for field applications of digested sludge, equipment for making uniform applications on small research plots could not be obtained. Thus, a self-propelled irrigation machine for uniformly applying digested sludge on the 3.1 x 15.2 m plots was designed and constructed specifically for the research project. Flexible tubing (5-cm diameter) is used to convey sludge or water from the risers to the irrigation machine. However, during the first year of operation when plots were irrigated 2 to 3 times each week during the growing season, it was observed that plants being continuously coated with sludge by spray irrigation were stunted. Sludge particulates coated leaf surfaces and blocked light absorption to such an extent that photosynthesis rates were reduced to very low levels. To avoid coating leaf surfaces, the method of sludge application was later changed to the ridge and furrow system.

<u>Description of instrumentation for volumetrically measuring and sampling runoff and drainage water from field lysimeter plots</u> - To collect descrete samples of runoff and drainage water from lysimeters of field-plot size, an electrically controlled sampling system was designed, constructed, and put into operation. The system is used to measure rate and total flow of both runoff and drainage water from each of the 44 lysimeters and to collect 400 ml samples after selected volumes of flow have occurred. The sampling instrumentation can be controlled to collect a sample from as little as 2.4 up to 50 percent of the total flow.

The collection and monitoring system consists of five major components: (1) tipping bucket, (2) sample collector mechanism, (3) electrical circuits for counting and control, (4) event recorders, and (5) automatic turn-on and turn-off system. Except for some common circuit elements schematically shown in Figure 2, each major component was duplicated eighty-eight times to provide complete instrumentation for the forty-four lysimeters. All of the instrumentation is located on the ground floor of the instrument house, except for tipping buckets and sample collectors which are positioned below the end of 10.2-cm diameter plastic pipes used to convey runoff and drainage water from the lysimeters to the basement of the instrument house. The arrangement of the tipping buckets and sample collectors may be seen in the cross-sectional view presented as Figure 3.

A tipping bucket consists of two end pieces, a divider, two studs, and two counterweight brackets all made from stainless steel and fabricated by welding. The studs welded on the sides below the center line of the buckets serve as axles. When a tipping bucket is mounted in its frame, as shown in Figure 4, the axles are inserted in holes in teflon pads that serve as bearings. The holes in the teflon pads were drilled just slightly larger than the diameter of the axle studs. The portions of

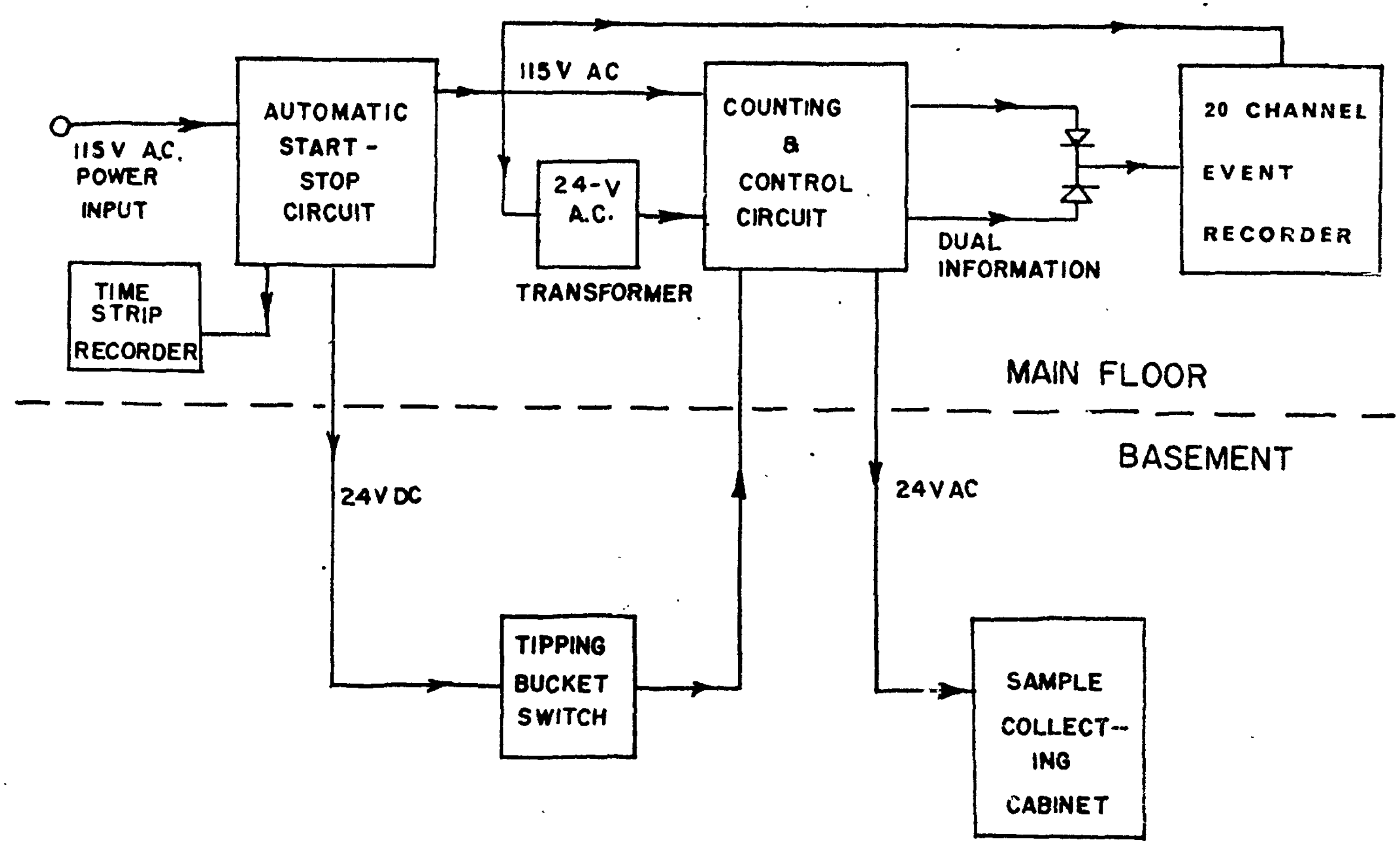


Figure 2. Schematic diagram of system for data and sample collection.

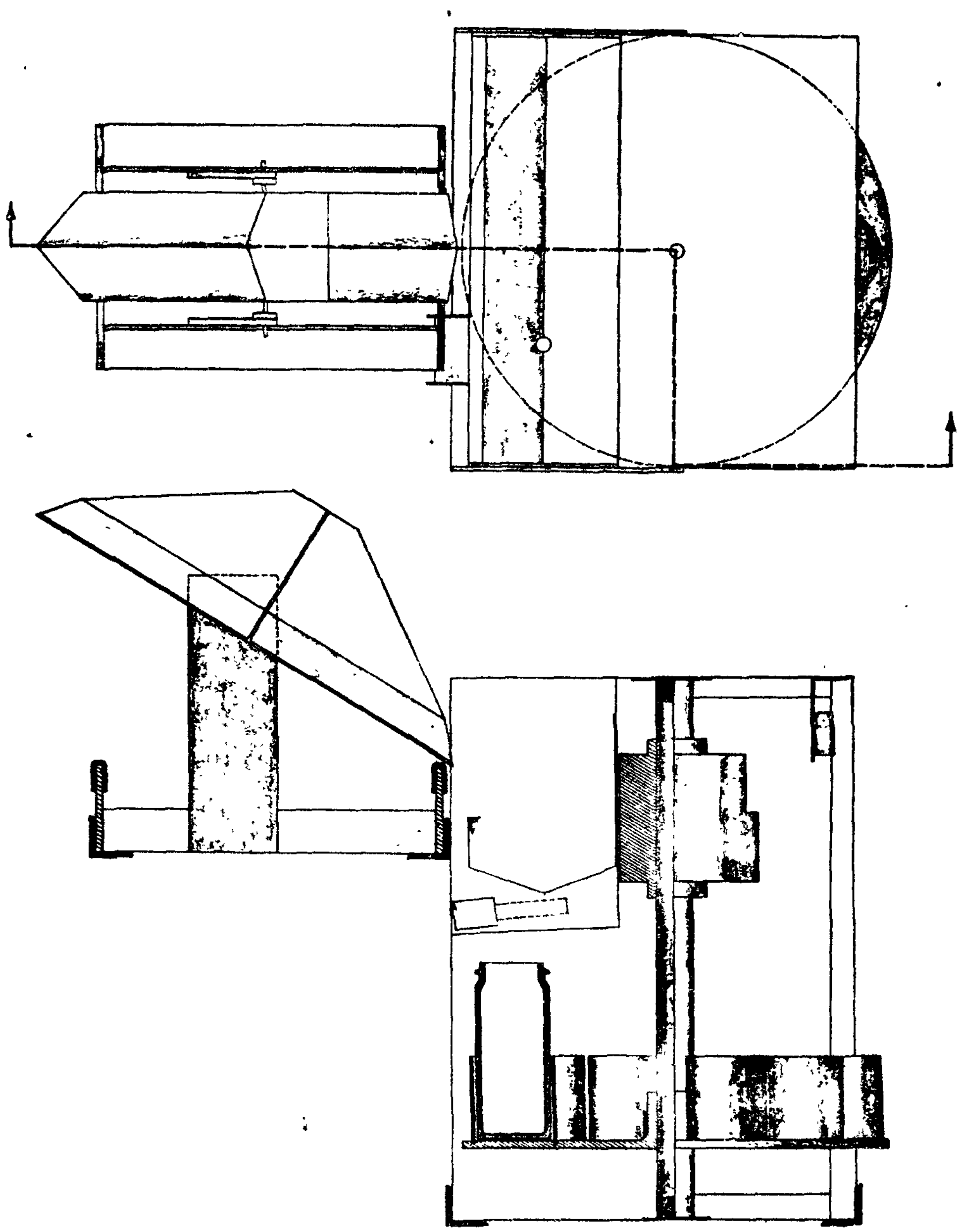
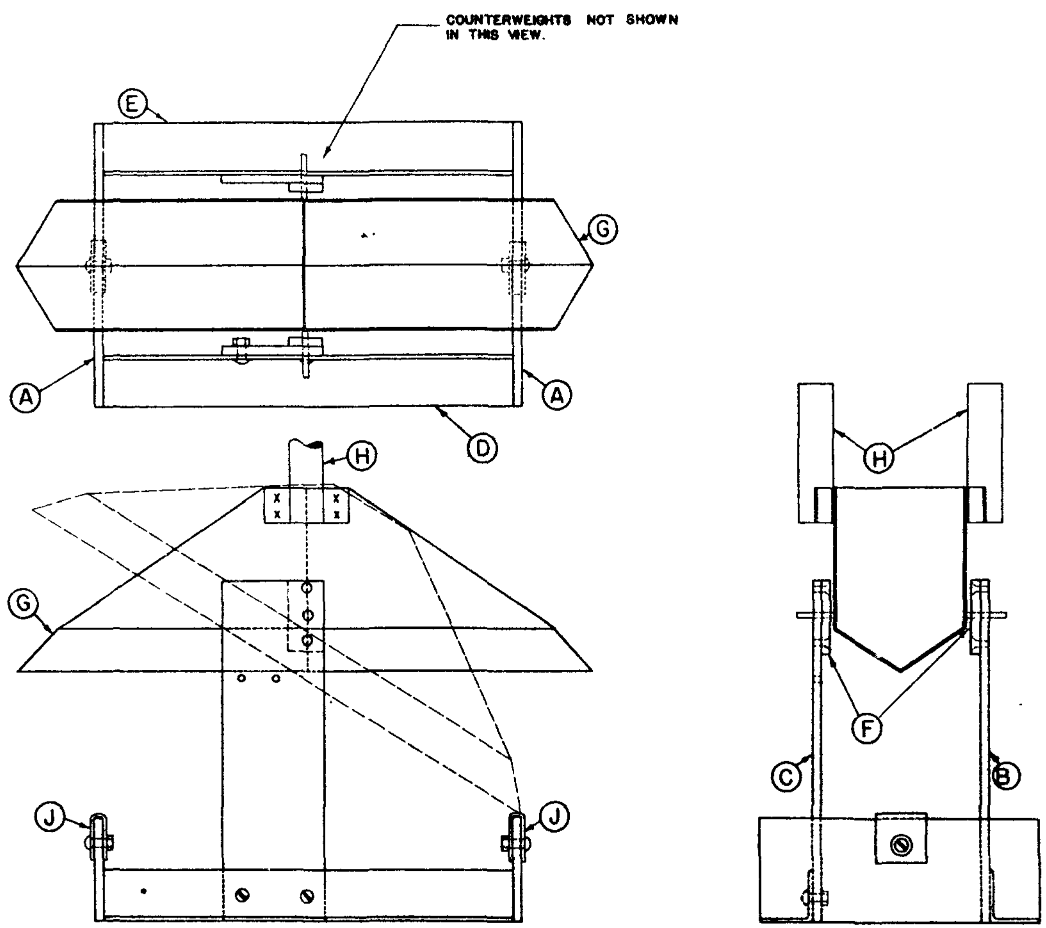


Figure 3. Top and side views of tipping bucket and sample collector assembly.



COMPONENT	QTY	MATERIAL
A END PLATE	2	MILD STEEL
B VERT. PLATE (WELDED)	1	MILD STEEL
C VERT. PLATE (BOLTED)	1	MILD STEEL
D BASE ANGLE (BOLTED)	1	ANGLE IRON
E BASE ANGLE (WELDED)	1	ANGLE IRON
F BEARING	2	TEFLON
G BUCKET	1	STAINLESS STEEL
H COUNTERWEIGHT	2	BRASS
J PAD	2	RUBBER

Figure 4. Top, side and front views of tipping bucket used in water collection.

the threaded studs in contact with the teflon bearings were sheathed with shrinking electrical spaghetti (a heat sensitive plastic tubing), therefore, frictional forces between the plastic covered axle studs and the teflon bearings are small. Also, since the teflon pads are mounted on the inside of the framework which supports the tipping bucket with at least 0.32-cm clearance in the direction of the axles, the tipping buckets are never in contact with the metal frame. The tipping buckets are restrained in their framework only by occasional bumping against the teflon pad. Although mercury switches are mounted on one end of the axle studs, they contribute very little frictional forces. The mercury switches do not come in contact with the metal frames and the electrical leads are coiled in such a manner between their points of attachment to the frame and the switch terminals that they present a minimal restraint to the pivoting of the buckets. However, because the mercury switches deteriorate under the corrosive conditions of the instrument house basement and result in malfunctions of the sampling equipment, they are gradually being replaced with a light source and photocell switch. Except for modification of the light to be reflected onto the photocell during the pivotal motion of the tipping bucket, the photocell switch is similar to that to be described later in connection with the water sampling equipment.

During calibration of the tipping buckets for volume versus flow rate or tipping rate, it was observed that when all buckets were adjusted to yield the same volume of water when tipped at a dropwise flow rate, one calibration curve for a uniform given size of buckets could be used for data interpretation. During the filling of a bucket, the moment due to the fluid increases until it exceeds the balancing moment of the counterweight causing the bucket to tip. Thus, by adjusting the moment of the counterweights, the volume of water that the buckets will collect can be varied. The counterweights are lengths of brass bar stock which have a 3-cm vertical slit cut through the center of one end. When the counterweights are mounted on the tipping buckets by means of the slits and the brackets welded on each side of the top center of the tipping buckets, their moments are adjusted by raising or lowering on the brackets. The counterweights are then held in position by tightening two set-screws. The counterweights are also held in a stable perpendicular position with respect to the top edge of the tipping buckets by the fact that the one-half diameter portion of each counterweight installed between the bracket and bucket walls, as shown in Figure 4, is a press-fit.

When a bucket tips, its bottom strikes and comes to rest against a pad (J in Figure 4) made of rubberized belting material bolted to the tipping bucket frame. The pad cushions shock to the tipping bucket axle and reduces noise level. Shock to the tipping bucket axles can be reduced to a minimum by placing the cushioning pad at the center of percussion, after it is located by trial and error for a normal flow rate.

Figures 5 and 6 are the calibration curves for the large and small tipping buckets. The large and small tipping buckets are used for measuring volume and flow rates of runoff and drainage water, respectively. The large buckets are adjusted to hold 1725 g and the small buckets 585 g before tipping at a minimal flow rate. At high flow rates (tips per second) the volume, or weight per tip, increases. The dashed lines in Figures 5 and 6 are the upper and lower ranges of values from the average values represented by the heavy line. It appears that the volume delivered by a tip at a given flow rate varies more between small buckets than for the larger buckets, although, except for size, fabrication was similar.

Volume is unpredictable for flow rates that produce more than one tip per ten seconds of small buckets. However, it is not expected that drainage flows from the lysimeters will ever be great enough to produce small bucket tipping rates greater than one tip per 10 seconds.

The automatic sample collecting mechanism shown in Figure 7 will take ten samples before full sample bottles are replaced with clean, sterilized empty bottles. The sample collector mechanism (Figure 7) consists of the following:

1. A motor driven turntable (E) that holds ten 450 ml widemouthed bottles.
2. A pair of enclosed microswitches (V), one of which stops the rotation of the turntable so the sample bottle is correctly positioned to take a sample, while the other is used as a means to control the volume of sample collected in the bottle.
3. A solenoid (X) operated flow control mechanism (M) which diverts the water collected in a reservoir or trough (A), during a tip of a tipping bucket either to a sample bottle or the basement floor drains.

The turntable is moved by a modified antenna rotator motor (Aliance Model T-45) that rotates about one RPM. All motors were modified by removal of the mechanical stops and circular wire wound resistors and associated sliding contacts. One further modification was the installation of the phase shift capacitor inside each motor housing; a component which is normally located in the control box.

The motor (W) is mounted (with clamps provided) on a length of pipe with a diameter of 3.16 cm which serves as a shaft (P) for the turntable. The shaft is mounted through the hub at the center of the turntable wheel. The turntable is secured to the shaft by two set screws in the hub. The shaft and motor assembly is held in place by stainless steel bearings (Q) which fit inside the shaft ends and are attached near the center of the top and bottom of the cabinet or frame. The turntable consists of the wheel, bakelite cylinders, and brass plate springs.

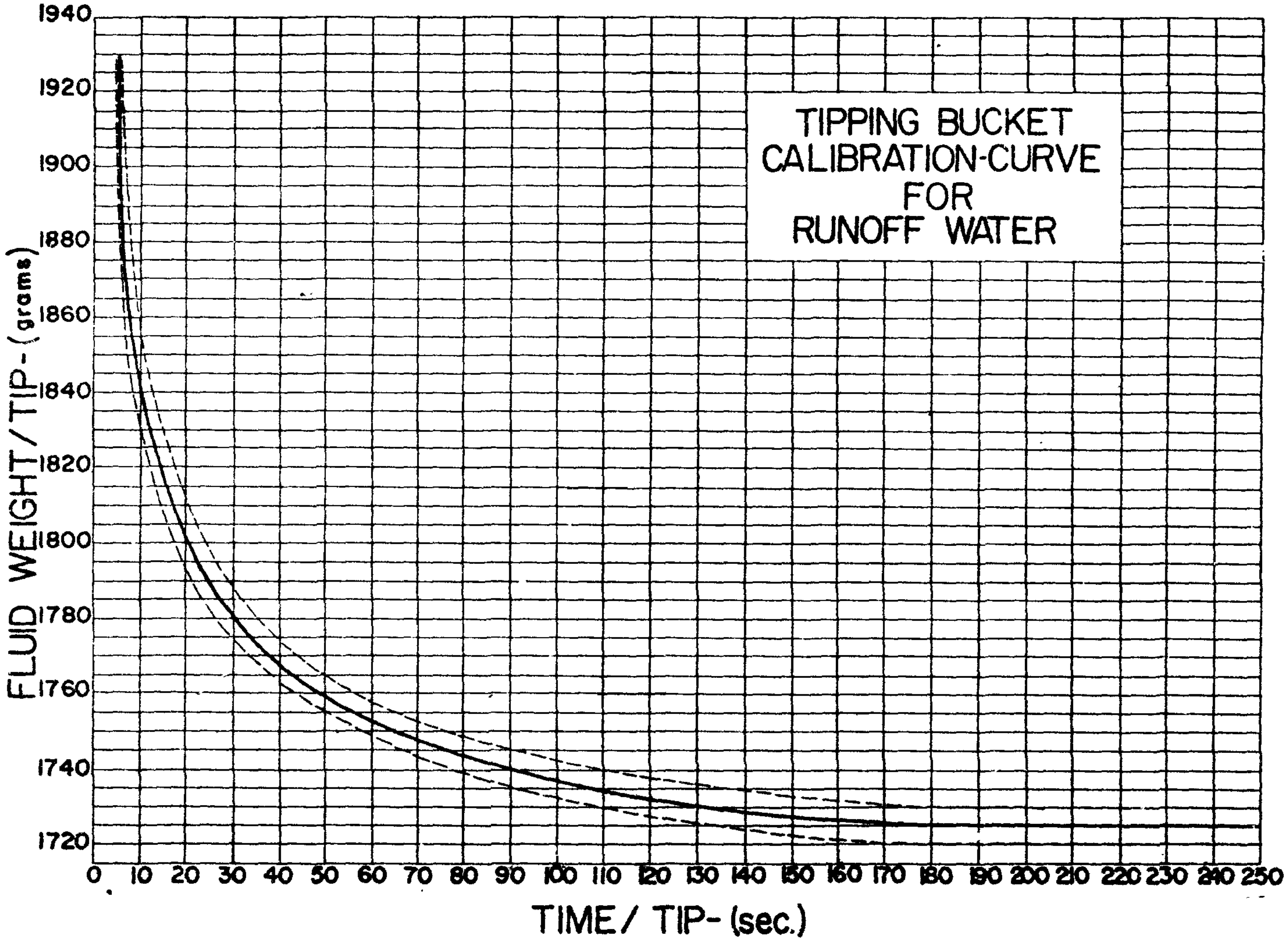


Figure 5. Weight of liquid contained in buckets at various rates of tipping.

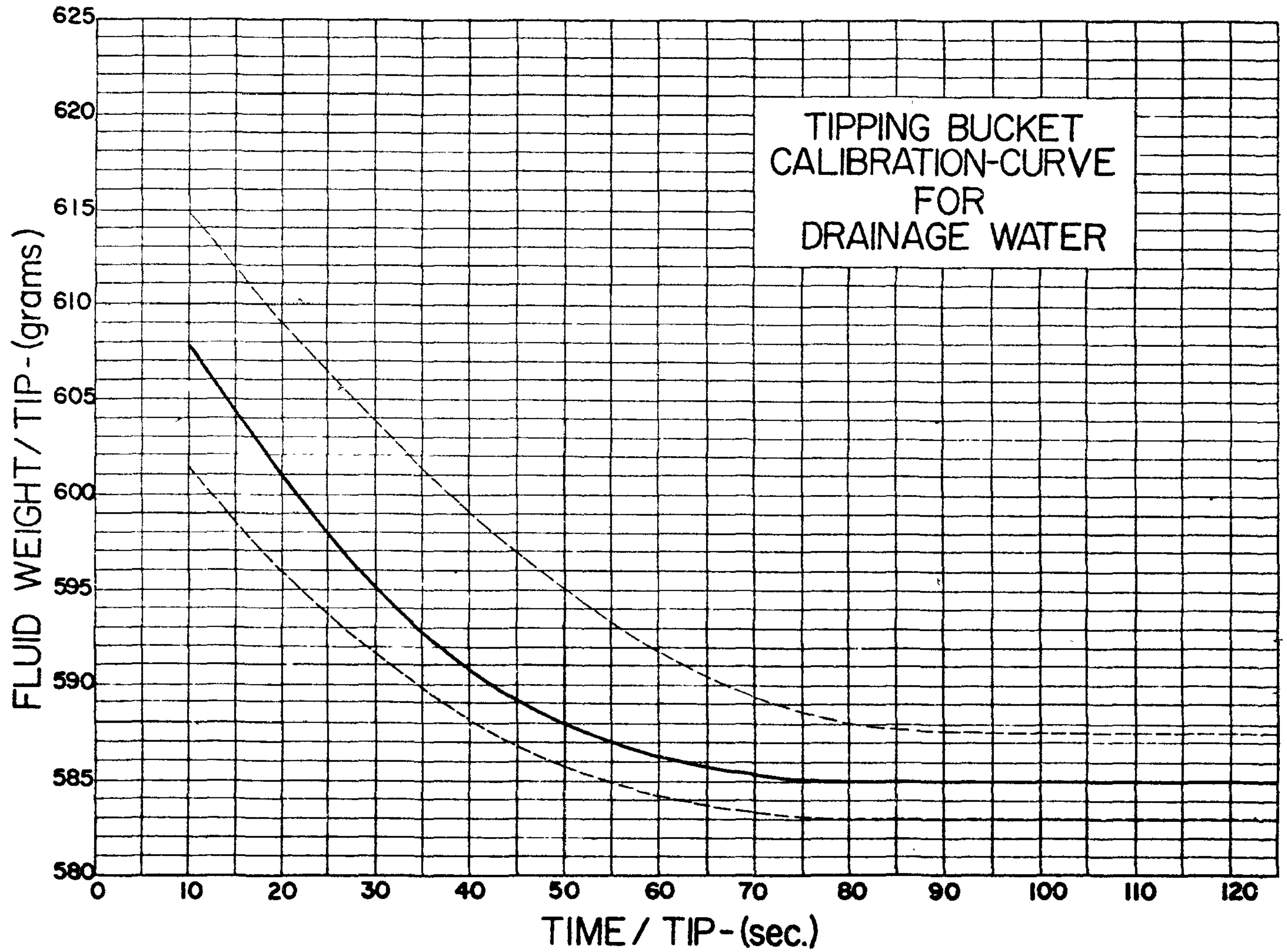
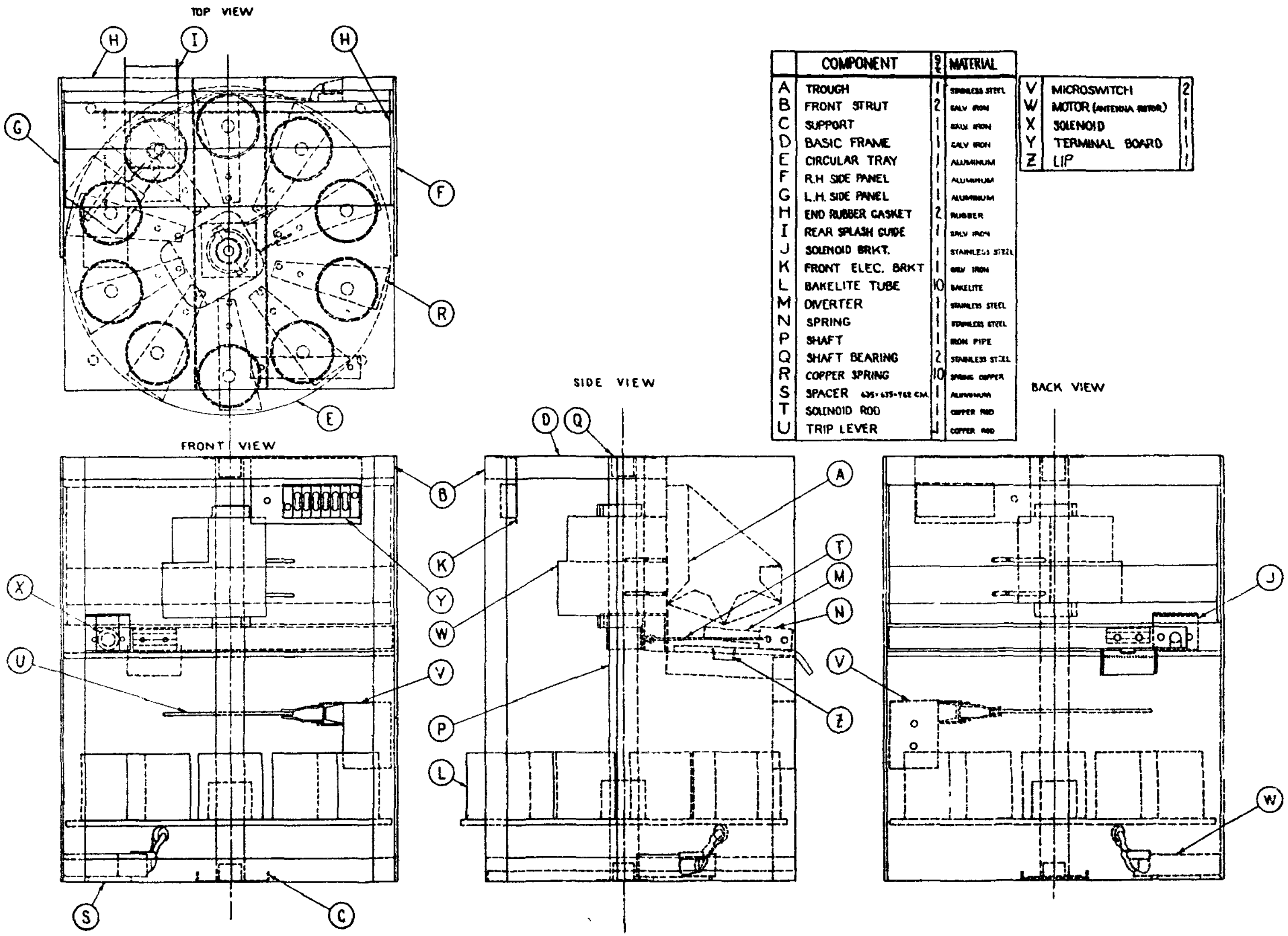


Figure 6. Weight of liquid contained in buckets at various rates of tipping.



	COMPONENT	QTY	MATERIAL
A	TROUGH	1	STAINLESS STEEL
B	FRONT STRUT	2	GALV IRON
C	SUPPORT	1	GALV IRON
D	BASIC FRAME	1	GALV IRON
E	CIRCULAR TRAY	1	ALUMINUM
F	R.H SIDE PANEL	1	ALUMINUM
G	L.H. SIDE PANEL	1	ALUMINUM
H	END RUBBER GASKET	2	RUBBER
I	REAR SPLASH GUIDE	1	GALV IRON
J	SOLENOID BRKT.	1	STAINLESS STEEL
K	FRONT ELEC. BRKT	1	GALV IRON
L	BAKELITE TUBE	10	BAKELITE
M	DIVERTER	1	STAINLESS STEEL
N	SPRING	1	STAINLESS STEEL
P	SHAFT	1	IRON PIPE
Q	SHAFT BEARING	2	STAINLESS STEEL
R	COPPER SPRING	10	SPRING COPPER
S	SPACER 635-635-762 CM	1	ALUMINUM
T	SOLENOID ROD	1	COPPER ROD
U	TRIP LEVER	1	COPPER ROD

V	MICROSWITCH	2
W	MOTOR (ANTENNA ROTOR)	1
X	SOLENOID	1
Y	TERMINAL BOARD	1
Z	LIP	1

Figure 7. Schematic views sample collector for water.

The wheel is a disc, 35.6-cm diameter, cut from aluminum plate 0.624-cm thick. Figure 8 shows an aluminum hub welded in the center of the wheel and ten equally spaced circular grooves near the outer edge of the upper surface. The circular grooves, 7.62-cm diameter and 0.32-cm deep, accept bakelite cylinders, 35.6-cm long, that are epoxy cemented into the grooves for the purpose of holding sample bottles.

In the original design, when a sample bottle was in position to be filled it rested on a spring plate directly over a sensitive microswitch (V in Figure 7) which was enclosed in a moisture-tight casement. When the weight of the bottle and its contents were sufficient to overcome the resistant force of the spring the microswitch, through an arm and roller positioned below the spring plate, was tripped. The activated microswitch de-energized the solenoid that operates the flow control mechanism, causing the flow of water to be diverted from the sample bottle to the basement floor drains. Because of the corrosive basement environment, a great deal of time had to be expended in adjusting the tension of the spring plate. Therefore, the mechanically operated microswitch and its associated spring lever plates were replaced by a light bulb and photocell in conjunction with a semi-conductor controlled rectifier. The light bulb and photocell are placed on opposite sides of the sample bottle when it is properly positioned to take a sample. The light source and photocell are arranged by means of a mounting bracket (not shown in Figure 7) so that light striking the photocell passes through only one side of the sample bottle. That is to say, it does not pass through the center of the sample bottle. When the sample bottle is empty, the sensitivity of the electrical circuit is adjusted so that the amount of light striking the photocell is just sufficient to perform as a closed switch to keep the solenoid that operates the water inlet valve in an energized state. When the solenoid is energized it opens the water valve (flow control mechanism, M) under the reservoir (A) allowing water to flow into the properly positioned sample bottle. As the water level rises, it intercepts the light passing through one side of the sample bottle. The light is first intercepted at a point where the sample bottle contains about 400 ml of water. The moment that the water level intercepts the light passing through the bottle, the optical properties of water are sufficient to deflect enough of the light from the photocell to cause its electrical circuit to perform as an open switch, which de-energizes the solenoid causing the water valve to shut off flow from the reservoir to the sample bottle.

The electrical circuit controlling water level in the sample bottles consists of three major components - a light bulb, a photocell and a semiconductor controlled rectifier. The light bulb and a 100 ohm adjustable resistor reduces the voltage in the circuit to not more than 3 volts AC. To gain greater sensitivity in controlling the semi-conductor controlled rectifier, the voltage is converted to DC voltage by a diode (IN55) and filtered with a 200 Mfd electrolyte capacitor. The photocell and a 1.1 K resistor perform as a voltage divider, determining the amount of voltage acting on the gate of the semi-conductor con-

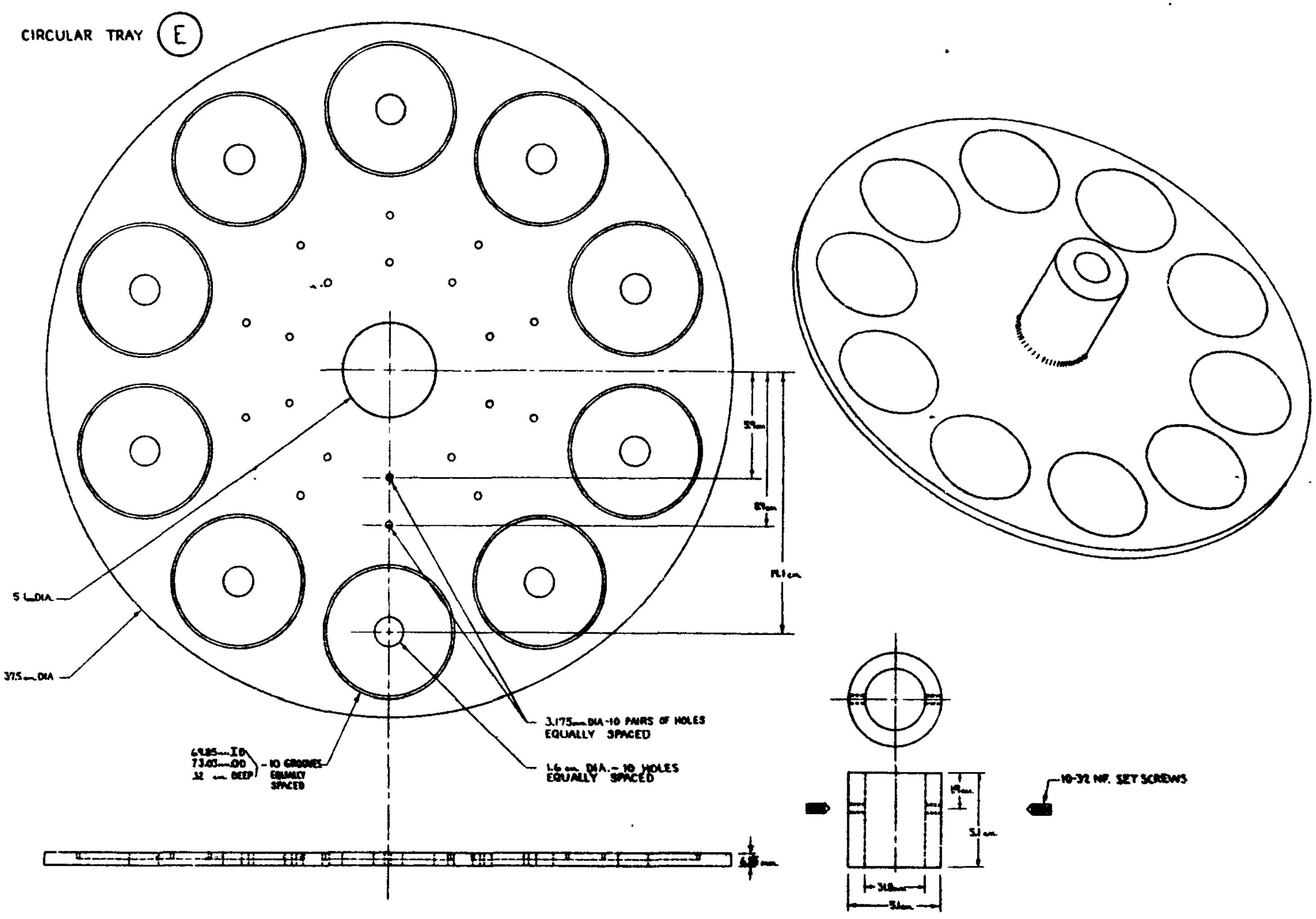


Figure 8. Detail plan view and oblique projection of sample collector turn-table used in water collection.

trolled rectifier. When the gate potential is 0.5 volts more positive or higher than the cathode, current can flow from anode to cathode. When the gate potential is less than 0.5 volts higher than that on the cathode, the effect is that of an open circuit. Since the photocell resistance changes in an inversely proportional manner to the light energy striking its surface, when water in the sample bottle attains sufficient height to intercept and deflect the light from the photocell surface the potential on the gate is reduced to less than the 0.5 volt threshold value. Thus, the open circuit effect produced by deflecting the light cuts off power to the solenoid which, in an energized state, holds the water valve open.

When the turntable motor is energized by the control circuits (to be discussed later) an empty sample bottle is properly positioned to take a sample as it is carried by the turntable contact rod (U) with sufficient force to actuate the microswitch(SPDT), which is mounted on the back wall of the cabinet. This microswitch (V) transfers the 24 VAC power from the rotor motor to the solenoid which operates the flow control mechanism (M). Rotation of the turntable stops and the energized solenoid aligns a series of 3 holes in the flow control mechanism to permit water from the reservoir (A) to pour into the properly positioned sample bottle. When a volume of about 400 ml of liquid has been collected, the photocell switch de-energizes the flow control solenoid, as discussed above.

Power flow from the source to the event recorders and sampling cabinets is sketched diagrammatically in Figure 2. The counting and control circuits for each of 88 sensing and recording units have the same design. Forty-four units are housed in a given rack. The 44 control units in each of two racks are contained in 11 chassis of 4 control units per chassis. The control units in one rack are employed in the measurement and sampling of runoff water, while those contained in the other rack serve the same purpose for drainage effluent.

Three views of an individual chassis containing four control units may be seen in Figure 9. In the front view, each of the four sets of double circles represents a twenty-one position rotary switch and associated dial. Both of the 115 and 24 volt circuits are protected by means of accessible fuses in holders mounted on the front of the chassis. In the top view, the approximate position of four 115 volt magnetic digital counters, one for each of the control units, are shown. Also shown in top view are four 24 VAC indicator lights used for a quick check of power to the sample collection equipment. On the side of the casssis, the input from the automatic start-stop circuit is connected by means of a banana jack. One five connector socket for each of the four control units is located on the back panel along with a six connector socket which serves the four control units as a connection to the event recorders. Jacks for 24 and 115 VAC inputs are also located on the back panel.

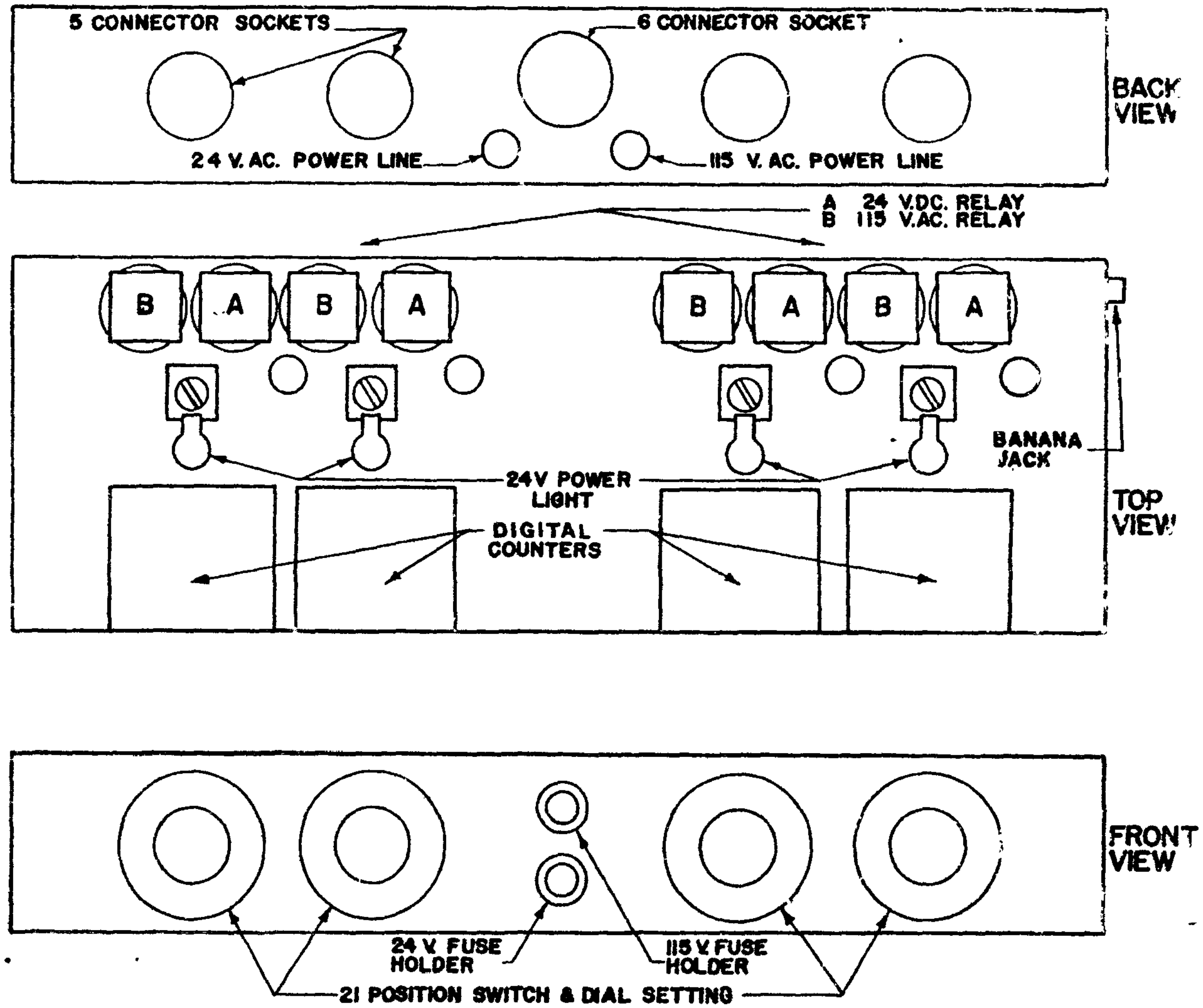


Figure 9. Back, top and front views of control chassis showing component location.

Figure 10 shows the underside or inside view of a control chassis and the correspondence between the control circuits and cable connections for the event recorders.

In Figure 11, a schematic view of a circuit for a control and counting unit is presented. Everything above the dashed line in Figure 11 is common to four identical control units. The 24 VDC power from the automatic start-stop system is supplied to the control unit circuits by means of a banana jack. One banana jack serves as a power link up for four K2 relays contained in a chassis. The current through a given 24 VDC relay (K2) continues to the mercury switch, mounted on the axle of the tipping bucket, with which the particular control unit is associated. The current goes to ground when momentary contact closure of the mercury switch occurs as a result of the tipping of the bucket. Closure of the mercury switch results in a momentary actuation of the K2 relay contacts. The main reason for using 24 VDC power was to give a more positive operation of the primary K2 control relay. When contacts of the K2 relay close, a 115 VAC signal is presented to the digital counter and to the proper channel of one of the event recorders. At the same time, the K3 latching relay is energized. Each time the K3 relay is energized, it reverses contact position so that the stepping relay K4 coil is energized only once for each two times the latching relay K3 is actuated. The alternating action of the latching relay K3 prevents the stepping relay K4 from being energized, except when the tipping bucket tips toward the sample collector reservoir. Thus, the usefulness of the stepping relay K3 contacts are doubled.

To prevent the coil of relay K3 from over-heating when it is energized, a special circuit arrangement using a 10 MFD capacitor is charged through a rectifier diode during the time when relay K2 is energized. Then, when relay K2 is in a relaxed position, the charge on the capacitor is dissipated through the latching relay K3 coil producing a brief but positive actuation.

When the closed contact of the stepping relay K4 is in one-half correspondence with the dial setting of the 21 position rotary selector switch mounted on the front of the chassis, power is supplied to a high resistance DC relay K5. Also, current is supplied through a rectifier diode from the stepping relay K4 to a 100 MFD capacitor in parallel with the coil of the relay K5. The capacitor was added to increase the contact closure time of relay K5 to about 3 seconds.

With the closure of the contacts of relay K5, three different functions are performed. First, the reset coil of the stepping relay K4 is energized. Secondly, another longer signal is presented to the same channel of the event recorder which received the initial signal when the bucket tipped. The additional signal to the event recorder passes through a

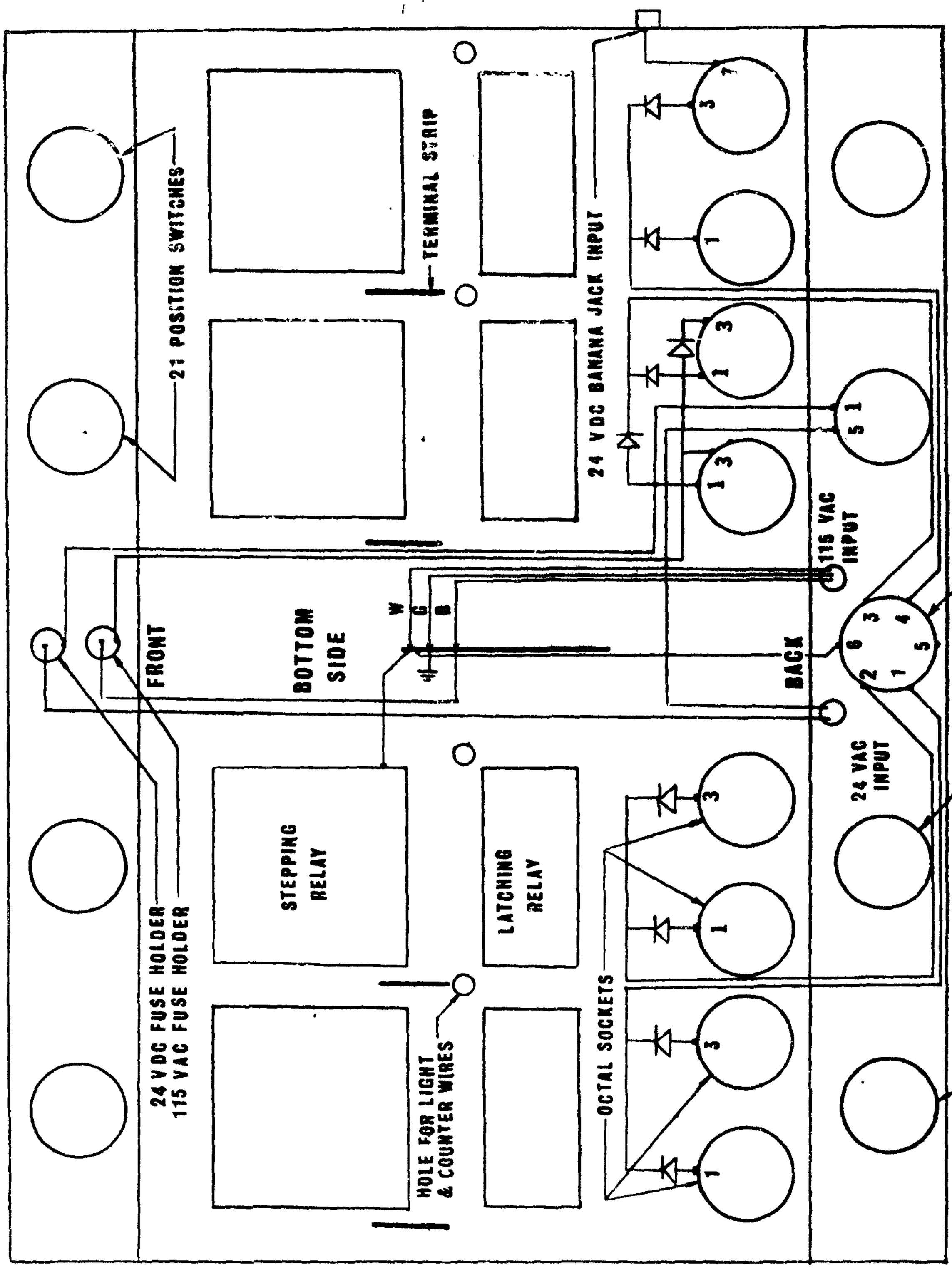
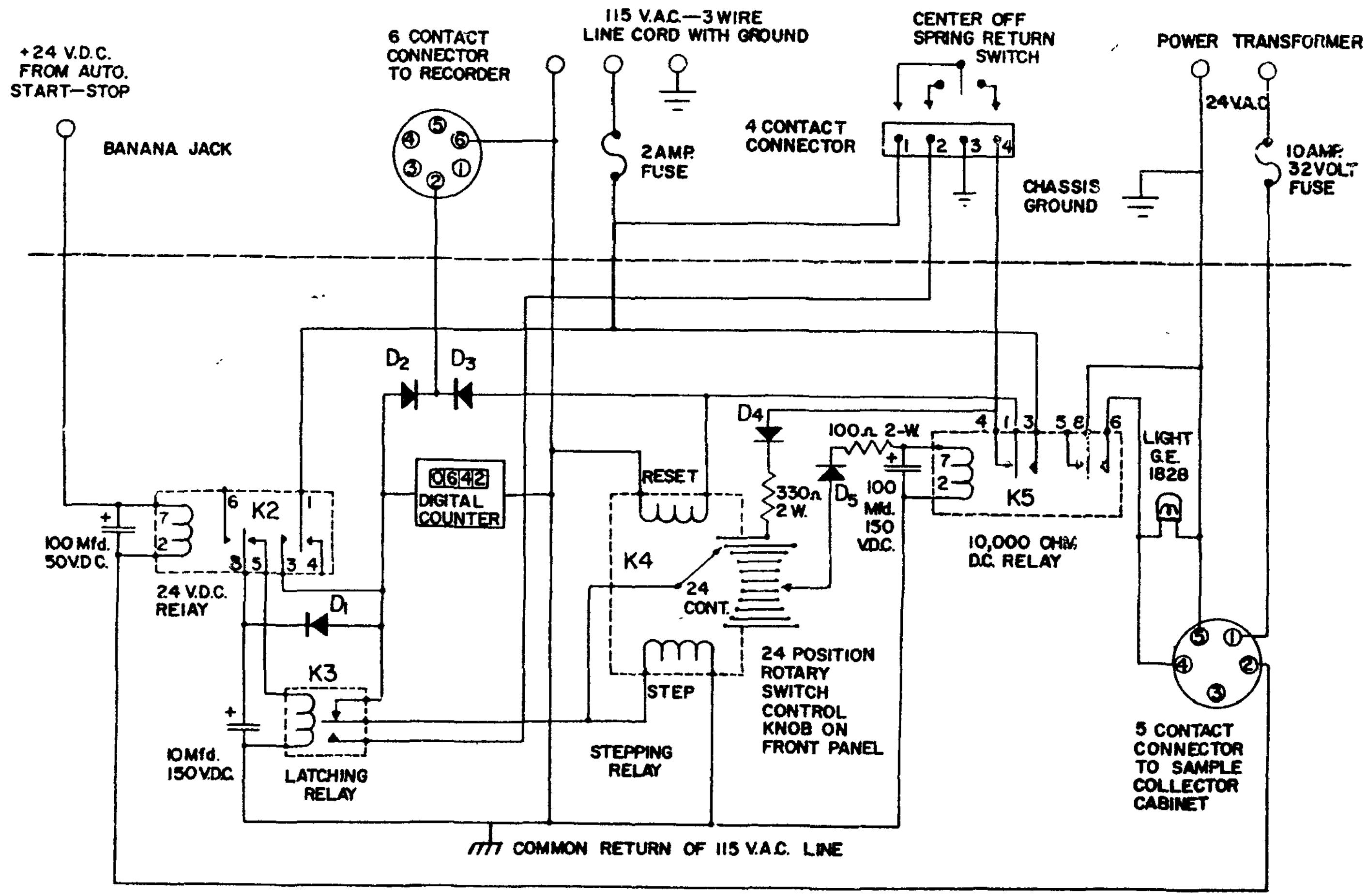


Figure 10. View from below of control chassis.



ALL DIODES ARE TYPE IN2484.
•ALL RELAY CONTACTS ARE SHOWN IN THE NON ENERGIZED STATE.
ALL ITEMS ABOVE DASHED LINE ARE COMMON TO 4 IDENTICAL CIRCUITS BELOW IT.
THIS COMPRISES ONE OF 22 COMPLETE CHASSIS IN THE SYSTEM.

Figure 11. Wiring diagram of data and sample collecting system.

blocking diode and amounts to a dual information system. The addition of the long duration signal to the particular channel of the event recorder results in a distinctive event mark on the chart, indicating that a sample was collected. The third function performed by the closure of one set of the relay K5 contacts is to energize the rotor motor in the sample collector for a three second period of time. During the three second period, the turntable rotates or advances a distance sufficient to release the rod U in Figure 7, permitting the sample bottle positioning microswitch to relax to its normally closed state. Power is supplied to the rotor motor through the part of the circuit containing the positioning microswitch after the contacts of relay K5 are open and until the next sample bottle is properly positioned. Holding the contacts of the relay K5 closed for a three second time interval by means of a capacitor and resistor combination is merely a means of initially over-riding the open positioning microswitch. The light, used as an indicator that 24 VAC power is present at the sample collectors, is shorted by the relay K5 contacts when the rotor motor is energized.

The 115 VAC and 24 VAC electrical power inputs shown in Figure 11 originate from a distribution panel that contains, among other things, a step-down transformer. Except for lighting, all of the electrical circuits in the instrument house basement are powered with either 24 VDC or 24 VAC.

The correlation between the stepping relay contacts and the front panel selector switch for a few of the 21 connections are shown in Figure 12. Also, the connections between a latching relay and stepping relay are shown, along with the dual information circuitry from a relay to an event recorder input.

Figure 13 displays the part of the counting and control circuitry that is specific for a sample collector and tipping bucket. The small numbers used to identify the cables in Figure 13 are the same numbers shown on the five-contact connector in Figure 11. On the terminal strip shown in Figure 13, terminal number one is used as a common ground.

The closed circuit jack (Figure 13) is used as a means of manually controlling power to the turntable motor. By inserting a portable pushbutton switch, the control circuit is disabled and power to the motor is supplied only when the portable switch is closed. With the portable switch, the turntable can be rotated to a convenient position for changing sample bottles.

The slide switch was included in the circuit (Figure 13) as a means of removing the 24 VAC power from the sample collector when a malfunction occurs or during cleaning and repair operations.

The turntable motor shown in Figure 13 is the modified antenna rotary motor previously described. The 10 ohm resistor between points 1 and 4 on the motor terminal board is used as a means of limiting the current

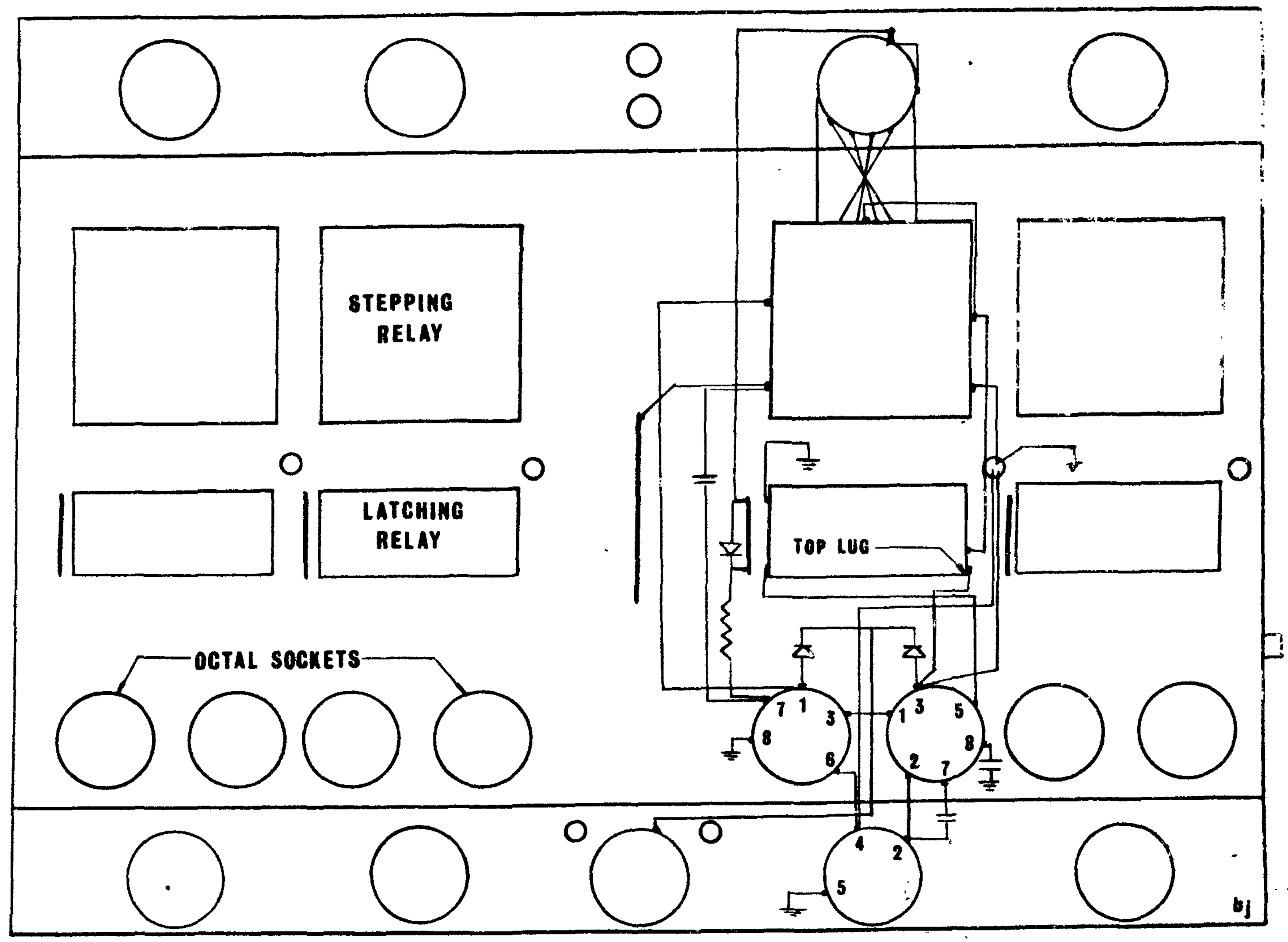


Figure 12. Wiring diagram of a control and counting unit.

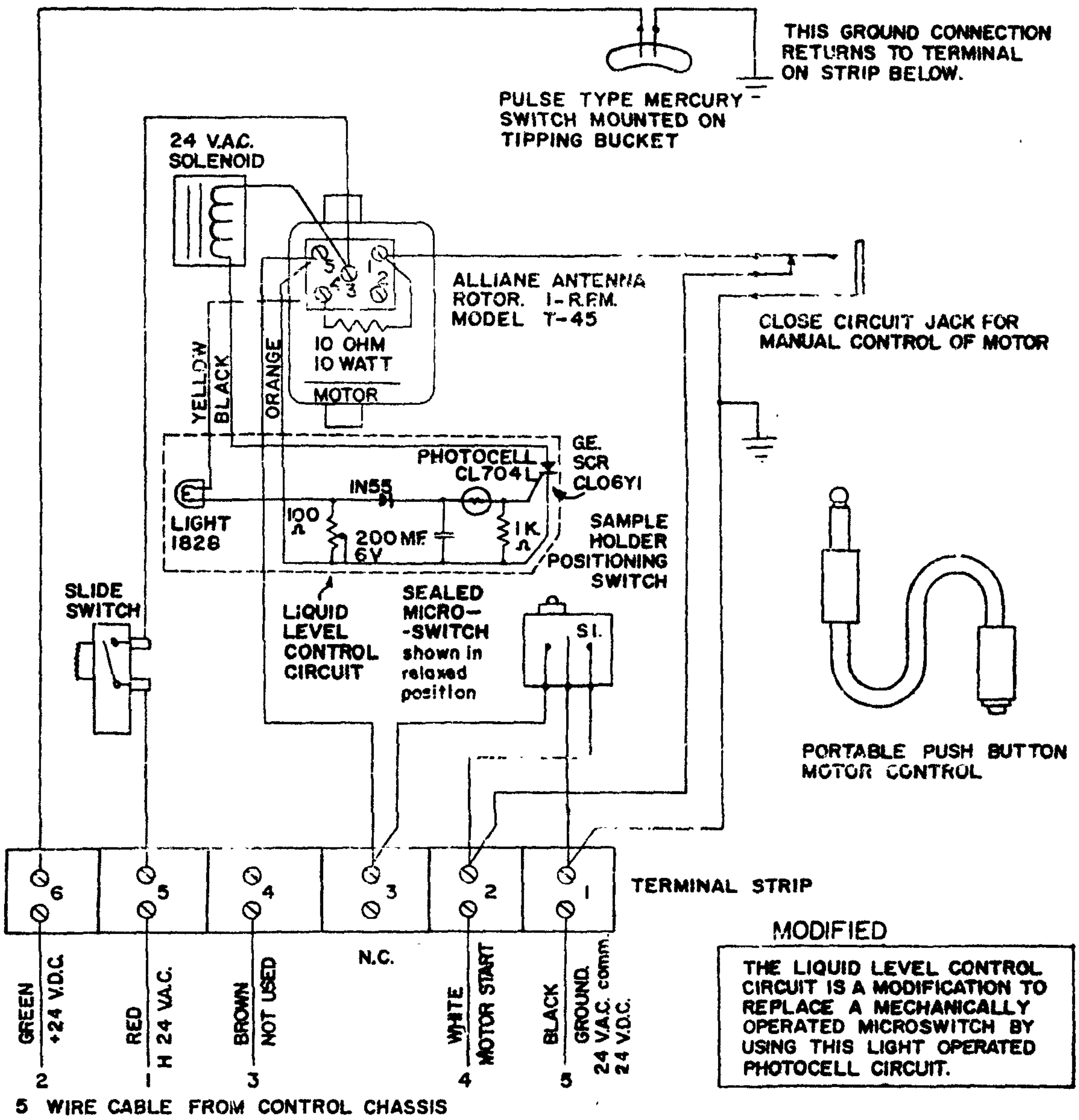


Figure 13. Wiring diagram for sample collector cabinet and tipping bucket circuits.

to prevent the motor from overheating. Also shown in Figure 13, as previously described, is the solenoid that operates the flow control mechanism; the photocell switch circuit which replaced the microswitch that formerly controlled volume in the sample bottle; and the modified microswitch that controls the positioning of the sample bottle.

Five 20-channel Esterline Angus event recorders provide a continuous, accurate record of bucket tips with time. The recorders employ heated styluses to produce records on non-wax, heat-sensitive paper. The heating circuit is rated for operation on 120 VAC. The electromagnet-actuated writing elements are fully deflected by the application or absence of a voltage lasting only 15 milliseconds. Writing elements will record signals up to 20 "on-off" cycles per second, a capability which far exceeds that needed for the tipping buckets.

The chart is 24.7-m long and 15.24-cm wide. Each channel is approximately 0.63-cm wide. Although chart speed can be varied, the most appropriate speed for recording bucket tips under a variety of conditions appears to be about 58 cm/hr. Thus, about 42 hours of continuous operation of an event recorder is possible before the chart must be replaced.

Figure 14 is a block diagram of the circuitry for the automatic turn-on and turn-off system. Electrical power is supplied to the counting and control circuits and recorders by the system only when the tipping buckets are receiving runoff or drainage water. When a bucket collecting runoff or drainage water tips, the signal to the 24 VDC sensing relay results in the activation of the reset timer in its respectively associated circuit, labeled upper or lower drains in Figure 14. Each time any one of the 44 buckets associated with a given circuit tips the timer is reset for the power to remain on after the last tip for some designated period of time that determined by setting the timer to a prescribed time which may be varied from 0 to 3 hours.

To prevent sticking of the recorder styluses to the chart paper, the stylus heater must be turned off 2 minutes before the recorder motor stops. Thus, the early-off circuit within the reset timer is utilized to provide a cooling period for the styluses before electrical power is removed from the control units and recorder motors.

The isolation relays shown in Figure 14 are necessary because one of the five multi-channel recorders is used with both of upper and lower drain circuits. Four channels of the fifth 20-channel recorder E are used to record the tips of buckets receiving runoff water while another four channels of the same recorder are used to record the flow of drainage water.

On the left side of the schematic diagram, presented as Figure 15, 115 VAC is supplied to the circuits and the motor of a recorder which is used as

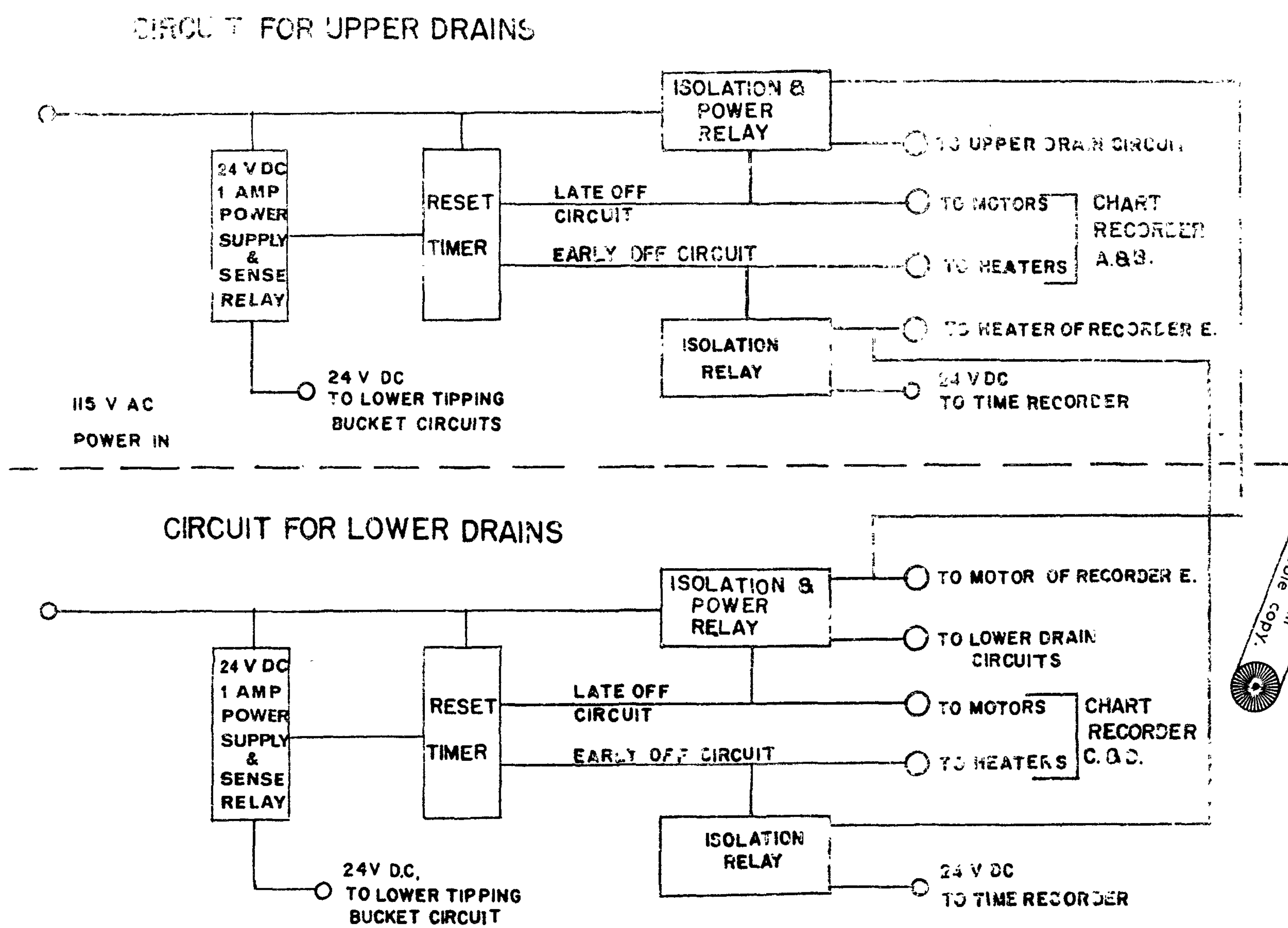
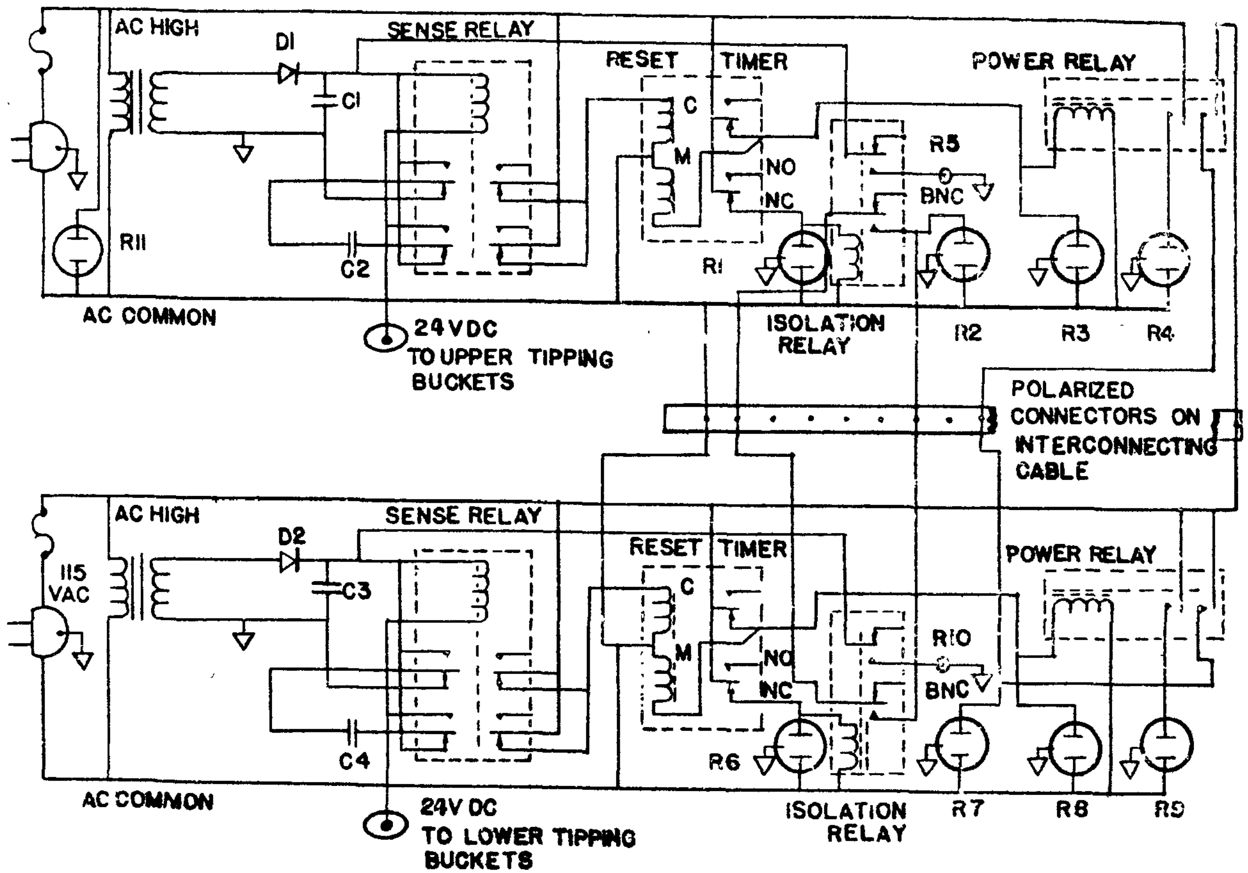


Figure 14. Schematic diagrams for automatic turn-on and turn-off systems for upper and lower drains.



R1 EARLY SHUT-OFF CKT.
R2 RECORDER E. HEATER
R3 RECORDER A AND B. MOTORS
R4 TO RACK-UPPER DRAINS
R5 24 V.DC. SIGNAL TO TIMING CHART RECORDER

R6 EARLY SHUT-OFF CKT.
R7 RECORDER E. MOTOR
R8 RECORDER C. AND D. MOTORS
R9 TO RACK-LOWER DRAINS
R10 TO TIMING RECORDER

Figure 15. Wiring diagram of turn-on and turn-off circuit.

a clock. The recorder used as a clock is a 4-channel Rustrak chart recorder. Channels 1 and 2 of the Rustrak recorder receive 24 VDC power from one set of contacts through receptacles R5 or R10 from its associated isolation relay in each of the upper or lower turn-off and turn-on circuits when the control and counting circuits are energized. Another channel of the Rustrak recorder is used to indicate when power is being supplied to all circuits from an auxiliary gas operated motor and generator power supply.

The step-down transformers shown in Figure 15 are rated at 25.2 volts at one ampere. The voltage is rectified by the diodes and charges the 500 MFD capacitors. The sense relays have an operating voltage of 6 VDC with an internal impedance of 40 ohms. The contacts of the sense relay are shown in the non-energized position. When a bucket tips, the contacts open and allow the reset timer to operate, thus turning on the power to the associated equipment. The power remains on until the reset timer removes it. That is, in the absence of further tipping during a prescribed time by any bucket associated with the circuit, the power is removed.

The upper set of contacts of the reset timer in the upper tipping bucket circuit is connected to the reset timer motor and, through receptacle R3, to the motors of the event recorders designated as B and D. The same kind of circuit arrangement is used to supply power through receptacle R8 to the motors of event recorders A and C which are associated with the lower tipping bucket collecting drainage water. The upper set of contacts of the upper reset timer are also used to supply power to the 115 VAC high side of the power relay. From the power relay, current is supplied through receptacle R4 to the step-down transformer that is the 24 VAC source of power for the upper drain sample collectors. Again in a similar manner, power is supplied to the lower drain sample collectors through receptacle R9, shown in the lower part of the circuit presented in Figure 15.

The lower contacts of the reset timers supply 115 VAC power to the stylus heaters of event records A, B, C, and D through receptacles R1 and R3 in the upper and lower drain circuit elements, respectively (Figure 15). Power is supplied from the isolation relays to the stylus heater of event recorder E through the receptacle R2 and to the motor of the same event recorder E through the receptacle R7.

After the water measuring and sampling equipment had been in operation for several months, it seemed expedient to take water samples at the beginning of a period of runoff. To insure that a sample would be taken from the water causing the second tip of a tipping bucket, the 21 point stepping-relay had to be adjusted to step 20. To accomplish the proper positioning of the contact arm of the stepping-relay and latching relay, a small modification in the control circuit was required. With the modification, the synchronization operation can now be conveniently performed with a special toggle switch. The toggle switch is a single pole double throw with center off and double spring return, made portable by mounting on a small metal box with a one meter length of cable from the switch to an attached four-contact connector. The four-contact connector mates with the four-contact connector installed in each chassis, serving four-control circuits. Thus, the switch mechanism causes the four identical circuits contained in a particular chassis to function simultaneously during the synchronizing process. To modify the circuits for easy synchronization of the tipping buckets for sample collection, it was necessary to install two diodes for the purpose of isolating the circuits so that erroneous bucket tips would not be recorded on the event recorders and a 330 ohm, 2 watt resistor to prevent the step coil of the stepping-relay from being actuated during the process. The location and functions performed by the added circuit elements can best be understood by tracing the power flow through Figure 11.

In Figure 11 it can be seen that, if the portable switch contacts are closed to the right-hand side, current is carried from terminal #1 through the switch to terminal 4 of the 4 contact connector. The current goes to relay K5 where terminals 4 and 1 convey current to the reset coil of the stepping relay K4. The energized reset coil causes the rotating contact arm of relay K4 to come to rest at the starting position or zero contact. Immediately upon closure of the zero contact, current passes through diode D4, the resistor (330 ohm, 2 watt) and the rotating contact arm to the center contact terminal of relay K3 which is an impulse latching relay. If the center terminal is in contact with the lower terminal nothing happens, but if it is in contact with the upper terminal, current then passes through diode D1 to contacts 8 and 5 of relay K2 and eventually energizes the coil of relay K3. Energizing the relay K3 causes the contact to flip to the lower contact terminal and then nothing more happens. All of the reactions described happen in less than a second and the portable switch which initiated the action is released, allowing it to return to its center-off position.

To complete the rephasing or synchronization procedure for any four circuits contained in a particular chassis, it is only necessary to momentarily press the portable control switch in the opposite direction. This is done to impulse the step coil of stepping relay K4. The switch is pressed as many times as necessary to position the rotating contact arm to the desired position. Since we want to collect a sample from the second tip of a bucket at the beginning of the runoff period, the switch must be sequentially pressed 20 times.

To insure against the loss of water samples and data during a storm in which an electric service failure might occur, a 120 volt, 2500 watt auxiliary power plant was installed in the instrument house. This self-starting power plant is automatically activated by a transfer panel if a failure occurs and is automatically turned off when line power is restored. The auxiliary power plant provides power for only the sample and data collection equipment in the instrument house.

To summarize, the special features of the equipment designed to collect water samples and record flow data, it should first be pointed out that, to our knowledge, this is the only lysimeter installation with the capabilities for collecting water samples from known volumes of flow. Automatic sampling equipment, in the past, was designed to collect samples on a time basis, regardless of flow rate. An outstanding feature of the counting and control circuits is the dual information system that provides a distinctive rectangular mark on the proper channel of an event recorder chart when a sample is collected. The counting and control circuits also include electrical digital counters which accumulate the total number of tips for each bucket. The digital counters provide a "back-up" system for collecting total flow data if an event recorder does not function properly. A four-channel event recorder is used as a clock from which the time of day that runoff and/or drainage water was first received and its duration can be determined. Thus, knowing the time of day when flow first occurred, the time when a particular sample was collected can be precisely calculated from the sequential record on the associated channel contained in one of the 20 channel recorders. By means of the turn-on and turn-off circuit, power is supplied to the counting and control circuits only when runoff and/or drainage water is being received by the tipping buckets, thus conserving expensive chart paper. Since runoff and drainage water is not always received at the same time, the control and counting circuits for each group of 44 tipping buckets and associated sample collectors for a particular source of water are powered separately. After some modifications were made to overcome problems caused by the humid basement conditions, the equipment has been very reliable and requires only a nominal amount of time for maintenance.

Disposition of water flow data - As mentioned above, the four-channel Rustrak recorder is used as a real time clock. At the beginning of each month the field technician replaces the Rustrak recorder chart and enters the appropriate date and time on the old chart and on the beginning of the new chart. A particular channel of the Rustrak recorder indicates the on and off operational periods for an event recorder. For example, channel one of the Rustrak recorder is used to identify operating time periods for the 20-channel event recorder labeled C. Each 61 cm length of Rustrak recorder chart represents a 24-hour period. Thus, after each 24-hour segment of the real time chart has been labeled with the proper date, days in which runoff and/or drainage water flows occurred can be readily identified. Each 18.3-m Rustrak recorder chart contains a continuous "on-off" operating record of the 20-channel event recorders for a 30-day period. When operating, a 20-channel event recorder chart travels at a rate of 58 cm per hour, and the 24.7 m long chart will record a runoff or drainage period of about 42-hours duration. Thus, one event recorder chart may contain runoff or drainage water flow data from several storms of short duration or several event recorder charts may be required during a storm lasting several days. Each time the expended recorder charts are changed the ending date and time is recorded on the end of the chart. By matching the dates and time of data recording periods as indicated on the event recorder charts with the "on-off" or operational periods shown on the appropriate channel of the Rustrak recorder, the exact time at which runoff or drainage water flows began and ended are determined. After the correspondence of date and time of runoff and/or drainage water flows have been established between the two types of recorder charts, the date and time are transcribed onto the 20-channel event recorders. Then, each flow period recorded from a particular lysimeter plot is divided into 15-minute time intervals. For each 15-minute time interval, the total number of bucket tips recorded is determined and transferred onto standard FORTRAN coding forms. Finally, the flow data is punched on data cards and the number of bucket tips per 15 minute intervals is converted to volume per unit time by making use of the calibration curves (Figures 4 and 5). Flow rate and total flow are then correlated with total rainfall, rainfall intensity, evaporation rates, total radiation intensities, and the several chemical and biological water quality parameters determined in the laboratories.

As mentioned earlier, each control unit has associated with it an electrical counter for recording total tips of a particular tipping bucket. The electrical counters are, for the most part, a backup for the 20-channel event recorders. However, at the end of a given recording period following a rainstorm the total counts accumulated on the electrical counters are transcribed onto the event recorder chart paper. Total counts are written in the skip space that occurs on the recorder paper as a result of the automatic turnoff of the heat to recorder pens before the recorder is switched off by the delay timers. Thus, during an intensive portion of a storm causing such rapid tipping of the buckets as to result in marks on the recorder chart which are not easily distinguishable as separate events, the total counts are readily available for obtaining an estimate of counts by difference. To date we have not experienced such a difficulty in reading the charts, but this was one of the reasons for installing electrical counters.

Cropping systems and water, sludge and inorganic fertilizer applications - Originally, when the spray irrigation system was used, it was found that infiltration of sludge water was so highly dependent on the initial soil moisture and sludge solids concentrations that a given application rate could not be specified. Thus, the most feasible method of determining treatments was to start each time with the maximum treatment rate for each soil type, which was determined by the amount of sludge that could be applied within a period of about 20 minutes without producing runoff. After this application to the maximum-treated plots, the sludge was applied to the other three treatments in the ratio of one-half maximum, one-quarter maximum, and zero for each soil type. The zero sludge treatments, or check plots, are fertilized annually for high yields and irrigated with well water each time sludge treatments are made. Check plots are irrigated with well water at a rate equivalent to the amount of water supplied with the sludge at the maximum rate. During 1971, the method of irrigation was changed to a ridge and furrow system to facilitate greater loading rates. The crop and treatments are outlined on the plan presented as Figure 16 and are as follows:

The 22 plots located on the north side of the installation are planted to soybeans on or about May 15.

Blount silt loam

3 check plots - annually treated with inorganic fertilizer and irrigated with water.

3 plots irrigated with the maximum amount of sludge permitted by weather conditions and infiltration capacity.

3 plots irrigated with sludge on the same day as above, but with only one-half as much as the maximum rate.

3 plots irrigated with sludge on the same day as above, but with only one-fourth as much as the maximum rate.

12 plots

Plainfield sand

2 check plots - annually treated with inorganic fertilizer and irrigated with water.

1 plot irrigated with the maximum amount of sludge possible.

1 plot irrigated with one-half maximum rate.

1 plot irrigated with one-fourth maximum rate.

5 plots

Simulated Elliott silt loam

Number of plots and treatments are the same as for Plainfield sand.

The 22 plots located on the south side of the installation are planted to corn on or about May 1. Distribution of soil types and treatments are the same as for the north side. All lysimeter plots receive a broadcast application of 269 kg/ha of K before spring plowing.

All check plots planted to corn receive broadcast applications of 336-269-0 kg/ha before spring plowing, except for the sand check plots. The sand check plots receive the same total application, but N is equally divided between three applications. Before plowing, a broadcast application of 112-269-0 kg/ha is made on all sand check plots and about June 20 and again on or about July 15, broadcast applications of 112-0-0 kg/ha are applied.

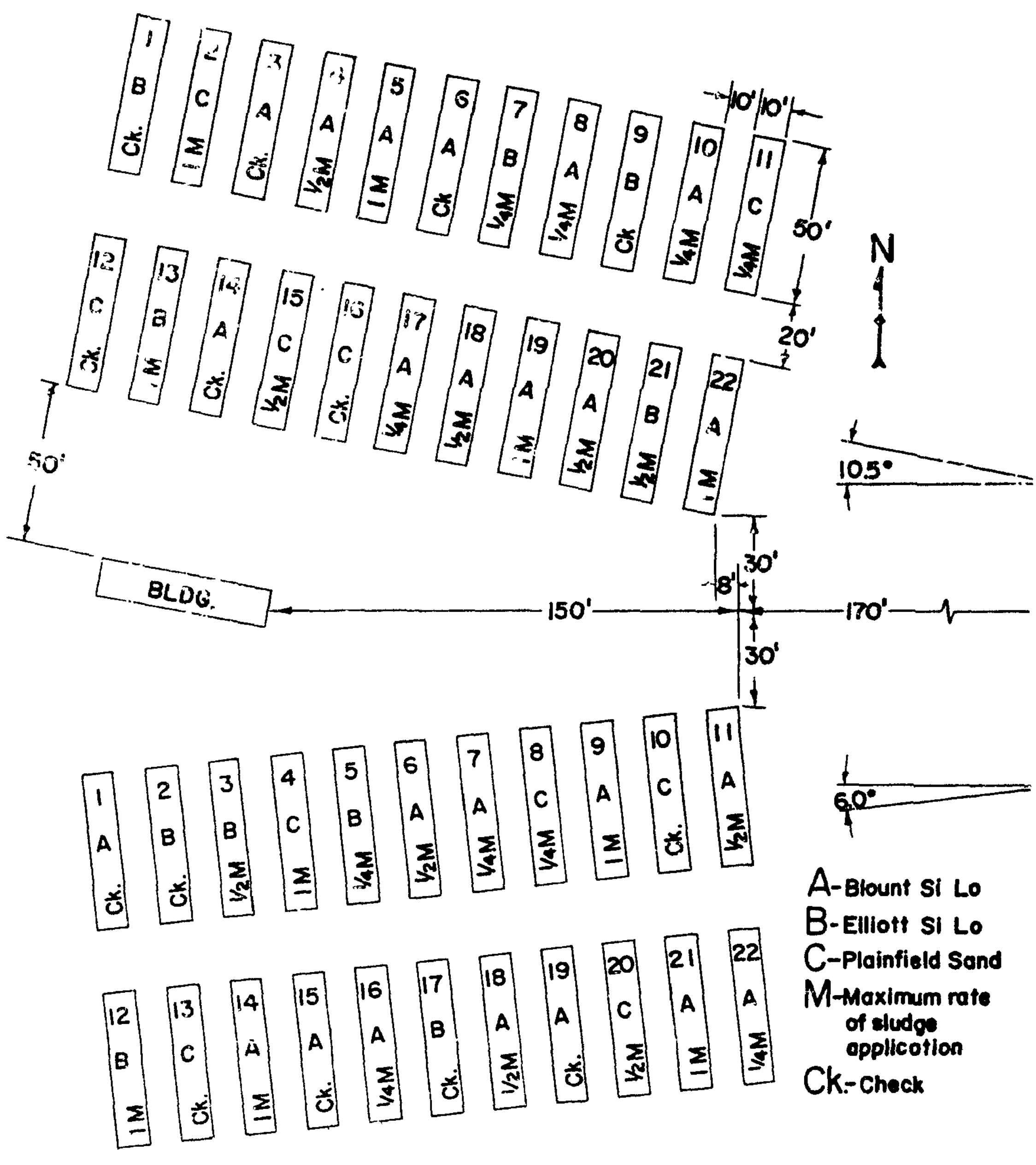


Figure 16. Schematic plan view of lysimeter facility showing soil type and treatment for each lysimeter.

All check plots planted to soybeans receive a broadcast application of 0-269-0 kg/ha before plowing. Also, all plots receive a broadcast application of pre-emergence herbicide at the time or immediately after the planting operation.

<u>Operating procedures and collection of the several kinds of samples</u> - It has been found that sufficient water samples are collected by setting the selector switch, on the front of the control chassis, on position number 21. At this setting, a sample of water will be obtained every forty-second tip of a particular tipping bucket. Depending on flow rate, this is approximately equivalent to taking one water sample for each 0.16 cm of water that runs off the plot surface and 0.11 cm of water lost through the drains.

In the diagrams labeled upper and lower drain systems and presented as Figures 17 and 18, respectively, upper refers to runoff and lower to the drainage water collection and volumetric recording systems. As discussed previously, each chassis contains four individual control units for collecting water samples and recording effluent volumes from runoff or drainage water pipes. Thus, 11 chassis in one rack contain the control units for the runoff water collection and recording equipment provided for each of the 44 lysimeter plots. Another rack houses the 11 chassis serving the drainage water collection and recording equipment. The individual chassis are numbered from 1 through 11 from top to bottom in both of the racks. In a given chassis, each of the four control units are numbered from left to right. Thus, the control unit for sampling and measuring the runoff water from a particular lysimeter plot is identified in the "Upper Drain System" rack by a chassis and position number. The control unit that is sampling and measuring drainage water from a particular lysimeter plot has the same relative position in the rack labeled "Lower Drain System". For example, referring to Figures 17 and 18, there is one control unit in each of the two racks associated with water collection and recording equipment for plot 20 on the north side of the instrument house and coded as plot N-20. Its position or location in each rack is control unit number 3 in chassis number 8. Bucket tips resulting from runoff water from plot N-20 are recorded on channel 11 of the event recorder identified by the letter D, as shown in Figure 17. In a like manner, bucket tips resulting from tile drainage water from plot N-20 are recorded on channel 11 of the event recorder C, as shown in Figure 18.

Water sample bottles are washed and rinsed in a 20 percent nitric acid solution, further rinsed several times with distilled water, and finally with double-distilled water. Clean bottles and screw-on type lids are dryed in a heated cabinet maintained at a temperature of 45 to 50°C. Originally, some of the bottles were autoclaved and used to collect samples of water from which fecal coliform counts were determined. However, after making several comparisons of fecal coliform counts in sterilized versus those in routinely cleaned and dryed bottles, the differences in counts were found to be insignificant.

Chassis Number	Control Unit Position Number in a Chassis 1	2	3	4	Event Recorder Letter Designation	Event Recorder Channel Numbers			
1	N 2	N 11	N 12	N 15	B	1	2	3	4
2	N 16	S 4	S 8	S 10		5	6	7	8
3	S 13	S 20	N 1	N 7		9	10	11	12
4	N 9	N 13	N 21	S 2		13	14	15	16
5	S 3	S 5	S 12	S 17		17	18	19	20
6	N 3	N 4	N 5	N 6	D	1	2	3	4
7	N 8	N 10	N 14	N 17		5	6	7	8
8	N 18	N 19	N 20	N 22		9	10	11	12
9	S 1	S 6	S 7	S 9		13	14	15	16
10	S 11	S 14	S 15	S 16		17	18	19	20
11	S 18	S 1	S 21	S 22	E	1	2	3	4

The above blocks represent the controls on the rack. The letter-number sequence Designates associated plot.

Figure 17. Diagram for determining location of a control circuit and event recorder channel associated with sampling and recording runoff water flows from a lysimeter plot.

Chassis Number	Control Unit Position Number in a Chassis: 1	2	3	4	Event Recorder Letter Designation	Event Recorder Channel Numbers			
1	N 2	N 11	N 12	N 15	A	1	2	3	4
2	N 16	S 4	S 8	S 10	A	5	6	7	8
3	S 13	S 20	N 1	N 7	A	9	10	11	12
4	N 9	N 13	N 21	S 2	A	13	14	15	16
5	S 3	S 5	S 12	S 17	A	17	18	19	20
6	N 3	N 4	N 5	N 8	C	1	2	3	4
7	N 8	N 10	N 14	N 17	C	5	6	7	8
8	N 18	N 19	N 20	N 22	C	9	10	11	12
9	S 1	S 6	S 7	S 9	C	13	14	15	16
10	S 11	S 14	S 15	S 16	C	17	18	19	20
11	S 18	S 19	S 21	S 22	E	1	2	3	4

The above blocks represent the controls on the rack. The letter-number sequence designates associated plot.

Figure 18. Diagram for determining location of a control circuit and event recorder channel associated with sampling and recording drainage water flow from a lysimeter plot.

The fieldmen are cautioned to take care not to contaminate bottles during the operation of removing the lids and placing them in the bakelite cylinders mounted on the water sampling cabinet turntable. All sampling cabinet turntables are enclosed with polyethelene plastic curtains to minimize contamination by dust. Before placing sample bottles in the turntable holders, each bottle is identified by the lysimeter plot number and the number of the bottle in the sequence. For example, UN-5-4 identifies the sample as being the fourth runoff water sample from plot 5 on the north side of the house. For drainage water from the same plot, the letter L will replace the letter U. When the bottle containing a water sample is removed from the sampling equipment a sterilized lid is immediately screwed firmly into place. The data the sample is removed is recorded on the bottle below the identification number. The date is recorded as a four digit military number. For example, April 14, 1969 is written 9104 where the first digit identifies the year the the next three digits are the number of days starting from January 1.

Water samples are removed from the sampling equipment within 12 hours and transferred to the laboratory at Urbana no later than 30 hours after collection. During storage and transit, samples are kept in the original sampling bottles in a cool dark place. Upon arrival at the laboratory, the samples are recorded in the "water samples received" data book.

Soil samples are collected annually during the latter part of April after spring plowing. Samples are collected with a 2.5-cm diameter stainless steel soil probe. Six 76.2 cm deep probes are made per plot. The soil extruded from the probes is divided into 0 to 15, 30 to 46, and 61 to 76 cm depth sections. The six samples, representative of a given soil depth, are composited in a sample box appropriately labeled with the plot number, soil depth and date of collection. The soil samples are transported immediately to the laboratory where they are air-dried and pulverized in a soil grinder, and stored in sealed sample bottles.

When corn plants are 30- to 46-cm tall, 10 plants are cut at 5 cm above the soil surface from each plot in the same random fashion used in collecting soil samples. Immediately after harvest, the plants are dipped in a plastic bucket filled with distilled water. The plants are gently rubbed to loosen soil, especially in leaf axils and whorls, while submerged in the bucket. After the plants have been rinsed in distilled water, they are shaken to remove as much free water as possible before they are placed in a paper bag and labeled with the appropriate plot number and data of sampling.

Later in the growing season when about 10 percent of the corn plants have tasseled, the leaf opposite the ear node from each of 10 plants is collected. The leaf is cut about 7.6 cm away from the stalk to avoid the leaf axil where dirt collects. The leaf samples are washed in the same manner as described above for whole plant samples, folded into a tight bundle, placed in a properly labeled sack, and transported immediately to the laboratory.

Whole plant samples of soybeans are collected when the plants are 15- to 25-cm tall. For this first tissue sample, 20 plants, selected at random, are cut 2.5 cm above the soil surface and composited as a sample from each plot. Later in the season, a second soybean tissue is collected from each plot as the plants reach the full-growth stage. The leaf and petiole from the 3rd and 4th leaf positions from the top of the plant are harvested from 20 plants randomly selected in a plot. All soybean tissue samples are washed and placed in a labeled bag for immediate shipment to the laboratory in the same manner as corn tissue samples are handled.

A randomly selected subsample of about 250 g of alfalfa or grass plant materials is collected from each plot at the time of clipping. Alfalfa and grass samples are cleaned, placed in properly labeled bags and immediately transported to the laboratory in the same way corn and soybean tissue samples are processed. From the corn grain or soybean seed harvest from each plot, a 200 g subsample is randomly collected, placed in a properly labeled bag and transported to the laboratory. Upon receipt at the laboratory, all tissue and grain samples are dried at 60°C to a constant weight in a forced draft oven, ground in a Wiley Mill and stored in acid-cleaned and sealed sample bottles.

From 350 to 400 ml of sludge is collected at the beginning and near the end of an irrigation from a single batch (one storage tank) of sludge. The samples are collected in acid-cleaned glass water sample bottles and handled in the same manner as water samples. The sample bottles are labeled with the date (day batch was used for irrigation), north or south plots, and identified with a B for beginning and an E for ending irrigation sample. Like water samples, sludge samples are transported to the laboratory within 30 hours after collection. Special care is taken to keep the samples cool during transport and they are refrigerated immediately upon their arrival at the laboratory.

Sample analyses - The several kinds and methods of analyses routinely performed on water, plants, sludge, and soil samples received from the lysimeter facility are presented in a summarized form in Table 6. The analytical work is carried out in laboratories on the University of Illinois campus at Urbana-Champaign.

Table 6. Methods of determination for waters, plants, sludge, and soils received at the analytical support laboratory. Sample collection, handling and storage follow guidelines laid out in FWPCA Methods for Chemical Analysis of Water and Wastes, 1971, EPA, AQCL, Cincinnati, or as noted.

Determination or Analyte	Method
	LYSIMETER WATERS
NO_3-N	Ion sensitive electrode or reduction, distillation with titrametric finish
NH_3-N	Gas sensitive electrode or distillation and titrametric finish
P, Total	Acid digestion, vanadomolybdate finish
pH	Conventional glass electrode
Conductivity	Dipping cell
Zn, Cd, Fe, Mn, Cu	Determined in a concentrate obtained by evaporation of suitable volume, hot hydrochloric acid digestion, and chelation and organic-phase extraction. Atomic absorption used.
K*, Na*	Flame emmission photometry
Ca*, Mg*	Atomic absorption with Sr or La release
Fecal coliform	MFC method (Geldreich, E. E., et al., 1965. Fecal-coliform-organism medium for the membrane filter technique. J. Am. Water Works Assoc., 57:208-14)
Hg	FWPCA method
	PLANTS
N	Kjeldahl digestion; ammonia distillation finish
P	Dry-ash sample with vanadomolytdate-yellow method finish

Table 6 (cont). Methods of determination for waters, plants, sludge, and soils received at the analytical support laboratory. Sample collection, handling and storage follow guidelines laid out in <u>FWPCA Methods for Chemical Analysis of Water and Wastes</u>, 1971, EPA, AQCL, Cincinnati, or as noted.

Determination or Analyte	Method
	PLANTS (cont)
Ca, Mg, Zn, Cd, Cu, Fe, Mn, Cr	Atomic absorption on dry-ashed sample (500°C)
K, Na	Flame emmission photometry
B	Spectrophotometric determination of borofluorate-methylene blue complex (Weir and Jones, Trop. Ag., 47(3):261-64.
Hg	Vapor absorption after nitric acid digestion at low temperature
Ni, Pb	Atomic absorption on sample concentrated with chelate and organic phase extraction or absorptiometric
	SLUDGE AND SOILS
Organic C	Dichromate and sulfuric acid digestion with titration finish, the Walkley-Black procedure
P, Total	Acid digestion with vanadomolybdate finish
P, Available	Portion extracted by .025 N HCl and .03 N NH_4F, the Bray P_1 extractant
Ca, Mg, Zn, Cd, Cu, Fe, Mn, Ni, Cr, Pb	Determined in a filtered 0.1 N HCl extract by atomic absorption analysis. Where suitable, elements are concentrated by chelate extracted into an organic phase. The acid extractable portion is used to evaluate plant-available fractions of elements. Total in sludge and soil found in HF-HCl digest after careful ashing at 500°C.

Table 6 (cont). Methods of determination for waters, plants, sludge, and soils received at the analytical support laboratory. Sample collection, handling and storage follow guidelines laid out in FWPCA Methods for Chemical Analysis of Water and Wastes, 1971, EPA, AQCL, Cincinnati, or as noted.

Determination or Analyte	Method
	SLUDGE AND SOILS (cont)
B	Total with HF digest and hot water extractable (plant available), spectrophotometric finish as in plants (above)
Hg, Total	Vapor absorption after nitric acid digestion
K, Na	Flame emmission photometry on 0.1 N HCl extract, total after dry ashing and acid digest
Se*	Acid digestion with diaminobenzidine finish (follows Cummins, et al., Analyt. Chem., 36(2):382-84)
Sn*	Acid digestion with catechol violet finish (follows Newman and Jones, Analyst 91:406-10)
Mo*, Co*	Acid digestion with absorptiometric finish (follows Sandell, 1959, Interscience Pub.)
Cation Exchange Capacity	Ammonium ion saturation, distillation
Moisture Holding Capacity	Water held between 1/3 and 15 atmospheres, pressure membrane apparatus
Aggregate Stability	Weight percent by size, wet sieving using water
Infiltration Capacity	Cylinder infiltrometer

* Each of these elements not determined on every sample received.

Crop and water quality responses to digested sludge applications on the field lysimeter plots (Northeast Agronomy Research Center) - The first sludge applications to the Elwood lysimeter plots started in May 1969, and have been continued to be made as often as possible. The maximum sludge rate is determined by rainfall, the crop to be grown and its tillage requirement, and the dewatering rate of sludge following application onto soils. In seasons of high rainfall and for crops like corn and soybeans where the soil has to be relatively dry for plowing, seeding, and cultivation the amount of sludge that can be applied is severely restricted. Similarly, from the latter part of November until the middle of March, low evaporative losses of sludge water, frozen soil, and snow essentially stop all sludge applications. As of November 1970, a maximum of approximately 25.4 cm of digested sludge has been applied to the lysimeter plots. The dates and amounts of sludge applied to the lysimeters in 1969 and 1970 are listed in Tables 7 and 8. This listing will facilitate interpretation of the data from crop and water analyses presented in this report.

The check plots annually fertilized with inorganic N, P and K fertilizers at rates previously specified provide plant nutrients in quantities more than adequate for maximum crop production. No heavy metal trace elements such as Zn, Cu, Fe, Mn, are added to the check plots in the form of fertilizers. However, as previously mentioned, all check plots are irrigated with well water at the same time and in amounts equivalent to that applied as a constituent of sludge on the maximum treated plots.

The runoff and leaching water samples collected from the lysimeter facility are analyzed for NH_4-N, NO_3-N, PO_4, Cu, and electrical conductivity on a routine basis. During the early period of the investigation before appreciable quantities of sludge could be added to the lysimeter plots, the composition of the drainage waters was relatively unaffected by sludge rates. However, with time and sludge accumulation the influence of sludge rates on nitrates and conductivity became apparent. Toward the end of the growing season in 1970, the accumulation of sludge influenced the results of water analyses more than was apparent earlier. Runoff water was generally higher in P and lower in NO_3-N than corresponding tile drainage water from check plots.

For the analysis of water samples, certain plots were assigned higher priorities than others in terms of analytical sequence for runoff water. Thus, when plots S-14 (Blount soil, maximum treatment) or N-2 (Plainfield sand, maximum treatment) produced seven or more samples, each sample in the series was analyzed in addition to a composite. If fewer than seven samples were produced during a sampling period, composites were made for analyses. In addition to these plots, water from Blount soil plots S-1,6,7,9,11,15,21,22; Plainfield sand plots N-15,11,16; and Elliott soil plots S-12,3,5,17 was analyzed in preference to that obtained from the remainder of the lysimeters. Water samples from these plots were usually composited over the sampling period prior to chemical or biological analysis.

Table 7. Sludge applications on the south (corn) 22 lysimeter plots (Northeast Agronomy Research Center)

Date	Sludge Application	Sludge Accumulation		
		1/4 Max.	1/2 Max.	Max.
	cm	cm	cm	cm
1969				
6/17-7/2	-	0.21	0.42	0.84
8/28	0.55	0.35	0.70	1.39
9/2	0.55	0.48	0.97	1.94
9/8	0.55	0.62	1.24	2.49
9/12	0.55	0.76	1.52	3.04
9/17	0.55	0.90	1.79	3.59
9/19	0.41	1.00	2.00	4.00
9/25	0.34	1.09	2.17	4.34
9/30	0.55	1.22	2.45	4.89
10/23	0.34	1.31	2.62	5.23
10/27	0.41	1.41	2.82	5.65
10/29	0.41	1.51	3.03	6.06
11/7	0.41	1.62	3.24	6.47
11/10	0.27	1.69	3.37	6.74
11/12	0.34	1.77	3.54	7.09
11/28	0.41	1.87	3.75	7.50
12/1	0.41	1.98	3.95	7.91
12/4	0.41	2.08	4.16	8.32
12/9	0.41	2.18	4.37	8.73
12/16	0.41	2.29	4.57	9.14
12/19	0.41	2.39	4.78	9.56
12/22	0.41	2.49	4.98	9.97
1970				
2/5	0.27	2.56	5.12	10.24
3/17	0.27	2.63	5.26	10.52
4/10	0.41	2.73	5.46	10.93
4/24	0.55	2.87	5.74	11.48
6/9	0.55	3.01	6.01	12.02
6/11	0.55	3.14	6.29	12.57
6/19	0.55	3.28	6.56	13.12
7/1	0.55	3.42	6.84	13.67
7/2	0.55	3.56	7.11	14.22
7/6	0.55	3.69	7.38	14.77
7/8	0.55	3.83	7.66	15.32
7/10	0.55	3.97	7.93	15.86
7/13	0.55	4.10	8.21	16.41
7/16	0.55	4.24	8.48	16.96
7/17	0.55	4.38	8.76	17.51
7/21	0.55	4.52	9.03	18.06
7/22	0.55	4.65	9.30	18.61
7/24	0.55	4.79	9.58	19.16

Table 7. (Cont'd) Sludge applications on the south (corn) 22 lysimeter plots (Northeast Agronomy Research Center)

Date	Sludge Application cm	Sludge Accumulation 1/4 Max. cm	1/2 Max. cm	Max. cm
1970				
8/12	0.55	4.93	9.85	19.70
8/14	0.55	5.06	10.13	20.25
8/17	0.55	5.20	10.40	20.80
8/21	0.55	5.34	10.68	21.35
8/26	0.55	5.48	10.95	21.90
8/28	0.55	5.61	11.22	22.45
8/31	0.55	5.75	11.50	23.00
9/2	0.55	5.89	11.77	23.54
9/11	0.55	6.02	12.05	24.09
2/10	0.53	6.16	12.31	24.62

Table 8. Sludge applications on the north (soybeans) 22 lysimeter plots (Northeast Agronomy Research Center)

Date	Sludge Application	Sludge Accumulation		
		1/4 Max.	1/2 Max.	Max.
	cm	cm	cm	cm
1969				
7/4-7/14	-	0.21	0.42	0.84
8/29	0.55	0.35	0.70	1.39
9/3	0.55	0.48	0.97	1.94
9/9	0.55	0.62	1.24	2.49
9/15	0.55	0.76	1.52	3.04
9/18	0.55	0.90	1.79	3.59
9/22	0.55	1.03	2.07	4.14
9/25	0.34	1.12	2.24	4.48
10/23	0.34	1.21	2.41	4.82
10/27	0.41	1.31	2.62	5.23
10/29	0.55	1.45	2.89	5.78
11/10	0.55	1.58	3.16	6.33
11/12	0.41	1.69	3.37	6.74
11/26	0.41	1.79	3.58	7.16
11/28	0.41	1.89	3.76	7.57
12/1	0.41	1.99	3.99	7.98
12/4	0.41	2.10	4.20	8.39
12/9	0.41	2.20	4.40	8.80
12/16	0.41	2.30	4.61	9.21
12/17	0.41	2.40	4.81	9.62
12/22	0.41	2.51	5.02	10.03
1970				
2/5	0.27	2.58	5.16	10.31
3/17	0.27	2.65	5.29	10.58
4/10	0.55	2.78	5.57	11.13
4/24	0.41	2.88	5.77	11.54
6/9	0.41	2.99	5.98	11.96
6/12	0.41	3.09	6.18	12.37
6/19	0.55	3.23	6.46	12.92
7/1	0.55	3.36	6.73	13.46
7/2	0.55	3.50	7.00	14.01
7/6	0.55	3.64	7.28	14.56
7/8	0.55	3.78	7.55	15.11
7/10	0.55	3.91	7.83	15.66
7/13	0.55	4.05	8.10	16.21
7/16	0.55	4.19	8.38	16.76
7/17	0.55	4.32	8.65	17.30
7/20	0.55	4.46	8.92	17.85
7/22	0.55	4.60	9.20	18.40
7/24	0.55	4.74	9.47	18.95

Table 8. (Cont'd) Sludge applications on the north (soybeans) 22 lysimeter plots (Northeast Agronomy Research Center)

Date	Sludge Application	Sludge Accumulation		
		1/4 Max	1/2 Max.	Max.
	cm	cm	cm	cm
1970				
7/27	0.55	4.87	9.75	19.50
7/28	0.55	5.01	10.02	20.05
7/29	0.55	5.15	10.30	20.60
8/3	0.55	5.28	10.57	21.14
8/5	0.55	5.42	10.84	21.69
8/7	0.55	5.56	11.12	22.24
8/10	0.55	5.70	11.39	22.79
8/12	0.55	5.83	11.67	23.34
8/14	0.55	5.97	11.94	23.89
8/17	0.55	6.11	12.22	24.44
8/21	0.55	6.24	12.49	24.98
8/26	0.55	6.38	12.77	25.53
8/28	0.55	6.52	13.04	26.08
8/31	0.55	6.66	13.31	26.63
9/2	0.55	6.93	13.86	27.73

Lower or tile drainage water samples were usually composited over each sampling period because it was assumed by some that the chemical composition of percolated water was more uniform in comparison to runoff water. We now know this is not a valid assumption. Beginning in 1971, after the analytical laboratory was reorganized, samples are not longer composited.

A statistical summary of the chemical analyses of water samples from the lysimeters is given in Table 9. Runoff rate is indicated by the number of bucket tips per 15 minute interval. In some storms this has exceeded 250 tips per 15 minutes. The sampling devices collect a runoff and tile drain water sample for analysis each time any one of the buckets go through a 42 tip cycle. The reader will recall from the description of sampling design that this is equivalent to a sample for each 0.157 cm of runoff water and each 0.117 cm of tile drainage water. Composite and series designations refer to water samples which were composited over the sampling period prior to analysis, and to samples in a series produced during a sampling period which were analyzed individually. Sample number refers to the order in which an individual sample was collected during the collection period.

Statistical correlations were made of ammonium, nitrate and phosphate with each other, with sample number, with sludge accumulation, with electrical conductivity, and with bucket tips per 15 minute interval. The statistical analysis was performed on data from each of the soil types separately instead of all data across soil types. This approach was chosen on the basis of substantial differences in runoff and drainage water distribution between silt loams and sand. Most of the possible correlations were determined but those which are not included did not show significance at .05 percent probability level or above.

Interaction of ions from samples collected in series is somewhat variable. Positive and negative correlations are found for ammonium vs nitrat, nitrate vs phosphorus and nitrate vs phosphate. Since correlations are not consistent for runoff or drainage water, it is questionable whether these correlations have any practical significance.

Correlations of ionic concentrations with sludge accumulation are more consistent. The only negative correlations occur in Blount and Elliott US samples. All other runoff samples show positive correlations. Ionic amounts (ion x bucket tips) either showed no significance or are positively correlated with sludge accumulation.

Ionic concentrations do not correlate consistently with sample position within a sequence of samples. Apparently intensity of precipitation, the sediment load, and rate of water flow varied too much during periods of collection to show trends of increasing or decreasing ionic concentration. The ionic amounts do not correlate any better with sample number.

Table 9. Results of statistical correlations calculated for series and composited water samples from the lysimeter plots (Northeast Agronomy Research Center)

Data Group	Variable	Correlation Coefficients: NH_4^+	NO_3^-	PO_4	Bucket tips/15 min.	Sludge accum. in.	EC	Sample No.
Blount US series	NH_4		-.43		-.49	-.69		.44
	NO_2	-.43				.30*		-.45
	PO_4							.32*
	NH_4 x bucket tips					-.52		.43
	NO_3 x bucket tips					.33		
	PO_4 x bucket tips					.49		
Blount UN series	NH_4		.78	.79		.94	.78	
	NO_3	.78				.68	.88	
	PO_4	.79	.59	.55		.76	.73	
	NH_4 x bucket tips					.64	.45	
	NO_3 x bucket tips					.59	.41	
	PO_4 x bucket tips					.61	.42	
Elliott US series	NH_4		-.94	.74	.67*	.94		
	NO_3	-.94		-.82		-.99		
	PO_4	.74	-.82		[illegible]	[illegible]		.63
	NH_4 x bucket tips							.65*
	NO_3 x bucket tips							
	PO_4 x bucket tips							.70*

Table 9. (Cont'd) Results of statistical correlations calculated for series and composited water samples from the lysimeter plots (Northeast Agronomy Research Center)

Data Group	Variable	Correlation Coefficients NH_4^+	NO_3^-	PO_4	Bucket tips/15 min.	Sludge accum. in.	EC	Sample No.
Elliott UN	NH_4			.92		.83		
series	NO_3			-.56*				-.54*
	PO_4	.92	-.66*			.71*		
	NH_4 x bucket tips					.69*		
	NO_3 x bucket tips							
	PO_4 x bucket tips					.66*		
Elliott LN	NH_4		-.60	.27	-.41*			
and LS	NO_3	-.60			.50	.84		
	PO_4	.87						
	NH_4 x bucket tips	.76	-.52	.54				
	NO_3 x bucket tips	-.47	.80			.71		
	PO_4 x bucket tips		.51			.61		
Plainfield	NH_4		.50		-.38*			
sand UN	NO_3	.50			-.51			
	PO_4					.69		
	NH_4 x bucket tips							
	NO_3 x bucket tips					.54		
	PO_4 x bucket tips					.61		.42*

Table 9. (Cont'd) Results of statistical correlations calculated for series and composited water samples from the lysimeter plots (Northeast Agronomy Research Center)

Data Group	Variable	Correlation Coefficients						
		NH_4^+	NO_3^-	PO_4	Bucket tips/15 min.	Sludge accum. in.	EC	Sample No.
Plainfield sand LN and LS	NH_4			.57*				
	NO_3			-.92	-.90	.93		
	PO_4	.57*	-.82		.93	.87		
	NH_4 x bucket tips							
	NH_4 x bucket tips							
	PO_4 x bucket tips					.92		
Blount US composites	NH_4		.44	.82				
	NO_3	.44				.46	.61	
	PO_4	.82				.28	.29*	
	Sludge accum.		.49	.36			.37	
Blount US composites without snow melt	NH_4		.52	.20*				
	NO_3	.52		.29		.47		
	PO_4	.20*	.29			.43		
Blount UN composites	NH_4			.42				
	NO_3					.49		
	PO_4	.42				.45		
	Sludge accum.						.36	

Table 9 (Cont'd) Results of statistical correlations calculated for series and composited water samples from the lysimeter plots (Northeast Agronomy Research Center)

Data Group	Variable	Correlation Coefficients						
		NH_4^+	NO_3^-	PO_4	Bucket tips/15 min.	Sludge accum. in.	EC	Sample No.
Blount UN composites without snow melt	NH_4							
	NO_3					.49		
	PO_4					.50		
Blount LS composites	NH_4					.24*		
	NO_3					.78	.79	
	PO_4					.32		
	Sludge accum.	.24*	.78	.32			.66	
Blount LN composites	NH_4					.33		
	NO_3					.94		
	PO_4							
	Sludge accum.	.33	.94				.90	
Elliott US composites	NH_4		.79	.97			.82	
	NO_3	.79		.72			.78	
	PO_4	.97	.72				.84	
	Sludge accum.							

Table 9. (Cont'd) Results of statistical correlations calculated for series and composited water samples from the lysimeter plots (Northeast Agronomy Research Center)

Data Group	Variable	Correlation Coefficients						
		NH_4^+	NO_3^-	PO_4	Bucket tips/15 min.	Sludge accum. in.	EC	Sample No.
Elliott US composites without snow melt	NH_4		.34*	.40*				
	NO_3	.34*		.55				
	PO_4	.40*	.55					
Elliott UN composites	NH_4			.99		.83		
	NO_3						.69	
	PO_4	.99				.85		
	Sludge accum.	.83		.85				
Elliott UN composites without snow melt	NH_4			.99		.86*		
	NO_3							
	PO_4	.99				.83*		
Elliott LS composites	NH_4			.80				
	NO_3					.82	.88	
	PO_4	.80						
	Sludge accum.		.32				.80	

Table 9. (Cont'd) Results of statistical correlations calculated for series and composited water samples from the lysimeter plots (Northeast Agronomy Research Center)

Data Group	Variable	Correlation Coefficients						
		NH_4^+	NO_3^-	PO_4	Bucket tips/15 min.	Sludge accum. in.	EC	Sample No.
Elliott LN composites	NH_4					.53*	.70	
	NO_3					.85		
	PO_4					.60*		
	Sludge accum.	.53*	.85	.60*				
Plainfield sand US composites	NH_4		.69					
	NO_3	.69				.74		
	PO_4					.94		
	Sludge accum.	.94	.74				.73	
Plainfield sand UN composites	NH_4						.27*	
	NO_3					.32	.81	
	PO_4					.70		
	Sludge accum.			.76				
Plainfield sand LS composites	NH_4							
	NO_3						.98	
	PO_4							
	Sludge accum.							

Table 9. (Cont'd) Results of statistical correlations calculated for series and composited water samples from the lysimeter plots (Northeast Agronomy Research Center)

		Correlation Coefficients						
Data Group	Variable	NH_4^+	NO_3^-	PO_4	Bucket tips/15 min.	Sludge accum. in.	EC	Sample No.
Plainfield sand LN composites	NH_4							
	NO_3					.89	.56	
	Sludge accum.		.89					

*Coefficient shows significance at P = .05. Other values show significance at P = .01 or above.

Significant correlations of ionic concentration and rate of water loss (bucket tips/15 min.) are negative for ammonium in Blount US and Plainfield UN, and positive in Elliott US. Elliott US shows the only significant correlation between phosphate and water loss rate.

Ionic interactions from composited samples when significantly correlated are positive. Thus, all ions tend to increase or decrease in concentration together. Significant correlations between ions and sludge accumulation are also positive as would be expected. Nitrate and sludge accumulation correlate positively with conductivity.

Nitrate and phosphate concentrations in lower drain water are unaffected by discharge intensity, but are correlated with sludge accumulation.

The distribution of water collected as runoff and drainage from the lysimeters is shown in Table 10. The silt loam soils generally produced more water as runoff than drainage in 1969. In 1970, this trend was generally reversed in the north (soybean) plots, but not in the south (corn) plots. The latter had a greater proportion of water lost as lower drainage in 1970 than in 1969, however. Sand plots showed anomalous losses of water. Several lost more water than should be possible when allowance for evapotranspiration is made.

Excess water lost from sand plots can be explained by the construction of the plots. A 3 x 15 m excavation was made for the sand, but an additional 0.75 m of soil on each side was removed to a depth of about 0.3 m. This allowed sand to continue into the border area. Since the fiberglass plot barriers are only 15 cm deep, a gap between them and the sub-surface plastic liner exists. Thus, the plots have 1/3 more surface area effective in accumulation precipitation than the area of the plot on which calculations are based.

Soybeans were grown on the north set of 22 lysimeter plots and corn on the south 22 lysimeter plots established on the Northeast Agronomy Research Center. The soybean and corn yields for 1969 and 1970 are presented in Table 11. In 1969, average corn yields were somewhat greater on highly fertilized Elliott silt loam and Plainfield sand plots irrigated with water as compared to sludge-treated plots, but yields of soybeans were not affected by treatment. Again in 1970, average corn yields were greater on control plots, but yields were not affected by sludge application rates. Average soybean yields were also greatest on control plots, but unlike corn yields, were decreased with increasingly greater rates of sludge applications. Considering the frequency at which the plots were spray irrigated, it is not surprising that yields were decreased. While the leaves of sludge irrigated soybeans appeared to be healthy, they were considerably smaller with regard to surface area than those on plants irrigated with equivalent amounts of well water. Thus, it appears that yields were reduced by lower photosynthetic rates

Table 10. Distribution of runoff and drainage water from the lysimeter plots (Northeast Agronomy Research Center) in 1969 and 1970. Measurements of runoff and drainage water were initiated in June 1969

Year	Plot No.	Total Water & Rainfall		Recovery Total Applied	Distribution	
					Runoff	Drainage
		cm	cm	%		
			Blount Check			
1969	N-3	49.83	18.42	36.9	36.5	0.4
1970		72.47	29.29	40.4	34.6	5.8
1969	N-6	49.83	5.33	10.7	9.8	0.9
1970		72.47	20.07	27.7	10.1	17.6
1969	N-14	49.83	10.95	22.0	21.8	0.2
1970		72.47	22.96	31.6	26.8	4.8
			Blount 1/4 Maximum			
1969	N-8	43.15	10.34	24.0	22.0	2.0
1970		66.85	27.41	41.0	10.1	30.9
1969	N-10	43.15	11.81	25.4	17.6	7.8
1970		66.85	43.76	65.5	6.3	59.2
1969	N-17	43.15	14.83	34.3	33.8	0.5
1970		66.85	17.02	25.4	17.3	8.1
			Blount 1/2 Maximum			
1969	N-4	45.67	5.03	11.0	5.7	5.3
1970		72.49	17.93	24.7	8.4	16.3
1969	N-18	45.67	5.99	13.1	12.3	0.8
1970		72.49	19.15	26.4	10.3	16.1
1969	N-20	45.67	13.11	28.7	26.7	2.0
1970		72.49	27.64	38.1	14.5	23.6
			Blount Maximum			
1969	N-5	50.67	4.04	15.0	8.0	7.0
1970		83.74	32.16	38.4	4.5	33.9
1969	N-19	50.67	7.42	14.7	14.5	0.2
1970		83.74	19.15	22.9	9.3	13.6
1969	N-22	50.67	10.64	21.0	18.4	2.6
1970		83.74	96.27	44.6	15.0	29.6

Table 10. (Cont'd) Distribution of runoff and drainage water from the lysimeter plots (Northeast Agronomy Research Center) in 1969 and 1970. Measurements of runoff and drainage water were initiatated in June 1969

Year	Plot No.	Total Water & Rainfall		Recovery Total Applied	Distribution	
					Runoff	Drainage
		cm	cm	%	%	%
			Blount Check			
1969	S-1	49.35	9.40	19.0	14.7	4.3
1970		72.75	27.76	38.1	19.4	18.7
1969	S-15	49.35	19.33	39.2	38.8	0.4
1970		72.75	20.62	28.3	24.6	3.7
1969	S-19	49.35	6.91	14.0	13.0	1.0
1970		72.75	16.92	23.2	6.6	16.6
			Blount 1/4 Maximum			
1969	S-7	43.13	17.88	41.4	40.7	0.7
1970		63.96	25.58	39.0	31.3	7.7
1969	S-16	43.13	15.93	36.9	35.7	1.2
1970		63.96	17.58	26.8	19.6	7.2
1969	S-22	43.13	8.53	19.8	19.6	0.2
1970		63.96	21.64	33.0	4.8	28.2
			Blount 1/2 Maximum			
1969	S-6	45.62	9.98	21.8	21.6	0.2
1970		69.09	7.52	10.9	5.9	5.0
1969	S-11	45.62	10.54	23.1	19.9	3.2
1970		69.09	21.06	30.4	12.8	17.6
1969	S-18	45.62	15.60	34.2	33.4	0.8
1970		69.09	21.59	31.2	18.5	12.7
			Blount Maximum			
1969	S-9	50.62	16.43	32.5	32.1	0.4
1970		75.34	16.87	22.3	13.8	8.5
1969	S-14	50.62	18.92	37.4	35.6	1.8
1970		75.34	20.42	27.1	14.1	13.0
1969	S-21	50.62	17.30	34.2	32.5	1.7
1970		75.34	24.03	31.9	16.3	15.6

Table 10. (Cont'd) Distribution of runoff and drainage water from the lysimeter plots (Northeast Agronomy Research Center) in 1969 and 1970. Measurements of runoff and drainage water were initiated in June 1969

Year	Plot No.	Total Rainfall	Water &	Recovery Total Applied	Distribution Runoff	Drainage
		cm	cm	%		
			Blount Check			
1969	N-1	49.83	8.66	17.4	14.1	3.3
1970		72.47	34.52	47.7	18.5	29.2
1969	N-9	49.83	13.00	26.1	23.0	3.1
1970		72.47	28.35	39.1	19.3	19.8
			Elliott 1/4 Maximum			
1969	N-7	43.15	27.69	64.1	60.3	3.8
1970		66.85	13.72	20.6	2.4	18.2
			Elliott 1/2 Maximum			
1969	N-21	45.67	3.33	7.3	6.3	1.0
1970		72.49	32.26	44.5	17.6	26.9
			Elliott Maximum			
1969	N-13	50.67	4.19	8.2	7.4	0.8
1970		83.74	10.08	12.0	5.7	6.3
			Elliott Check			
1969	S-2	49.35	12.93	26.2	24.3	1.9
1970		72.75	16.43	22.6	18.2	4.4
1969	S-17	49.35	15.88	32.2	30.9	1.3
1970		72.75	20.87	28.7	26.4	2.3
			Elliott 1/4 Maximum			
1969	S-5	43.13	5.72	13.2	12.0	1.2
1970		63.96	12.45	19.0	14.3	4.7
			Elliott 1/2 Maximum			
1969	S-3	45.62	12.83	28.1	26.7	1.4
1970		69.09	11.33	16.4	7.4	9.0
			Elliott Maximum			
1969	S-12	50.62	17.15	33.9	27.1	6.8
1970		75.34	54.00	71.7	40.2	31.5

Table 10. (Cont'd) Distribution of runoff and drainage water from the lysimeter plots (Northeast Agronomy Research Center) in 1969 and 1970. Measurements of runoff and drainage water were initiated in June 1969

Year	Plot No.	Total	Water & Rainfall	Recovery Total Applied	Distribution Runoff	Drainage
		cm	cm		%	
			Plainfield Check			
1969	N-12	49.83	9.02	18.1	12.2	5.9
1970		78.66	70.54	89.6	16.7	72.9
1969	N-16	49.83	11.56	23.2	17.3	5.9
1970		78.66	50.77	64.1	18.4	45.7
			Plainfield 1/4 Maximum			
1969	N-11	49.83	37.69	87.4	18.1	69.3
1970		78.66	143.08	215.7	10.7	205.0
			Plainfield 1/2 Maximum			
1969	N-15	45.67	7.11	15.6	12.1	3.5
1970		71.42	38.20	53.5	10.3	43.2
			Plainfield Maximum			
1969	N-2	50.67	12.67	24.7	10.6	14.1
1970		81.64	74.57	91.3	14.4	76.9
			Plainfield Check			
1961	S-10	49.35	22.12	44.9	10.9	34.0
1970		76.53	95.48	124.7	11.7	113.0
1969	S-13	49.35	7.59	15.7	8.2	7.2
1970		76.53	44.40	58.1	9.0	49.1
			Plainfield 1/4 Maximum			
1969	S-8	43.13	9.65	22.4	15.4	7.0
1970		64.74	40.94	63.2	10.6	52.6
			Plainfield 1/2 Maximum			
1969	S-20	45.62	4.55	9.9	4.1	5.8
1970		68.28	41.78	61.2	14.7	46.5
			Plainfield Maximum			
1969	S-4	50.62	3.84	7.6	7.5	0.1
1970		75.34	13.74	18.2	4.9	13.3

Table 11. Average corn and soybean yields as tons per hectare on NEARC lysimeter plots by sludge treatment, soil types, and year.

Soil Type	Year	Control	Sludge Treatment Rate 1/4 M	1/2 M	Maximum
		Corn			
Blount silt loam	1969	5.83	5.82	3.86	5.23
Blount silt loam	1970	5.25	2.40	1.48	2.01
Elliott silt loam	1969	7.24	4.85	5.48	5.55
Elliott silt loam	1970	7.58	1.87	1.93	2.89
Plainfield sand	1969	1.41	0.18	0.05	0.43
Plainfield sand	1970	3.52	0.04	0.25	0.10
		Soybeans			
Blount silt loam	1969	1.55	2.06	1.89	1.65
Blount silt loam	1970	1.59	0.70	0.40	0.18
Elliott silt loam	1969	1.75	2.51	1.43	1.42
Elliott silt loam	1970	2.27	0.60	0.52	0.10
Plainfield sand	1969	0.25	0.15	0.13	0.26
Plainfield sand	1970	0.88	0.37	0.04	0.02

resulting from the reduction in light absorption. The reduction in light absorption by coating leaves with sludge seemed to be greater for soybeans than for corn. Soybeans, having pubescent leaves, are prone to accumulate more sludge solids on leaf surfaces than corn or at least the solids are not shed as rapidly after drying. Although it was noted that sludge spraying was causing an adverse light absorption effect, frequent applications were continued in the interest of obtaining high sludge loading rates on the soils. Following the 1970 growing season, the method of irrigating the lysimeter plots with sludge and water was changed to a ridge and furrow system. Soybeans and corn grown with ridge and furrow irrigation should provide a more accurate assessment of elemental uptake through the roots.

Chemical composition of plants - Plant chemical compositions were influenced by sludge application as shown in Tables 12, 13 and 14. Zinc, Fe, Al, and Cu increased in concentration in corn leaves. Zinc, Fe and perhaps Al increased in concentration in corn grain. Sludge addition caused no apparent depression in any element's concentration.

With increasingly greater sludge application rates, Zn, Fe, Ca, Al, and Cu increased in soybean grain, and Zn, Mn, Mg, P, Na, and Al increased in soybean leaves. No elements were depressed with sludge application. All plant analyses indicate a high level of plant nutrients in the soil; none indicates that crops might be detrimental to human or animal life.

South Farm Lysimeter Studies

Introduction - When the project was begun in April 1967 it was soon recognized that it was improbable that the large, sophisticated, lysimeter facility to be constructed on the Northeast Agronomy Research Center would be operational during the first years of the project. To obtain some preliminary information while the larger lysimeter facility was being constructed, the decision was made to use some small lysimeters for digested sludge utilization studies which had been established on the University of Illinois Agronomy South Farm, at Urbana, Illinois. The small lysimeters, on the Agronomy South Farm, were originally established to measure amounts of runoff and percolation water resulting from natural precipitation and to determine leaching losses of soil constituents. In 1937 triplicate profiles of each of eight soil types were collected in galvanized steel cylinders which are 0.91 m in diameter and 1 m long in as nearly an undisturbed condition as possible to be used in the small lysimeter facility. The encased soil profiles were set upright into the soil around the periphery of a basement like structure so that their surface was at the same elevation as the surrounding soil. Before the profiles were lowered into position, gravel and solid tubing were installed at the bottom of each cylinder to convey the water percolating through the soil profiles to outlets in the basement structure. At the surface of each profile a metal ring, fitted with a solid piece of tubing, was installed in such a manner as to insure the collection of runoff

Table 12. Composition of corn leaves from the lysimeter plots in 1970 (Northeast Agronomy Research Center)

Soil type and sludge treatment	Zn ppm	B ppm	Fe ppm	Mn ppm	Mg %	Ca %	P %	K %	Na ppm	Al ppm	Si ppm	Cu ppm
						Corn Leaves						
Blount												
Maximum	111	28.9	309	34.3	0.28	0.43	0.23	2.27	>1400	115	1.28	28.7
1/2 maximum	111	34.6	347	38.2	0.30	0.47	0.23	2.23	>1400	156	1.64	30.1
1/4 maximum	123	31.7	354	43.3	0.35	0.52	0.24	1.39	>1360	165	1.60	31.4
Check	32.[illegible]	39.2	131	58.5	0.35	0.53	0.24	1.05	>1400	55.3	1.37	2[illegible].8
Elliott												
Maximum	164	28.2	368	43.0	0.32	0.56	0.28	2.08	>1400	168	1.83	22.2
1/2 maximum	122	28.2	416	44.0	0.26	0.45	0.26	2.44	>1400	52.0	1.45	30.2
1/4 maximum	169	41.2	421	34.0	0.29	0.51	0.25	1.98	>1262	256	1.51	38.3
Check	36.2	30.2	132	59.0	0.31	0.54	0.24	1.64	>1400	43.8	1.22	22.2
Plainfield												
Maximum	169	42.2	490	43.0	0.19	0.44	0.29	2.25	>1400	244	1.59	39.2
1/2 maximum	131	50.8	388	30.0	0.22	0.45	0.23	2.21	>1400	215	1.73	32.7
1/4 maximum	109	38.2	340	31.0	0.20	0.39	0.24	2.41	>1400	204	1.70	32.3
Check	30.4	70.5	295	71.5	0.26	0.51	0.30	2.30	>1400	54.5	1.20	17.2

Table 13. Composition of corn grain from the lysimeter plots in 1970 (Northeast Agronomy Research Center)

Soil type and sludge treatment	Zn ppm	B ppm	Fe ppm	Mn ppm	Mg %	Ca %	P %	K %	Na ppm	Al ppm	S %	Cu ppm
					South Corn Grain							
Blount												
Maximum	99.6	4.7	83.5	14.7	0.26	<0.05	0.40	0.84	280	<10.0	<0.20	12.7
1/2 maximum	93.8	6.0	83.5	14.8	0.25	<0.05	0.45	0.74	196	<10.0	<0.20	10.3
1/4 maximum	73.6	4.5	68.3	<5.7	0.22	<0.06	0.41	0.53	<64.0	<10.0	<0.20	8.5
Check	43.5	4.4	62.7	11.7	0.26	<0.05	0.47	0.72	147	<10.0	<0.20	8.0
Elliott												
Maximum	129	5.0	82.0	9.0	0.28	<0.05	0.49	0.67	71.0	<10.0	<0.20	8.2
1/2 maximum	98.2	4.0	93.0	9.0	0.26	<0.05	0.47	0.73	80.0	<10.0	<0.20	22.5
1/4 maximum	102	5.5	107	18.0	0.28	<0.05	0.46	0.80	80.0	<10.0	<0.20	10.0
Check	49.0	5.5	67.0	14.0	0.26	<0.05	0.45	0.70	195	<10.0	<0.20	11.2
Plainfield												
Maximum	111	5.0	83.0	12.0	0.28	<0.05	0.50	0.74	161	<10.0	<0.20	8.5
1/2 maximum	130	6.0	89.5	16.0	0.30	<0.05	0.60	1.00	137	<10.0	<0.20	11.2
1/4 maximum	98.2	5.0	78.0	20.0	0.32	<0.05	0.61	1.07	323	<10.0	<0.20	14.0
Check	42.6	5.5	77.5	12.5	0.27	<0.05	0.51	0.78	125	<10.0	<0.20	6.9

Table 14. Composition of soybean grain from the lysimeter plots in 1970 (Northeast Agronomy Research Center)

Soil type and sludge treatment	Zn ppm	B ppm	Fe ppm	Mn ppm	Mg %	Ca %	P %	K %	Na ppm	Al ppm	Si %	Cu ppm
						North Soybeans						
Blount												
Maximum	204	72.9	418	94.0	0.45	0.72	0.92	3.96	1000	240	<0.76	35.6
1/2 maximum	197	73.8	232	91.0	0.47	0.70	0.89	3.97	981	62.2	<0.20	36.5
1/5 maximum	139	60.7	209	72.0	0.40	0.69	0.85	4.02	817	58.3	<0.20	32.6
Check	116	70.9	207	92.7	0.42	0.58	0.81	4.14	949	28.0	<0.20	26.8
Elliott												
Maximum	208	81.0	268	92.0	0.45	1.26	1.02	4.73	1184	52.0	<0.20	35.0
1/2 maximum	246	87.8	238	103	0.52	0.76	0.89	3.60	980	67.5	<0.20	39.2
1/4 maximum	190	73.2	224	88.5	0.53	0.73	0.95	3.67	926	47.0	<0.20	37.7
Check	142	76.0	197	80.5	0.50	0.59	0.92	4.11	1148	34.5	<0.20	26.4
Plainfield Sand												
Maximum	254	95.2	353	88.5	0.47	0.73	>1.15	4.81	1388	138	0.26	41.3
1/2 maximum	336	107	496	113	0.53	0.67	>1.15	4.22	>1400	213	0.62	49.8
1/4 maximum	170	79.8	270	75.0	0.48	0.70	0.88	4.40	860	81.5	<0.20	35.8
Check	81	91.5	234	98.5	0.47	0.63	1.06	4.87	1115	36.0	<0.20	20.9

water and its conveyance to an outlet in the basement structure. Thus, both runoff and drainage water could be collected, but the small lysimeters were never equipped to provide for measurement of flow rates or the collection of water samples representative of specific intervals of total flow events.

The eight soil series represented in the South Farm lysimeters are Brooklyn, Cisne, Cowden, Elliott, Soybrook, Herrick, Muscative, and Tama. These soils have been described in detail by Stauffer and Smith (146). Briefly, they may be described by saying they all are silt loams, developed under grass vegetation and, except for the Brooklyn series, occupy extensive areas in Illinois. Under natural conditions all occupy areas having slopes that vary from 0.5 to 3.5 percent, except the Tama series which occurs on 3.5 to 7.0 percent slopes. The main differences between the various soil profiles are their differences in permeability and internal drainage capacities.

Sludge, obtained from the Calumet Sewage Treatment Plant of the Metropolitan Sanitary District of Greater Chicago, was applied as received from the digesters, i.e., as a liquid with about 3 percent solids. Sludge treatments were chosen on an estimation of the maximum liquid volume which could be accommodated. In 1967, rates of 2.54 cm and 1.27 cm inch per eight days were chosen, and sludge application totals of 25.4 cm and 12.7 cm were realized. In 1968, 1969 and 1970 rates of 2.54 cm and 1.27 cm per week were adopted, and the maximum yearly totals were 25.4 cm and 18 m, respectively. Commercial fertilizer, at the rate of 224 kg/ha N, 112 kg/ha P_2O_5, and 112 kg/ha K_2O was used on the control plots in 1968, 1969, 1970. Where necessary, plots were irrigated with water to equal the liquid volume of the maximum sludge rate.

Crop yields - Soybeans were planted in 1967. In 1968, grain sorghum and Reed canary grass were grown, and in 1969 and 1970 corn and Reed canary grass were grown.

Whole soybean plants were harvested when the lower leaves began to yellow with maturity. There appeared to be a toxic condition, especially in the control lysimeters. The toxicity probably arose from a high Zn content in the soil (see Table 32) that apparently resulted from solubilization of Zn from the galvanized metal containers used in the construction of the lysimeters.

Yield data for soybeans are shown in Table 15. Sludge-treated plots produced significantly better grain and total plant yields than did the controls. Sludge additions ameliorated the toxic condition that was apparent in the controls.

Table 15. Soybean yields from South Farm lysimeters, 1967.

Sludge Treatment	Whole plant	Grain
	- - - - - - - - g(d.w.) - - - - - -	
25.4 cm	288.8**	88.3
12.7 cm	253.9**	83.0
Control	78.2	24.4

** Significantly different from the control at the 1% level.

Reed canary grass yields are presented in Table 16. Sludge-treated plots yielded significantly more than control plants from the first cutting in each year. Second cutting yields from sludge-treated and control plots were similar. Better physical condition of the soil and/or availability of residual nutrients in sludge-treated plots probably accounted for the higher first cutting yields on sludge-treated plots.

Table 16. Reed canary grass yield means in grams dry weight; South Farm lysimeters.

Sludge Treatment	7/19/68	9/9/68	5/26/69	9/18/69	6/2/70	9/29/70
	- - - - - - - - - - - - - g(d.w.) - - - - - - - - - - - - -					
Maximum	190.3**	165.5	239.1	106.3	115	287
1/2 Maximum	132.5**	140.1	231.6	91.4	117	296
Control	73.5	143.5	78.3	108.8	108	253

** Significantly different from the control at 1% level.

Means for sorghum grain yields were 430.4, 284.9, and 354.8 g for the maximum, 1/2 maximum, and control, respectively. Although the maximum sludge application produced the highest yield, the differences were not statistically significant at the 5 percent level (see Table 17). Sludge-treated plants matured a few days earlier than control plants.

Average corn yields are given in Table 17. Unfortunately, leaf blight affected the 1969 yield. Yields from sludge-treated plots were more severely reduced because the disease affected those plants earlier than the ones grown on control plots.

Table 17. Sorghum (1968) and corn grain (1969, 1970) yields for plants grown in South Farm lysimeters.

Sludge Treatment	Sorghum dry wt. g 1968	Corn 5% moisture g 1969	1970
Maximum	430.4	180.61	638.5
1/2 Maximum	284.9	260.93	474.9
Control	354.8	349.18	537.0

Leaf samples for chemical analysis were collected before the fungal disease symptoms appeared. Sourthern leaf blight affected the plants in 1970, but did not affect the yield as much as the blight in 1968. Contrary to 1969 conditions, sludge-treated plants showed blight symptoms later than the control plants.

Plant chemistry - Addition of 2.5 cm of sludge containing 2000 ppm N provides about 250 kg/ha of N approximately half of which is in the NH_4-N form. Therefore, during each of the first two seasons, the equivalent of several thousand kg/ha of N was added. Plants were analyzed for total N to determine how sludge application rates affected N content.

Nitrogen contents for soybean leaves and grain are presented in Table 18. Leaf values were 3.42, 3.75, and 4.45 percent for the three sludge application rates in ascending order. Nitrogen contents of the grain were 3.94, 4.61, and 4.87 percent for increasing sludge application rates. Thus, N in plant tissues increased as expected with increasing applications of N from the sludge.

Table 18. Nitrogen content of soybean plants grown in South Farm lysimeters, 1967. Data are reported on oven dry (60°C) basis.

Sludge Treatment	Leaves	Grain
	- - - - - - - - %	- - - - - - - -
25.4 cm	4.45	4.87
12.7 cm	3.75	4.61
Control	3.42	3.94

Total percent nitrogen content means for three cuttings of Reed canary grass, one from each of the three 1968, 1969, and 1970, are listed in Table 19. Nitrogen values ranged from 2.13 to 4.09 percent dry weight for a control and maximum sludge rate, respectively. The high N fertilization rates had no apparent deleterious effects on the grass crops, and total contents were not significantly changed by sludge applications.

Table 19. Total N content of Reed canary grass and sorghum leaves from South Farm lysimeters. Data are reported for oven-dried (60°C) tissue.

Sludge Treatment	Reed Canary Grass			Sorghum
	1968	1969	1970	1968
	- - - - - - - - - - - - - - % - - - - - - - - - - - - -			
Maximum	4.09	3.11	2.75	2.49**
1/2 Maximum	3.89	2.88	2.41	2.37**
Control	3.47	2.13	2.65	1.48

** Significantly different from the control at the 1% level.

Concentrations of total N in grain sorghum leaves are also listed in Table 19. They are 1.48, 2.37, and 2.49 percent with increasing sludge application rates, respectively. The significant increases in total N contents of the crop with the abnormally high fertility levels resulting from sludge irrigations were not very different from results determined in a state-wide plant nutrient survey (165).

Nitrogen concentrations in corn leaves and grain from the 1969 crop are presented in Table 20. Each of the plant parts contains more N as a result of sludge fertilization than was accumulated from inorganic fertilization. The higher fertility level achieved by addition of several tens of centimeters of sludge is reflected in unusually high concentration levels of N in corn grain.

Digested sludge has a rather high complement of heavy metals especially with regard to Cu, Zn and Mn which are essential in small quantities to plants and Cd, Pb and Cr which are nonessential. Both essential and nonessential heavy metals are usually toxic to plants at relatively low available soil concentrations. Because they are polyvalent, they are held rather tightly by the soil colloids which reduces their availability

Table 20. Nitrogen concentration of corn raised on South Farm lysimeters, 1969. Data are reported for oven-dried (60°C) tissue.

Sludge Treatment	Leaf	Grain
	%	
68.8 cm	1.99	2.31
34.4 cm	2.12	2.19
Control	1.84	1.75

to plants. In the case of sludge, they are present as hydroxide (76) or other precipitated or complexed form in the solid phase. As long as soil pH remains neutral, or above, heavy metals should not become very available to plants from a sludge source. However, the potential hazard of toxicity from extended application of digested sludge to cropped land is a distinct possibility because one incident has been reported where raw sewage was applied on the same area for several decades (130).

Copper, Mn, Ni, and Zn concentrations found in soybeans are listed in Table 21. Lead and Cr were not detected. Copper concentrations in the grain of 8.1 to 9.0 ppm were considerably less than the 29 to 32 ppm found in leaves. Concentrations measured in this study were rather high, but differences in Cu, Ni and Zn contents in tissues due to sludge treatment were not significant.

Manganese concentrations in soybean leaves were significantly different for sludge treatments while Mn levels in the grain were not. It is easier to influence the leaf composition than it is the grain. Concentration levels in leaf tissue ranged from 62 to 145 ppm for the control and maximum rate, respectively.

Zinc concentrations in leaves and grain of sludge-treated and control plants were unusually high. Since the control plants were also high in Zn, it was obvious that much of the Zn found in the plant samples came from the contaminated soils and not entirely from the sludge. Zinc levels in all plants, including controls, were sufficiently high to be considered in the toxic range.

Even though sludge treated plants contained higher concentrations of Zn than the controls, the treated plants showed less toxicity. These results support the theory that Zn and P interact and since the sludge added the equivalent of several hundred pounds per acre P, it may have restored a more normal ratio between the elements, thereby reducing toxicity effects.

Table 21. Selected microelement concentration levels in soybean leaves and grain from South Farm lysimeters, 1967. Data are reported for oven-dried (60°C) tissue.

Sludge Treatment	Mn	Zn	Cu	Ni
	ppm			
<u>Leaves</u>				
25.4 cm	145**	1186	29	7.2
12.7 cm	129**	1251	29	7.2
Control	62	827	32	5.2
<u>Grain</u>				
25.4	38	178	8.1	7.6
12.7 cm	58	178	9.0	7.3
Control	42	137	8.1	11.0

** Significantly different from the control at the 1% level.

Nickel concentrations in leaves and grain were approximately the same, and differences due to sludge treatment were not significant.

Micronutrient concentrations in Reed canary grass are presented in Tables 22, 23 and 24. Cadmium and Cr were not detected. Manganese concentrations were increased with increasing sludge application rates in both 1968 and 1969. Manganese concentration levels in second cuttings for all sludge treatments were higher than the controls. Copper concentration levels reflected the added sludge in the first cutting but not in the second. Zinc concentration levels in the grass tissue were exceedingly high in all cuttings but only in the second cutting of 1968 were the levels significantly associated with sludge applications.

Table 22. Chemical element concentration levels in two cuttings of Reed canary grass from South Farm lysimeters, 1968. Data are reported for oven-dried (60°C) tissue.

Sludge Treatment	Mg 7/19	Mg 9/9	Mn 7/19	Mn 9/9	Zn 7/19	Zn 9/9	Cu 7/19	Cu 9/9
	- - % - -		- - - - - - - - - - ppm	- - - - - - - -				
50.8 cm	.226	.198	104**	154	845	823**	33*	32
25.4 cm	.223	.350	49**	96	895	976**	25*	40
Control	.292	.300	31	30	1168	635	18	40

* Significantly different from the control at the 5% level.
** Significantly different from the control at the 1% level.

Table 23. Selected macroelement and microelements in two cuttings of Reed canary grass from South Farm lysimeters, 1969. Data are reported for oven-dried (60°C) tissues.

Sludge Treatment	Ca	Mg	Cu	Fe	Ni	Zn	Mn
	- - % - -		- - - - - - - - - - ppm	- - - - - - - -			
			5/26/69				
50.8 cm	.259	.194**	17	156**	5.1	595	114**
25.4 cm	.229	.196**	12	142**	4.3	585	62**
Control	.197	.112	9	125	4.0	550	32
			9/18/69				
68.8 cm	.142		5.0	76	4.6	1225	193**
34.4 cm	.386		5.1	71	3.0	1070	94
Control	.174		7.8	63	1.4	1030	36

** Significantly different from the control at the 1% level.

Table 24. Concentrations of macroelements and microelements in Reed canary grass from South Farm lysimeters, 1970. Data are reported for oven-dried (60°C) tissue.

Sludge Treatment	P	K	Ca	Mg	Fe	Mn	Zn	Cu	B	Na	Al	Si
	MACROELEMENTS				MICROELEMENTS							
	----- % -------				---------- ppm -------------							%
Maximum	.263	2.34	.40	.225	156	100.0	419	13	12	303	123	1.80
1/2 Maximum	.246	2.08	.39	.209	158	50.4	416	11	9.8	300	109	1.40
Check	.235	2.36	.36	.171	162	52.0	509	10	8.7	443	90	1.59

Concentrations of nutrient elements in sorghum leaves and grain are shown in Table 25. Calcium, Mg and Cu contents did not vary significantly with sludge treatment. Like soybeans, nutrient element contents of the grain were much lower than that of the leaves. This phenomenon could be useful if sludge application induces higher than normal heavy metal uptake, particularly where the edible plant part is the grain.

Zinc and Mn concentrations in leaves and grain and Fe concentrations in grain showed highly significant increases with sludge applications. Nickel, Cr and Pb were not detected in leaves or grain.

Table 25. Chemical element concentration levels in sorghum leaves and grain from South Farm lysimeters, 1968. Data are reported for oven-dried (60°C) tissue.

Sludge Treatment	Mg	Ca	Cu	Fe	Zn	Mn
	%	----------	----------	ppm	----------	----------
			Leaves			
50.8 cm	.414		32		717**	173**
25.4 cm	.422		41		589**	76**
Control	.220		38		252	16
			Grain			
50.8 cm	.162	62	3.25	56**	60**	14**
25.4 cm	.161	75	3.44	57**	58**	11**
Control	.137	67	2.86	32	30	5.8

** Significantly different from the control at the 1% level.

Chemical element concentrations found in corn leaves and grain are presented in Tables 26 and 27, and on Mn concentrations in leaves showing a significant response to sludge treatments. Only trace amounts of Pb were detected in a few samples, but Cr and Cd were not present in sufficient amounts to be detectable.

Table 26. Concentrations of macroelements and microelements in corn leaves at two growth stages and grain from South Farm lysimeters, 1970. Data are reported for oven-dried (60°C) tissue.

Tissue and sludge treatment	Zn	B	Fe	Mn	Cu	Na	Al	Mg	Ca	P	K	Si
	ppm							%				
1st Cutting Leaf												
Maximum	158	24.8	107	85.0	20.4	310	62.6	0.29	0.53	0.25	2.10	1.15
1/2 Maximum	166	23.2	83.8	46.0	16.6	155	51.0	0.31	0.53	0.24	1.67	1.21
Check	214	20.5	109	53.5	18.2	248	55.9	0.37	0.54	0.21	1.28	1.12
2nd Cutting Leaf												
Maximum	663	19.5	114	219	18.2	942	78.5	0.42	1.01	0.30	1.40	1.63
1/2 Maximum	563	17.6	104	162	18.8	823	76.0	0.51	1.01	0.34	1.08	1.74
Check	396	16.8	152	164	16.8	592	79.5	0.52	0.86	0.26	1.38	1.51
Grain												
Maximum	222	4.5	88.0	>9.0	8.9	239	<10.0	0.26	<0.05	0.55	0.76	<0.20
1/2 Maximum	179	6.6	103	>11.8	11.5	372	<10.0	0.24	<0.05	0.53	0.91	<0.20
Check	134	4.2	57.2	>8.0	7.3	<160	<10.0	0.21	<0.05	0.42	0.68	<0.20

Table 27. Chemical element concentration levels in corn leaves from South Farm lysimeters, 1969.

Sludge Treatment	Ca	Mg	Fe	Mn	Zn	Cu	Ni
	- - % - -		- - - - - - - -	- - ppm - -	- - - - - - - -		
68.8 cm	0.761	0.593	215	153**	1120	12	1.0
34.4 cm	0.797	0.869	147	45**	1031	13	1.1
Control	0.755	0.674	181	28	881	17	1.1

** Significantly different from the control at the 1% level.

Boron and Cu contents decreased or remained about constant from the first sampling to the second (at silking) in 1970 leaf tissues. The other elements increased in concentration in samples collected at the later growth stage. Most showed increases in concentration with addition of sludge, but concentration levels were not affected as much in grain as they were in leaf tissues.

Table 28 shows the oil concentrations in corn grain as determined on single kernels with nuclear magnetic resonance spectroscopy. In both years the oil concentration in sludge treated grain is higher than in control grain. However, the differences were not determined to be statistically significant.

Table 28. Oil concentrations in South Farm lysimeter corn grains.

Sludge Treatment	1969	1970
	- - - - - - - %	- - - - - - -
Maximum	4.32	3.88
1/2 Maximum	4.22	4.23
Control	4.20	3.70

The sludge-treated plants generally exhibited enhanced Zn, Mn and Fe uptake. This enhanced uptake may be partly a function of addition of the elements in sludge, but there is good evidence that some of it may be an indirect effect of sludge addition. In no case has there been evidence of plant toxicities resulting from sludge addition in the four years of this lysimeter study.

Chemistry of soils - Mean soil test values for pH and concentration levels of available P and K as determined for combined soil samples from all South Farm lysimeter plots are shown in Table 30. The 1967 values preceded planting and sludge applications. At the 0 to 10 cm depth, soil pH values appeared to be unaffected by sludge treatments except in 1969, when sludge treatments caused an increase in soil pH values. Available phosphorus and potassium concentration levels were increased by sludge treatments, except for potassium levels in 1970.

Table 30. Mean values for pH and concentration levels of available phosphorus and potassium as determined for soil samples collected from the 0 to 10 cm depth of soils represented in the South Farm lysimeters.

Sludge Treatment	pH	P kg/ha	K kg/ha
1967			
Maximum	5.7	197	400
1/2 Maximum	5.8	209	497
Control	5.8	204	437
1968			
Maximum	5.8	504**	721**
1/2 Maximum	5.6	[illegible]**	517**
Control	5.7	[illegible]	335
1969			
Maximum	6.2*	[illegible]**	804**
1/2 Maximum	5.9*	[illegible]**	568**
Control	[illegible]	[illegible]	578
1970			
Maximum	[illegible]	[illegible]**	417
1/2 Maximum	5.2	[illegible]**	329
Control	5.2	[illegible]	428

* Significantly different from the control at the 1% level.

** Significantly different from the control at the 5% level.

Concentration levels of organic carbon in South Farm lysimeter plots are given in Table 31. Organic carbon contents increased in sludge treated plots, while the controls remained relatively constant as expected. However, even with additional sludge applications soil organic carbon concentration levels were not increased above levels found in 1968.

Table 31. Organic carbon contents in the 0 - 10 cm depth of South Farm lysimeter soils (percent dry weight).

Sludge Treatment	Pre-treated	8/21/67	10/20/67	5/12/68	5/19/70
Maximum	1.90	2.51**	3.41**	5.98**	4.74**
1/2 Maximum	1.98	2.19**	2.95**	3.37**	3.17**
Control	1.91	1.90	1.78	1.82	2.21

** Significantly different from the control at the 1% level.

Chemical element concentration levels as determined by 0.1 N HCl extraction are presented in Table 32. All chemical element concentration levels determined increased, relative to the controls, in sludge amended plots.

Table 32. Heavy metal concentration levels (0.1 N HCl extractable) in South Farm lysimeter soils, sampled 5/19/70.

Sludge Treatment	Mg	Fe	Zn	Mn	Pb	Cd	Ni	Ca	Cr	Cu
	ppm									
94 cm	1005	3365	1234	354	179	21	12	4256	57	199
47 cm	624	1919	857	329	98	12	8	2784	29	121
Control	409	504	679	327	26	3	5	2641	<10	40

Two soils were sampled in the spring of 1970 at the depths of 0-10, 15.5-20 and 30-33 cm. The soil samples were extracted with 0.1 N HCl in the usual manner and analyzed for the several chemical elements presented in Table 33. Manganese was the only element which showed any greater concentration levels with depth than in the soil surface. Manganese uptake by plants has generally been enhanced with sludge addition as has its loss in leachate. Perhaps sludge has increased the lability of Mn and it has moved downward in the profile. The higher concentration levels of other elements in surface samples compared to levels in soil samples from lower depths reflect their addition in sludge.

Chemistry of leachates - Figure 19 shows mean monthly concentrations of nitrate-N in leachate (drainage water) collected from the South Farm lysimeters. Analysis showed ammonium-N to be negligible. August 1967 leachates from sludge-treated plots showed somewhat elevated nitrate-N concentrations, but it was November 1967 before the nitrate that had accumulated near the surface from sludge applications appeared in drainage water. After the initial flush, concentrations decreased until June and July, when they increased slightly again. Although the lower sewage treatment was equal to one-half of the maximum, nitrate-N concentrations for the lower sludge treatment were nearly as great as those for maximum sludge applications. Data for the next two seasons showed that the nitrate concentration differences more closely reflected sludge treatments. The reasons for the small difference the first year are not clear.

The 1968-69 and 1969-70 seasons (middle and right graphs, Figure 19) show approximately parallel patterns of nitrate-N concentration from sludge-treated plots. An initial flush of nitrate was produced in the late fall followed by a reduction in concentration during midwinter. Another increase in early spring was followed by a decrease in late spring. The lower winter and higher spring concentrations are probably in part a reflection of organic nitrogen mineralization and nitrification processes.

Nitrate-N concentrations in drainage water from sludge-treated plots have consistently been much higher than the USPHS drinking water standard of 10 ppm. Moreover, they have been several times the levels found in regular field tile drainage water. The amount of ammonium nitrogen added at the maximum rate, for example in 1969, was the equivalent of 1,764 kg/ha of nitrogen. The total N added through 1970 exceeded an equivalent N application of 3400 kg/ha. With N added in amounts 15 to 30 times that of the crop requirement, large leaching losses of N would be expected. The conclusion is that the first limitation to sludge loading rate is that of nitrogen content if nutrient losses to the environment are to be minimized.

Table 33. Heavy metal concentration levels (0.1 N HCl extractable) with depth in South Farm lysimeter soils, sampled 5/19/70.

Soil Type and Treatment	Depth cm.	Zn	Fe	Pb	Cd	Mn	Ca	Cr	Cu	Ni	Mg
		ppm									
Saybrook	0 - 10	1110	3450	157	20	315	4625	58	219	13.0	1075
94 cm. sludge	15.5 - 20	390	400	15	5	467	2050	<10	33	3.5	300
	30 - 33	30	350	15	5	295	1575	<10	29	5.0	380
Saybrook	0 - 10	910	1575	95	11.5	440	3400	30	124	9.0	730
47 cm. sludge	15.5 - 20	360	400	27	2.0	350	2575	<10	30	6.0	333
	30 - 33	185	425	20	1.5	190	1475	<10	27	2.0	438
Cowden	0 - 10	930	2025	95	13	172	2575	30	116	9.0	715
94 cm. sludge	15.5 - 20	710	350	20	3.5	152	1562	<10	27	6.0	208
	30 - 33	325	225	20	1.5	135	1875	<10	30	2.0	285
Cowden	0 - 10	770	1800	70	10	225	2225	22.5	87.5	8.0	480
47 cm. sludge	15.5 - 20	415	300	27	2.5	130	1688	<10	35	2.0	188
	30 - 33	110	250	20	2.0	370	1600	<10	30	3.5	238

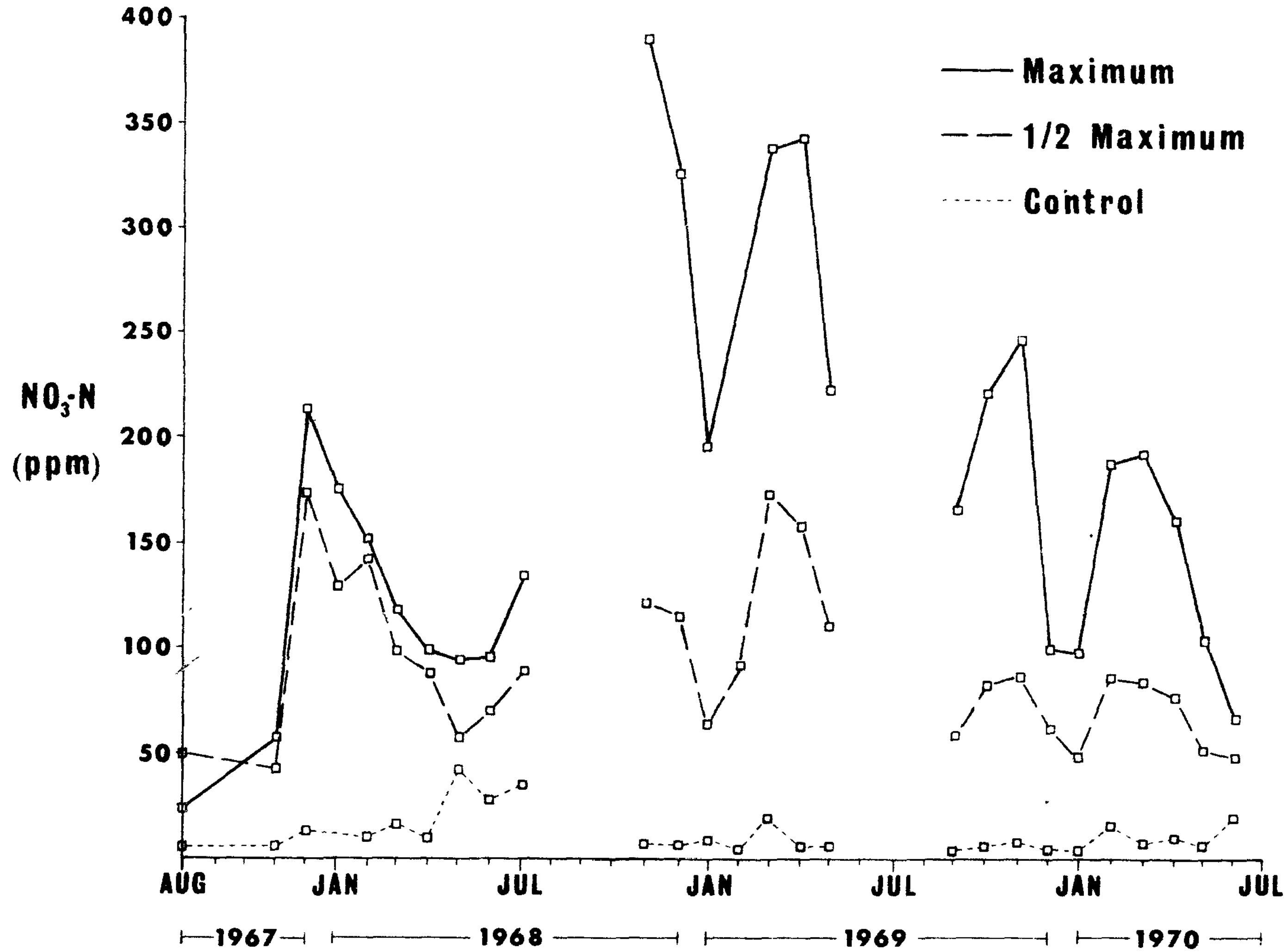


Figure 19. Average monthly concentrations of NO_3-N in drainage water from lysimeters at Urbana.

Drainage water from check plots remained at a level generally below 10 ppm of nitrate-N. The anomalous increase in nitrate-N concentration in May 1968, (Figure 19) was due to severe and almost immediate leaching of applied fertilizer. Almost 13 percent of the added N was leached in water collected within one week after the rain.

Toward examining the interaction of drainage water volume and nitrate-N concentration, the two parameters are diagramed as monthly means for the plots receiving the maximum sludge loading in Figure 20. The lower sludge treatments showed the same pattern. Generally nitrate-N concentration does not appear to be correlated with leachate volume. It might be expected that large volumes would produce a dilution effect and thus be inversely related to N concentration levels in leachate. This relationship is not evident in Figure 20 where the total drainage effluent was collected during a drainage event. More definitive information about interaction effects are forthcoming from the various kinds of water samples collected by the more sophisticated sampling equipment in use at the larger field lysimeter facility on the Northeast Agronomy Research Center.

Concentration levels for several chemical elements in leachate samples from two representative spring collections are presented in Table 34. Copper, Cr and Pb were not detected in leachate collected during these periods nor were they detected in other water samples collected from the South Farm lysimeters. Zinc and Mg showed a several fold increase in concentration under the influence of sludge additions, and also attained relatively high absolute concentrations in the leachate. Losses of these metals might be caused by ion exchange with soluble elements in sludge such as ammonium nitrogen. However, the evaluation of these data must be tempered by the fact that it is very likely that some of the metals which occur in the soils at exceedingly high concentration levels as a result of dissolution of the container surfaces, may have been more readily mobilized and leached following sludge applications than would have been the case for normal soils amended with sludge.

Table 34. Selected chemical element concentration levels in South Farm lysimeter leachate water.

Total sludge applications	Cd	Mn	Zn	Mg	Ni
	ppm				
<u>3/20/68</u>					
25.4 cm	0.08	0.26	25.2	141	0.12
12.7 cm	0.04	0.07	8.4	63	0.08
Control	<0.01	0.01	2.3	10	0.03

Table 34. (cont.) Selected chemical element concentration levels in South Farm lysimeter leachate water.

Total sludge applications	Cd	Mn	Zn	Mg	Ni
	ppm				
4/10/69					
50.8 cm	0.08	0.19	23.6	152	0.10
25.4 cm	0.04	0.06	7.1	65	0.07
Control	<0.01	0.02	3.0	9	0.02

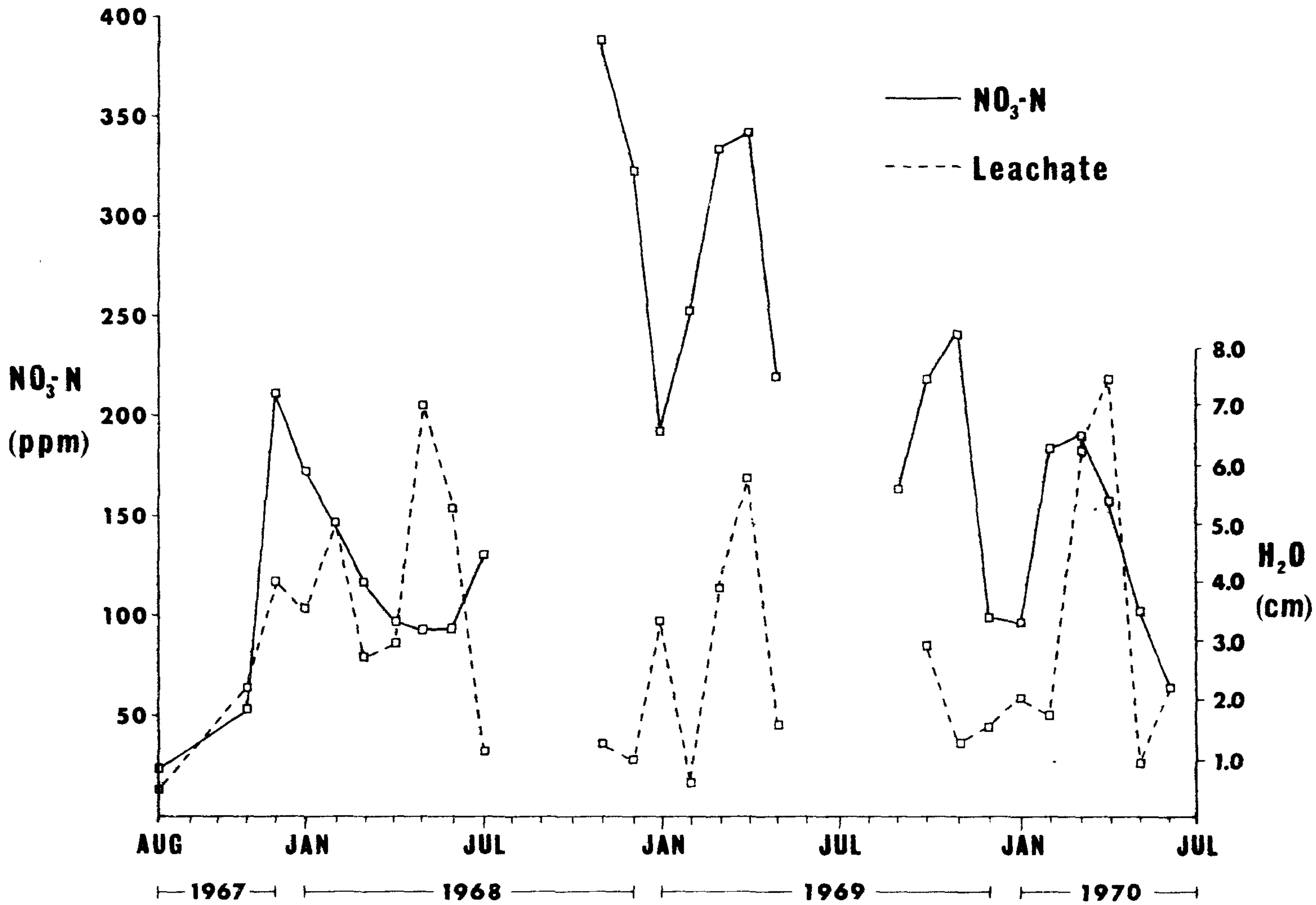


Figure 20. Average monthly concentrations of NO_3-N and leachate volume from lysimeters at Urbana. Data for NO_3-N also are shown in Figure 19.

SECTION V

LABORATORY AND SMALL SCALE FIELD STUDIES

Aeration Induced Changes in Liquid Digested Sewage Sludge

Introduction - The chemical composition of digested sludge varies to some extent depending on whether it is of domestic or industrial origin. There are some seasonal variations, too. The digested sludge can be grossly defined as a 2-5 percent solid suspension rich in N, about half of which is present as NH_4-N, P and small amounts of K. The organic C-N ratio is low (less than 5). The liquid phase of digested sludge contains most of the NH_4-N, only a few parts per million organic C, but is saturated with carbon dioxide and methane. The solid phase contains numerous metals such as Cu, Zn, Ni, Fe, Cr, Pb, as hydroxides and sulfides (69)(76).

Digested sludge is the product of a fermentation carried on in an oxygen-free medium at a low oxidation-reduction potential. Disposal of this material on soils will necessitate transportation and storage with exposure to air. The purpose of this investigation was to analyze the chemical and physical changes which occur in the digested sludge upon contact with the air, and to examine the effect of digested sludge on germination of plant seeds.

Materials and Methods - Organic C analysis was performed by the procedure of Mebius (103). This method based on the principle of wet carbon oxidation by a dichromate solution gives only the organic oxidizable C but does not include the C from the carbonates or from methane. Nitrogen was determined by the microKjeldahl procedure of Bremner and Keeney (23) and Bremner and Edwards (22). Values for oxidation-reduction potentials are given in terms of potentials between Pt and H electrodes and were determined by using a conventional calomel reference electrode. The metals (Ni, Cr, Zn, Cu, Pb) were determined with a Beckman Model 979 atomic absorption spectrophotometer.

Seed germination was accomplished by a method similar to that developed by Guenzi and McCalla (50) for their study of soil extract inhibitors.

The seeds were first immersed in the digested sludge for six hours. They were then incubated in a petri-dish on two layers of filter paper (Whatman No. 1) soaked with 5 to 6 ml of digested sludge.

Vigorous aeration of the digested sludge was carried out by passing compressed air through the slurry contained in a 15 liter bottle for 11 days. At different time intervals, aliquot samples were taken and analyzed for their C, N and metal content, pH, oxidation-reduction potential (Eh), sedimentation properties, and odors.

Changes in the chemical and physical properties of the digested sludge upon contact with the air - The results show that the digested sludge properties can be grouped into two categories, namely those that changed rapidly and those that remained unchanged by the aeration. Great and rapid changes were observed in pH, Eh, NH_4-N content (Figure 21), settleability, and odor. On the other hand, organic C, organic N, and nitrite plus nitrate nitrogen contents of the digested sludge were not significantly affected by the aeration process.

After forced aeration of the digested sludge for one day, its pH rose by about one unit, the distinctive smell disappeared, and a light colloidal solution developed that remained cloudy even following centrifugation of the material at 20,000 G for 30 minutes. After the second day of aeration, a sudden jump in the oxidation-reduction potential was observed. The NH_4-N content of the digested sludge regularly decreased to one-half of its original value after six days of aeration. In the course of 11 days of aeration, no decided decrease or increase in total organic C or organic N content was observed. Values fluctuated between the following extremes: 6.48-8.22 gm of organic C per liter and 732-822 mg of organic N per liter. During the course of those experiments, no significant amounts of nitrites or nitrates were detected.

Aeration by bubbling air is too drastic a treatment to simulate the conditions prevailing when digested sludge is transported in tanks and impounded. Therefore, another experiment was carried on with 400 ml of digested sludge placed in a 500 ml beaker which was clamped in a rotary shaker and gently swirled for two weeks. The results obtained were qualitatively similar to those found when compressed air had been passed through the digested sludge - no change in organic C or organic N content, an increase in pH, and a decrease in the NH_4-N content (Table 35). At the end of six days, it was verified that most of the organic C as well as the organic N was still localized in the solid phase of the sludge while the liquid phase contained the NH_4-N, and only small amounts of organic C (0.15 to 1.50 mg C per liter). After two weeks of incubation, the NH_4-N content had been reduced to 174 mg N per liter and no significant amounts of nitrite and nitrate could be detected. These values were for a digested sludge including industrial waste collected at the Calumet treatment plant, Chicago. Similar results were observed

Table 35. Changes in some properties of digested sludge upon contact with the air for 6 days.

	Organic Carbon (mg/liter)	Organic Nitrogen (mg/liter)	NH_4^+-N (mg/liter)	pH	Percent solid
Fresh digested sludge	3220	451	759	7.5	1.20
The same digested sludge aged for 6 days in contact with air	2970	478	322	8.8	1.17

with domestic-type digested sludge obtained at the Urbana-Champaign treatment plant. Whatever the conditions of aeration no solubilization of the metals was observed during a two-week period.

Seed germination in digested sludge - The inhibitory action of digested sludge on seed germination (corn, Illinois maize hybrid: WfgTMS x C103D, Wayne soybean) is indicated by the data in Table 36. The toxic properties were localized in the digested sludge supernatant liquid which was obtained as follows: centrifugation first at 6000 G for 15 minutes followed by centrifugation at 20,000 G for 20 minutes. Sulfide toxicity is unlikely in as much as less than 0.05 ppm S^{2-} were detected in the sludge supernatant liquid while 0.3 ppm are required to affect plants (47). The ash from the sludge supernatant liquor was not toxic. The partial inhibition by total sludge ash indicated the possibility of some salt or toxic metal interference.

Table 36. Inhibitory effect of digested sludge on seed germination.

	Percentage Germination	
	corn	soybean
Control*	100	100
Digested sludge	19	0
Digested sludge supernatant	0	0
Ashes** from digested sludge	50	66
Ashes** from digested sludge supernatant	100	100

* Seed germination in 10^{-4} M $CaCl_2$ aqueous solution.

** Ashes obtained by combustion of the dried material at 500°C for 12 hours.

Seed germination was still inhibited when the assay was performed aseptically with autoclaved digested sludge or with the digested sludge supernatant liquor sterilized by filtration, thus preventing the microbial proliferation observed around the seeds inside the petri-dish.

It was observed that upon storage in a cold room the digested sludge gradually lost its toxicity. An experiment was set up to investigate the persistance of the inhibitory property. For this purpose, three 200-ml aliquots of fresh digested sludge samples were aged for 5 days at room temperature, in the following ways: the first two samples were left undisturbed in a 500 ml beaker, one under vacuum, the other one in contact with the air; the third 200-ml aliquot sample was placed in a 500 ml Erlenmeyer flask and shaken by rotary action to provide for a more vigorous aeration. Table 37 shows that 5 days of aeration improved the digested sludge's capacity to support seed germination. Similar results were noted with the sludge samples obtained from the experiment reported in Fig. 21. The toxicity could not be solely attributed to the low redox potential of the sludge since some samples showing high redox potentials (samples at 2,3,4 and 6 days of aeration) were still toxic. Figure 21 indicates that the toxicity towards soybean seed germination was reduced with decreasing concentration of ammonium in the digested sludge. Ammonia toxicity towards plant development and seed germination have been reported for concentrations as low as 0.20 mH NH_3 (ag). Levels of ammonia in excess to this limit will be found in equilibrium with 700 ppm NH_4^+-N for any pH values above 6.8, assuming that the transformation NH_3 (ag) $\rightarrow$ NH_3 (g) does not occur. Considering the high pH of the digested sludge it is possible that ammonia was at the origin of this toxicity. To confirm this hypothesis, ammonium chloride as an aqueous neturalized solution was added to aliquot sludge samples which had been collected after 6 days of aeration with properties as shown in Fig. 21. The results indicated that, in order to reach toxic levels, ammonium had to be added in concentrations much higher than those usually encountered in fresh digested sludge. This indicated that ammonia was not the main toxic factor possibly because some was lost by volatilization (Table 38).

Table 37. Improvement of seed germination upon aging for 5 days of the digested sludge.

	Percentage Germination	
	Corn	soybean
Control*	100	100
Aged under anaerobic conditions	29	0
Aged in contact with the air	47	47
Aged and swirled in contact with the air	76	76

* Seed germination in 10^{-4} M $CaCl_2$ aqueous solution.

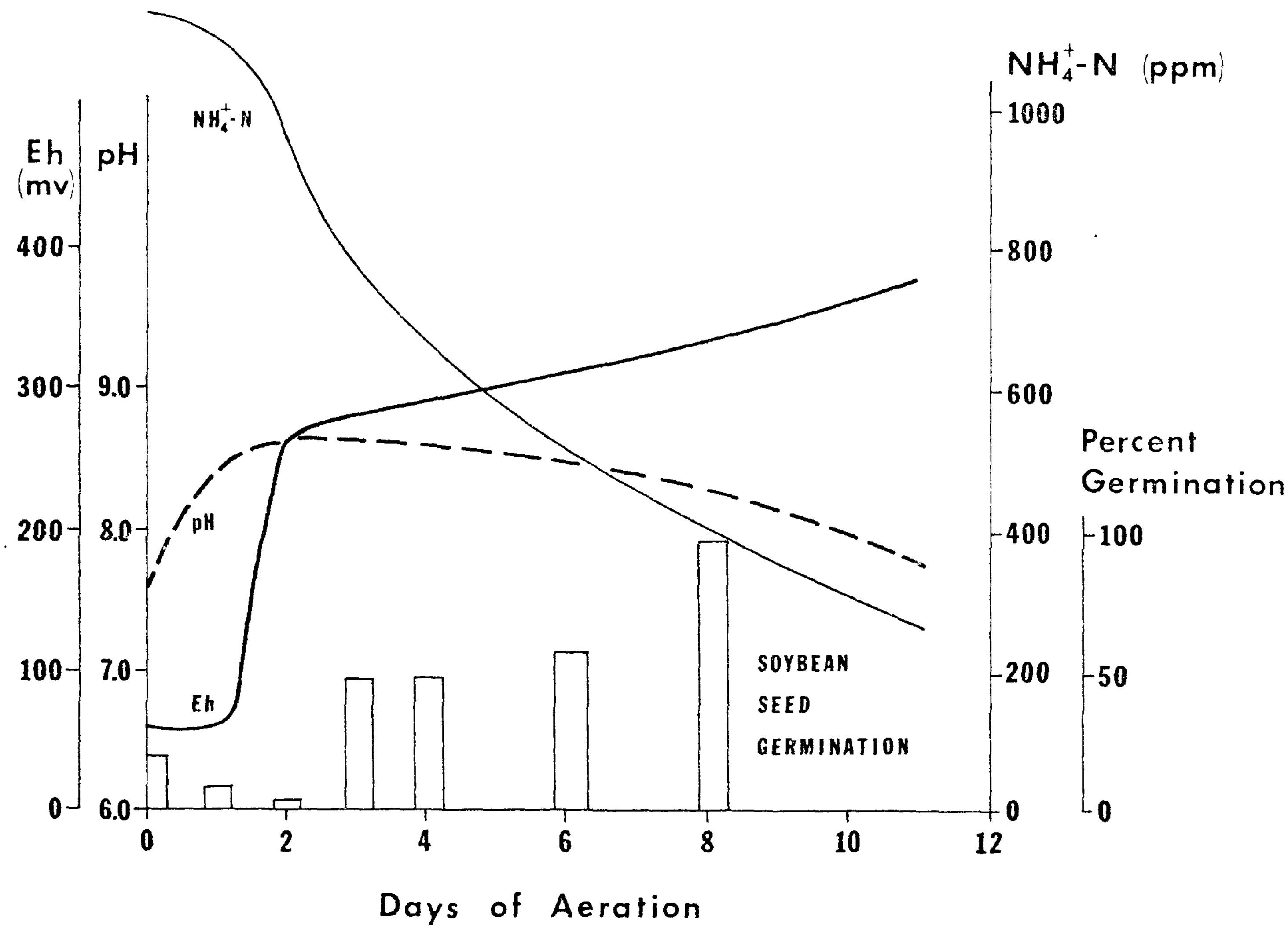


Figure 21. Effects of aeration of liquid digested sludge on Eh, pH and NH_4-N content of the sludge and effect on germination of soybean seed exposed to the sludge.

Table 38. Soybean germination in digested sludge supplemented with ammonium chloride.

Ammonium-nitrogen added to the digested sludge (mg/l)	Percent Germination
0*	60
300	60
600	70
1500	30
15,000	0

* The digested sludge contained 527 mg/l NH_4^+-N. It was the sample collected after 6 days of aeration with properties as shown in Figure 21.

Fresh sludge toxicity toward seed germination was confirmed in a greenhouse experiment. Corn seeds were planted 2.5 cm deep in sand which was supplemented with N, P, K {200 mg $(NH_4)_2SO_4$, 200 mg Ca $(H_2PO_4)_2$, and 200 mg KCl per kg of sand}. A mixture of water and digested sludge was added on the sand surface to bring the moisture content to a level optimum for seed germination. The equivalent addition of 2.5 cm (1 inch) per week of fresh digested sludge totally prevented seed germination, while the application of 2.5 cm of digested sludge aerated for one week did not interfere with the germination. Addition of 1.2 cm of fresh digested sludge reduced plant growth. The toxicity was not limited to seed germination since the addition of fresh digested sludge supernatant to the rooting system of a 1-week-old corn seedling induced degeneracy of the plant within 12 hours.

Discussion - Reduction in ammonium-nitrogen content of the digested sludge is brought about within a few days by aeration or by aging in contact with the air. Ammonium-nitrogen is volatilized since this occurs at the observed high pH and is further evidenced by the odor of ammonia. The ammonium-nitrogen is not oxidized (at least over a 2-week period) and the possibilities of nitrogen assimilation are slight in view of the low carbon-nitrogen ratio of the digested sludge (C/N = 4.3).

The mechanism by which the pH of the digested sludge increases on exposure to air is not fully understood. It is possible that, upon aeration, the concentration of the carbon dioxide dissolved in the digested sludge decreases. An aqueous solution of ammonium carbonate has a pH of 9.8 as opposed to the ammonium bicarbonate solution which shows a pH of 7.8.

No mineralization was observed. The organic C and organic N of the digested sludge are not in a form readily available to biochemical degradation under these laboratory conditions. A similar observation has been made by Premi and Cornfield who reported that no mineralization of the sludge organic N fraction occurred after an incubation period in soil of eight weeks at 30°C (121). Digested sludge is a stabilized material which is not amenable to immediate biodegradation, at least under laboratory study conditions.

One of the main rate limiting factors to the use of digested sludge on agricultural land will arise as a consequence of its high N content. Even though it is assumed that digested sludge organic N is mineralized very slowly, there is enough ammonium-nitrogen in a 3-4 cm application to satisfy the needs of a corn crop. Use of an excessive amount of digested sludge for irrigation may dangerously increase the nitrate content of the ground water. Yet, the possibility of removing N as ammonia by aging of the sludge (in a lagoon for example prior to irrigation) may permit application of up to 15 cm per year for irrigation. In this respect, Premi and Cornfield have noticed that a 4 to 13 percent N loss as ammonia could be expected following land application of liquid digested sludge (121).

Vigorous aeration of digested sludge for two weeks was insufficient to generate any biological exidation of the ammonium presumably because of the absence of nitrifiers. Seeding the digested sludge with activated sludge and aerating the mixture initiates a rapid oxidation of the ammonium (83). In soils, the ammonium added with digested sludge is oxidized to nitrate (121). If a soil harbors a low population of nitrifiers at the time of digested sludge addition, one may expect an accumulation of nitrite since the ammonium cation stimulates the development of Nitrosomonas spp., but ammonia inhibits the development of nitrite oxidizing organisms (1)(111). Premi and Cornfield noticed a lag phase for the appearance of nitrate in soils heavily amended with digested sludge, which they attributed to the presence of organic matter (121). However, the lag phase may have corresponded to an accumulation of nitrite which persisted until the ammonia concentration was reduced and Nitrobacter spp., were able to multiply.

Aging of the digested sludge in the laboratory for one week was sufficient to transform a material otherwise toxic into a medium suitable for seed germination. This would explain why erratic results have been reported about seed germination in soils amended with heavy loads of digested sludge (97)(106)(150). It is likely that high rates of fresh digested sludge on soils which have just been seeded may retard or prevent germination. However, if seeds are planted one week after the first fresh sludge application, or if the sludge has been aged in contact with air, germination may proceed normally. Digested sludge toxicity could be avoided also by planting on ridges with furrow irrigation. It is also possible that fresh digested sludges differ in toxicity - there was a marked difference in the level of toxicity between the Calumet and the Champaign-Urbana fresh digested sludges. The latter gave a maximum of 40 percent inhibition even though it had the same ammonium content as the Calumet sludge. Improvement of seed germination in soils amended with lagooned digested sludge, as opposed to fresh digested sludge, has already been observed by Lunt (97).

Effect of Sludge Application on Soil Atmosphere

Introduction - The fate of nitrogen in sludge which is added to soil is highly dependent on the level of dissolved oxygen in the soil-water. With normal soil conditions, ammonia is oxidized to the mobile nitrate form which may be lost by leaching. If anaerobic conditions are created in soil containing nitrates, denitrification occurs with the usual evolution of nitrogen gas to the atmosphere.

A preliminary laboratory study was conducted to evaluate the effect of sludge application on oxygen concentrations in soil. Rates of denitrification of nitrates added to sludge-soil mixtures were also evaluated. The work reported here is a part of a master's thesis prepared by Sze-Ern Kuo, Dept. of Civil Engineering, University of Illinois.

Description of experiment - Experiments were conducted with 19 cm diameter laboratory columns containing 1.65 m of Plainfield sand. A free water surface was maintained at the bottom of the columns. The columns were equipped with provisions for obtaining samples of the gas in the soil at various depths. Also, provision was made for obtaining liquid samples at various depths by applying a vacuum to sampling tubes. Liquid digested sludge from the Champaign-Urbana waste treatment plant was added each week to the top of the columns without mixing. Four columns were used and rates of application were 0.64, 1.3, 1.9, and 2.5 cm/wk. In addition, 1.3 cm of water was added to each of the columns weekly to simulate rainfall.

Discussion of Results - Samples collected at a depth of 5 cm below the surface of the soil following 0.64, 1.3, and 1.9 cm sludge applications indicated that total anaerobic conditions were not created during the first several hours following sludge application. No gas analysis for the column receiving 2.5 cm/wk of sludge could be obtained because soil pores at the 5 cm level were filled with moisture. Hence anaerobic conditions would be expected near the surface of that column.

Figures 22 and 23 show the variation of the carbon dioxide content in the gas in soil pores at the 15.2 cm and 45.7 cm depths as a function of time during the first four days following application of sludge. Note that the elevation in CO_2 concentration was most pronounced at the 15.2 cm depth soon after the application of sludge and that the concentration continued to decrease with time. During the first day following sludge application the carbon dioxide level was highest in the columns receiving

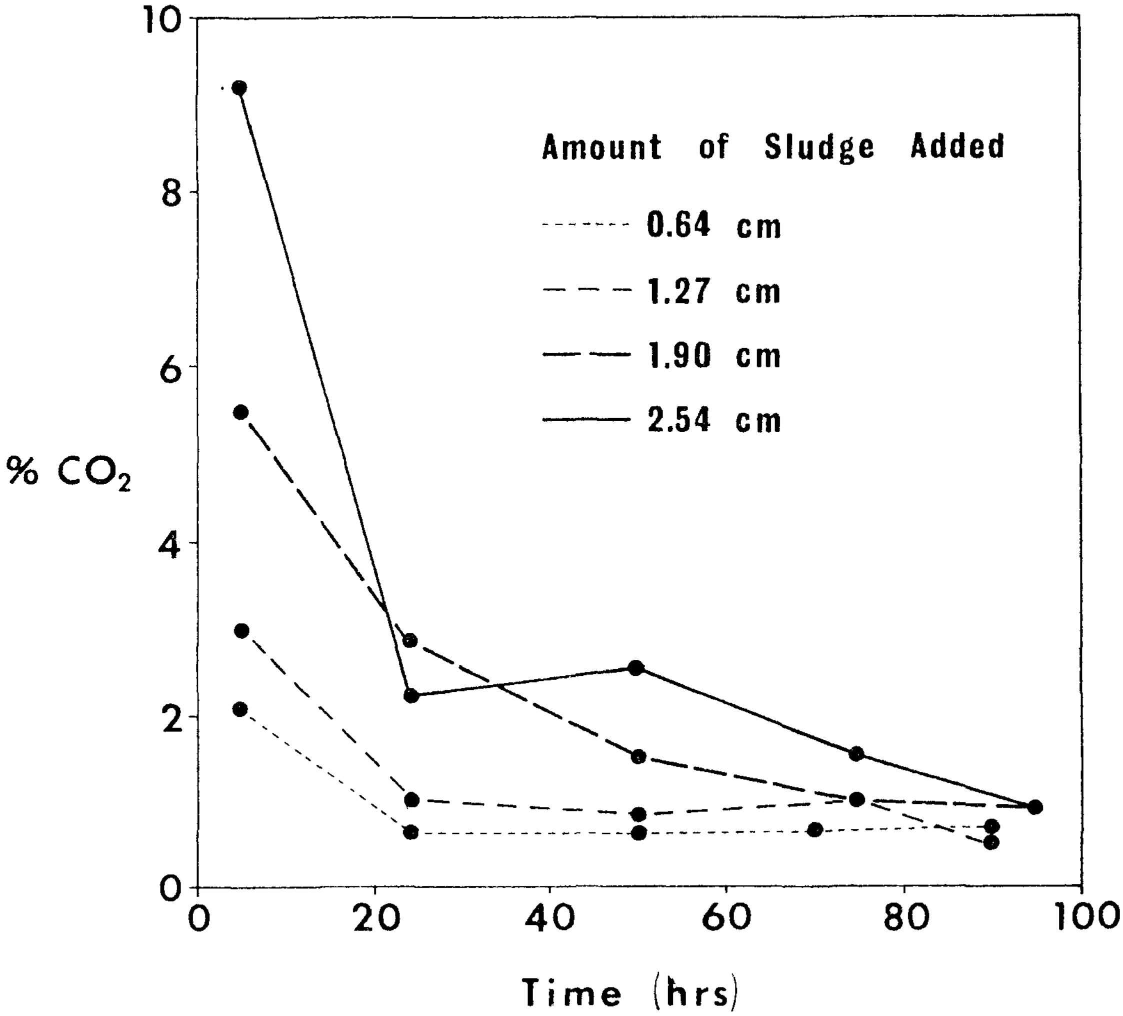


Figure 22. Carbon dioxide concentrations in soil atmosphere at 15.2-cm depth after various sludge applications were made on the soil's surface.

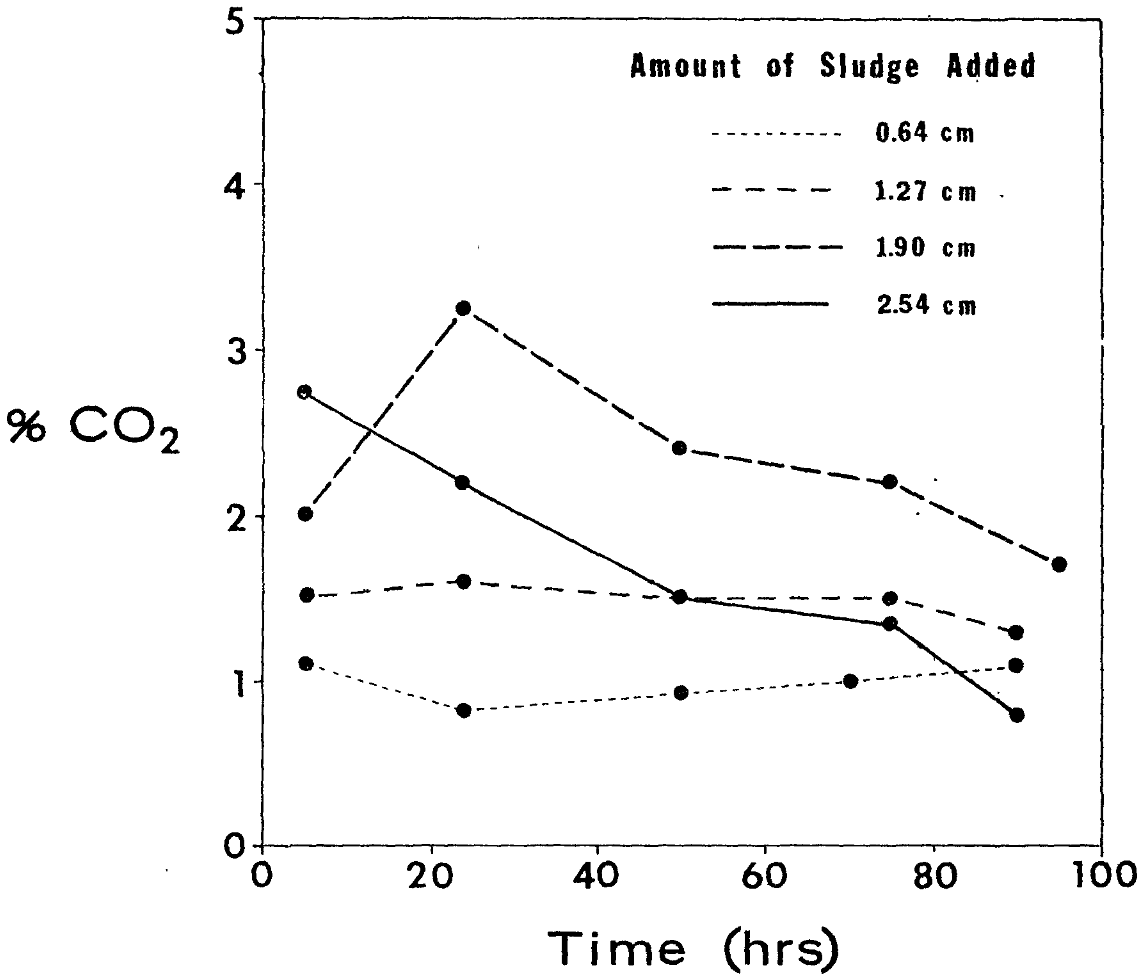


Figure 23. Carbon dioxide concentrations in soil atmosphere at 45.7-cm depth after various sludge applications were made on the soil's surface.

the largest amount of sludge indicating that the amount of microbial degradation of organic materials was greatest in those columns. However, then the carbon dioxide level decreased in the heavily dosed columns to levels below those in the columns receiving less sludge. A possible explanation for this observation is that soil structure was imporved by increasing sludge applications to the extent that gas in soil pores in the columns receiving heavy sludge doses was afforded greater opportunity for interchange with the atmosphere and concentration differences were equalized. However, such a pronounced change in soil structure, especially as it relates to an increase in porosity, was not expected in such a brief period of time.

Figure 24 shows the results of gas analyses at various depths in the columns one week after the first application of liquid digested sludge. In all of the columns, 0 concentrations in the air within the soil decreased with depth and the abundance of carbon dioxide increased with depth. Note that after a week the depletion of 0 in the soil was greatest in the lysimeter receiving 0.64 cm/wk of sludge and the least in the lysimeter receiving 2.5 cm/wk of sludge. This confirms the trend noted in Figures 22 and 23.

The extent of oxygen depletion and the amount of carbon dioxide in the air within the soil at the 0.75 and 1.35 m levels are shown in Figures 25 and 26 for a period of five weeks. Sludge additions were made weekly and the values shown were obtained one week after the latest sludge application *(just prior to addition of new sludge)*.

In the colunns which received 0.64 to 2.5 cm/wk of sludge along with 1.3 cm/wk of water, nitrates were first detected in the column leachate after three weeks of operation. Nitrate profiles in the soil column at the end of the third, fourth and fifth weeks are shown in Figure 27. Note that differences in the nitrate concentration of the soil moisture in the four columns were not proportional to the differences in the amount of sludge which the columns received. Note also that at the end of five weeks soil moisture nitrate concentrations had reached maximum levels of from 300 to 800 mg/l although high levels had not yet been detected in the leachate.

It is of interest to know whether nitrate formed in the soil illustrated in Figure 27 could be evolved as a gas through creation of controlled anaerobic conditions. In separate laboratory studies, nitrates were added to digested sludge to assess the probable maximum rate at which denitrification might be expected to occur. The rate of denitrification of nitrates in sludge was found to be independent of the nitrate concentration until the nitrate level reached 1 or 2 mg/l. The maximum rate of the zero order reaction observed was about 10 mg/l of nitrate per hour at room temperature. The rate depends on the characteristics

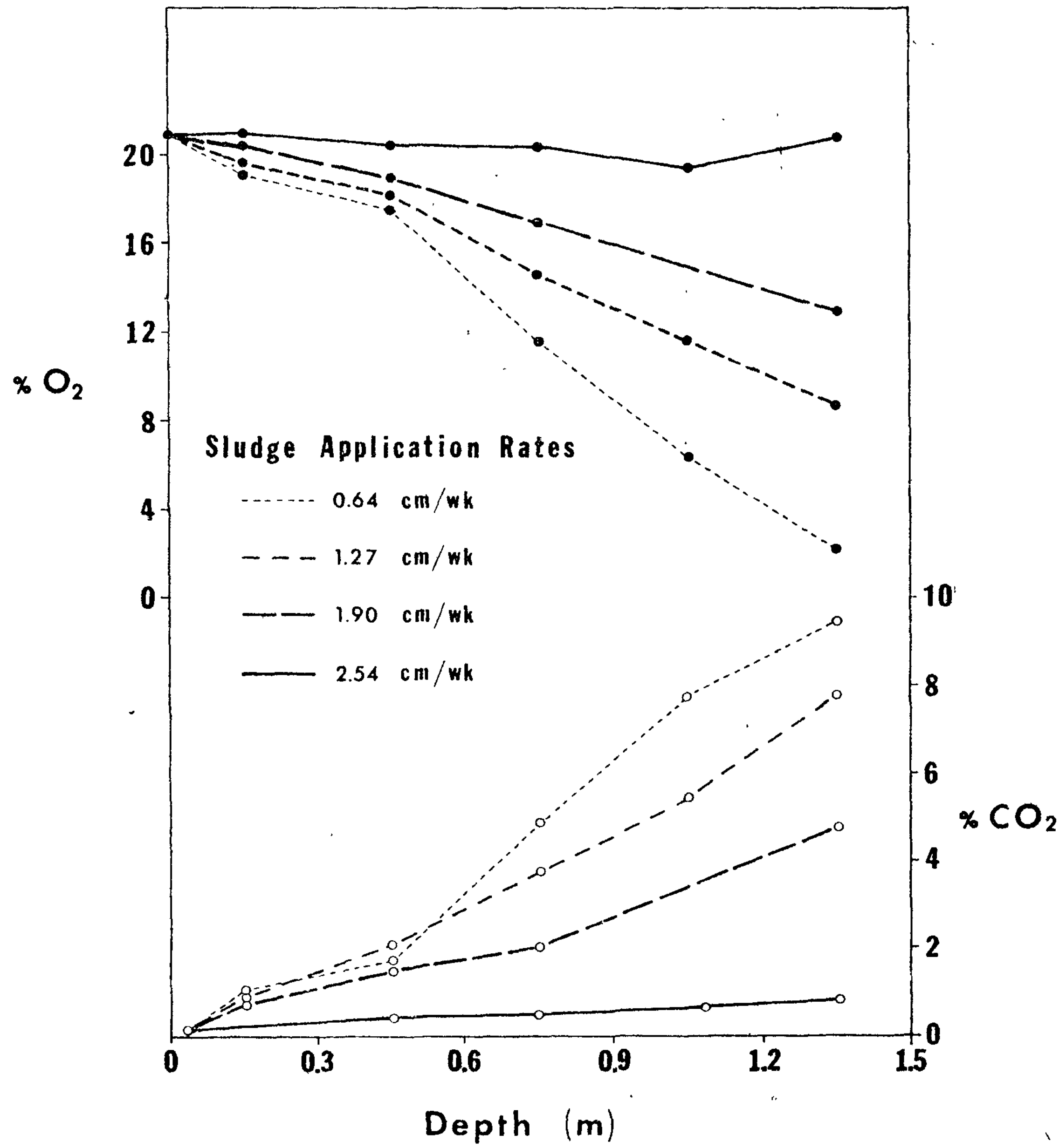


Figure 24. Oxygen and carbon dioxide concentrations of soil atmosphere with depth one week after different thicknesses of sludge had been applied to the surface.

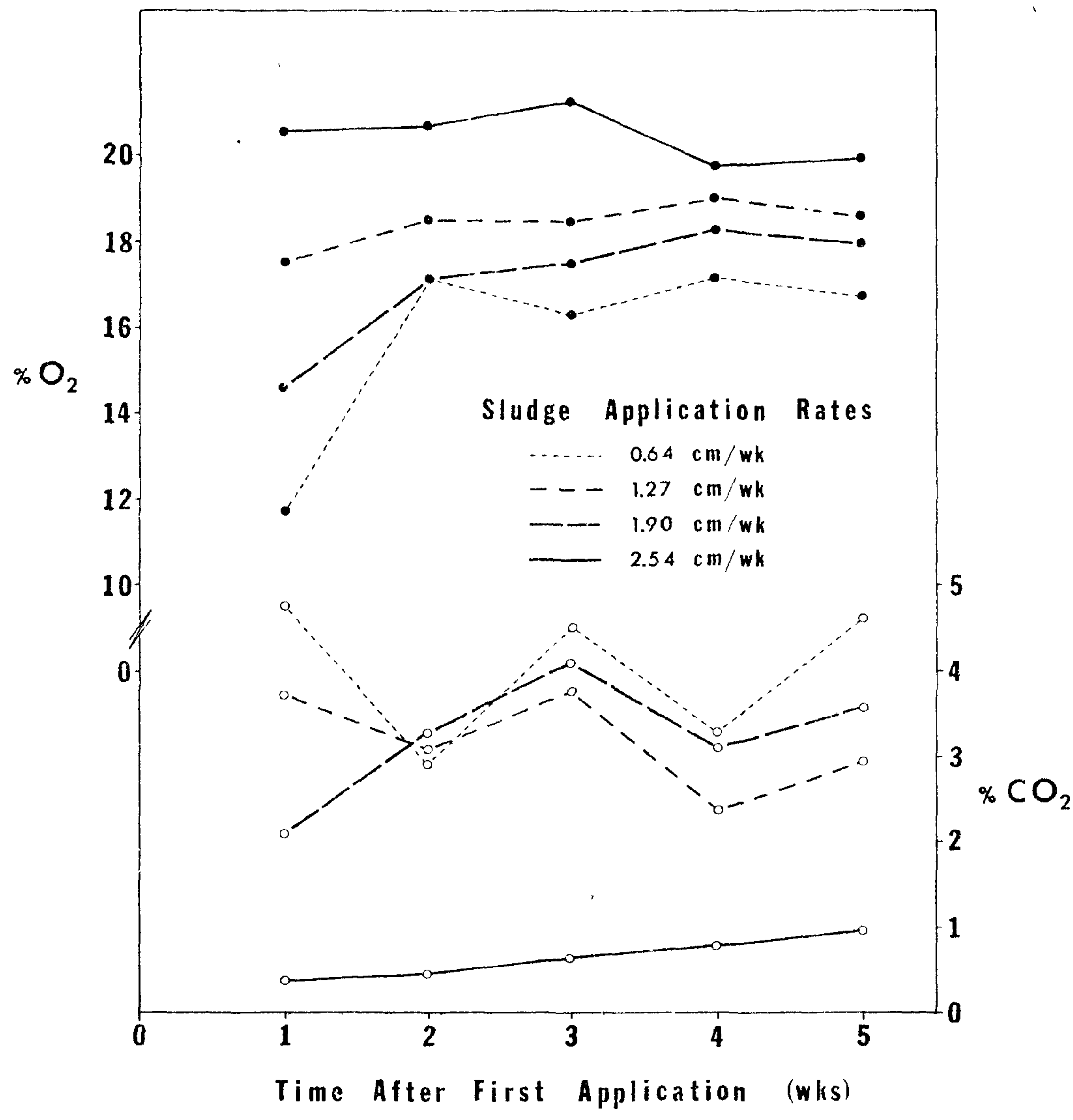


Figure 25. Oxygen and carbon dioxide concentrations of soil atmosphere at 0.75-m depth with time and thickness of sludge applied on the surface as the independent variables.

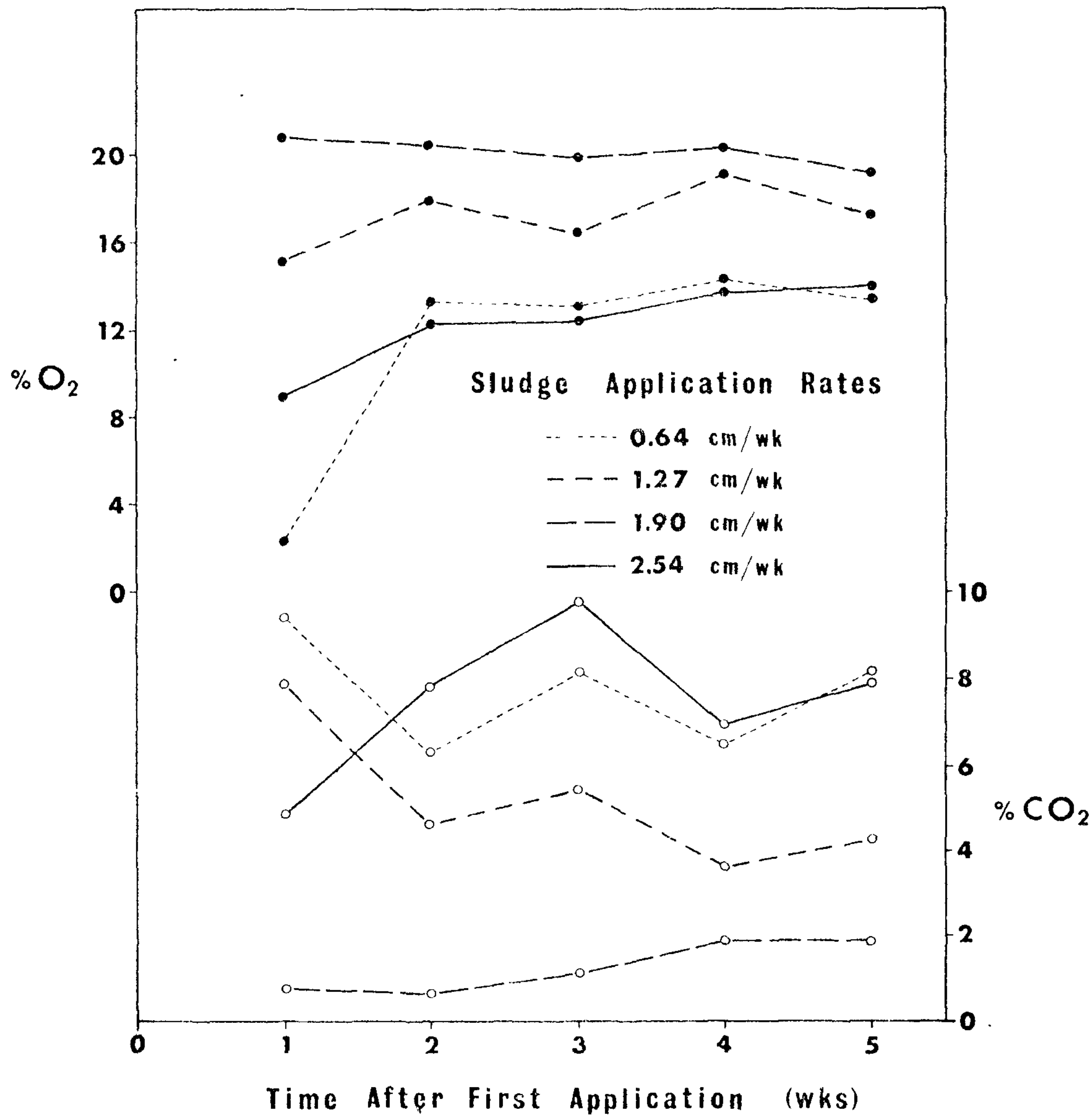


Figure 26. Oxygen and carbon dioxide contents of soil atmosphere at 1.35-m depth with time and application rate of sludge as independent variables.

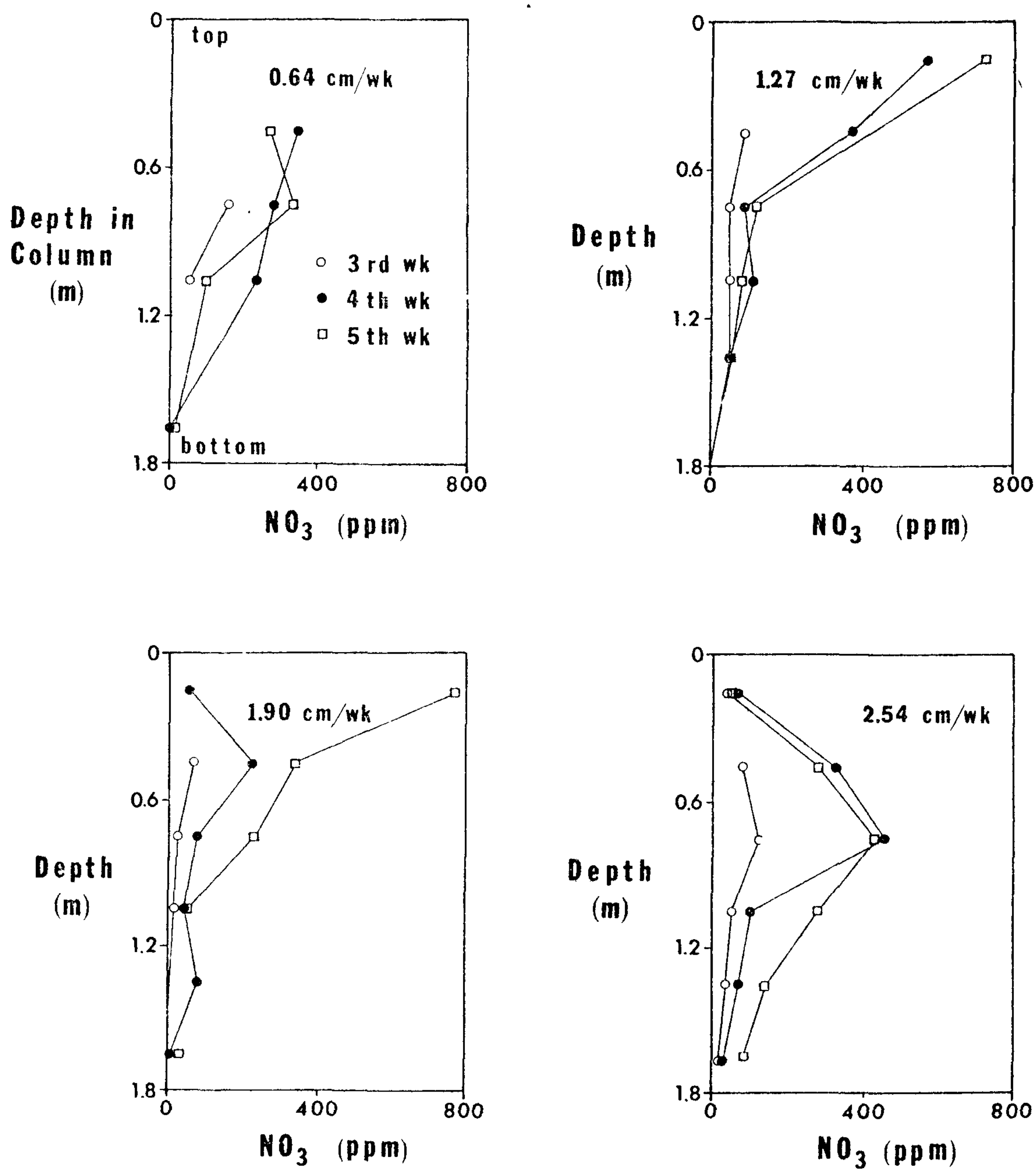


Figure 27. Distribution of NO_3 with depth in soil lysimeters after 3, 4 and 5 weeks. Sludge was added to the lysimeters in the amounts of 0.64, 1.27, 1.90, and 2.54 cm at the beginning of the experiment.

of the digested sludge as illustrated by Figure 28. The figure shows the zero order denitrification curves obtained at various times from the digested sludge maintained at 35°C in a laboratory digester for five days. As seen, the rate of denitrification decreased with storage, i.e., continued digestion. The pH of the sludge was constant at about 7 during the five day period, although the Eh (oxidation-reduction potential) increased from -110 mv to -60 mv during the period.

Figure 29 shows the rate of denitrification of nitrate added to a mixture of 50 gm sludge and 30 gm soil as compared to denitrification in sludge alone. In the sludge-soil mixture the rate of denitrification continued to be independent of nitrate concentration, but denitrification proceeded at a slower rate than in sludge alone.

Summary - Depression of the O content and elevation of the carbon dioxide content of gas in soil pores occurs as a result of sludge application. The deviation from normal atmospheric conditions initially is greatest for soils receiving the greatest amount of sludge, but after a few days, the trend reverses.

Extensive anaerobic conditions, as a result of biological utilization of O,were not created as a result of any of the sludge application rates studied (2.5 cm/wk maximum). High application rates may produce only brief anaerobic conditions by filling the soil pores with moisture.

Loss of N through denitrification of nitrate as a result of anaerobic conditions in soil would be expected to follow zero order kenetics. The rate of denitrification in soil is less than in sludge alone.

Ammonia Volatilization From Digested Sewage Sludge as Related to Land Applications

Introduction - When the economic aspects of a land application system are considered, the possible shortage of available land or the desire to minimize the initial cost of sludge spreading equipment may make desirable the application of more than just enough sludge to provide for the supplemental nitrogen needs of the crop. In view of the probable limitations imposed by water pollutional characteristics of N in sludge, studies of possible inoffensive losses of N are pertinent. A loss which has been poorly quantified is the escape of ammonia from liquid sludge by gas transfer. Such losses could occur in digested sludge storage facilities, during application and following application.

When applied to land, N in digested sludge can have various fates depending on the environmental conditions. It may be lost in runoff from the soil, leached to the groundwater, adsorbed on soil particles, taken up by plants, used in the synthesis of microorganisms, volatilized and lost to the atmosphere, or retained in an unavailable organic form. Its

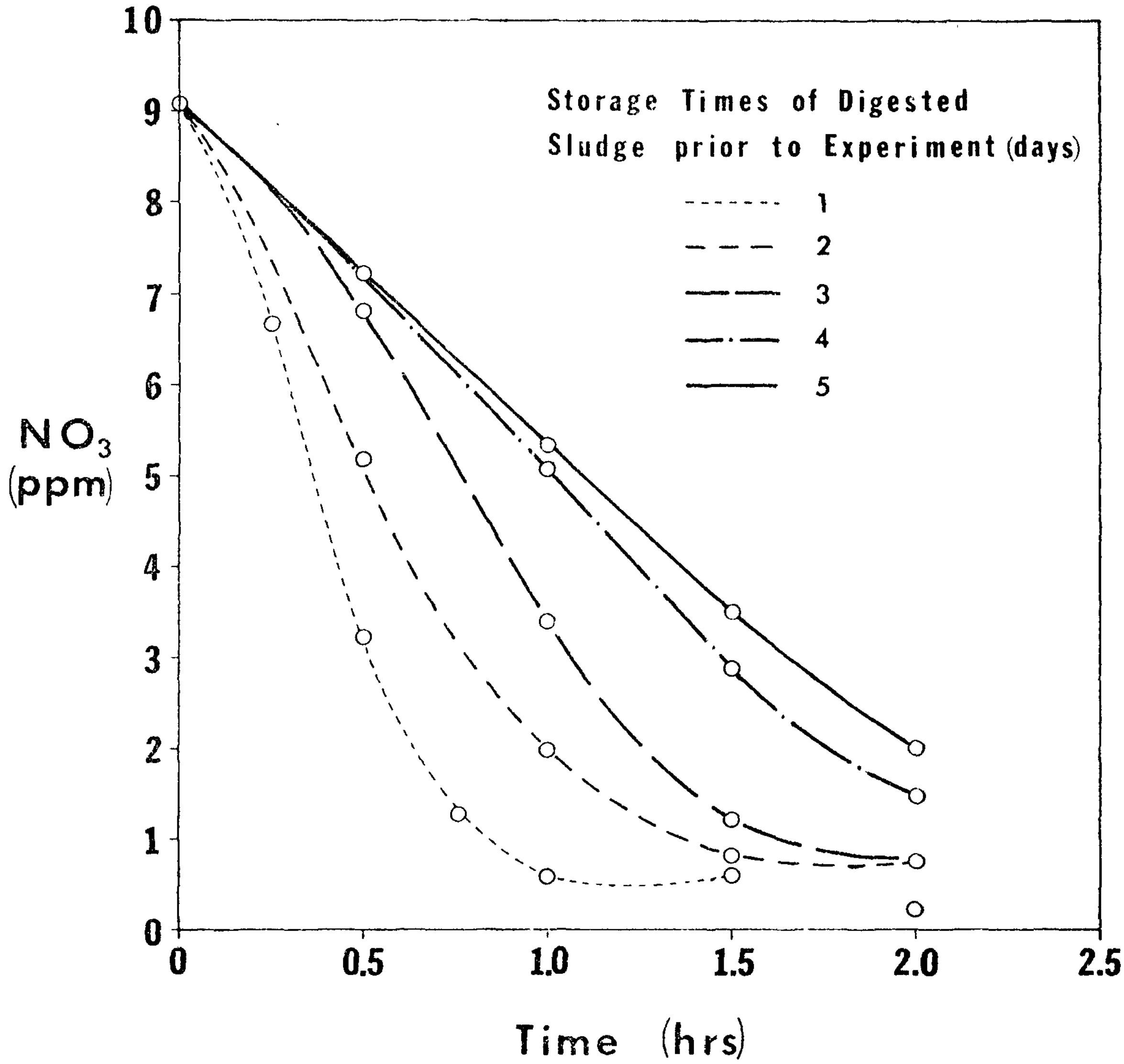


Figure 28. Denitrification of NO_3 added to sludge held for different lengths of time.

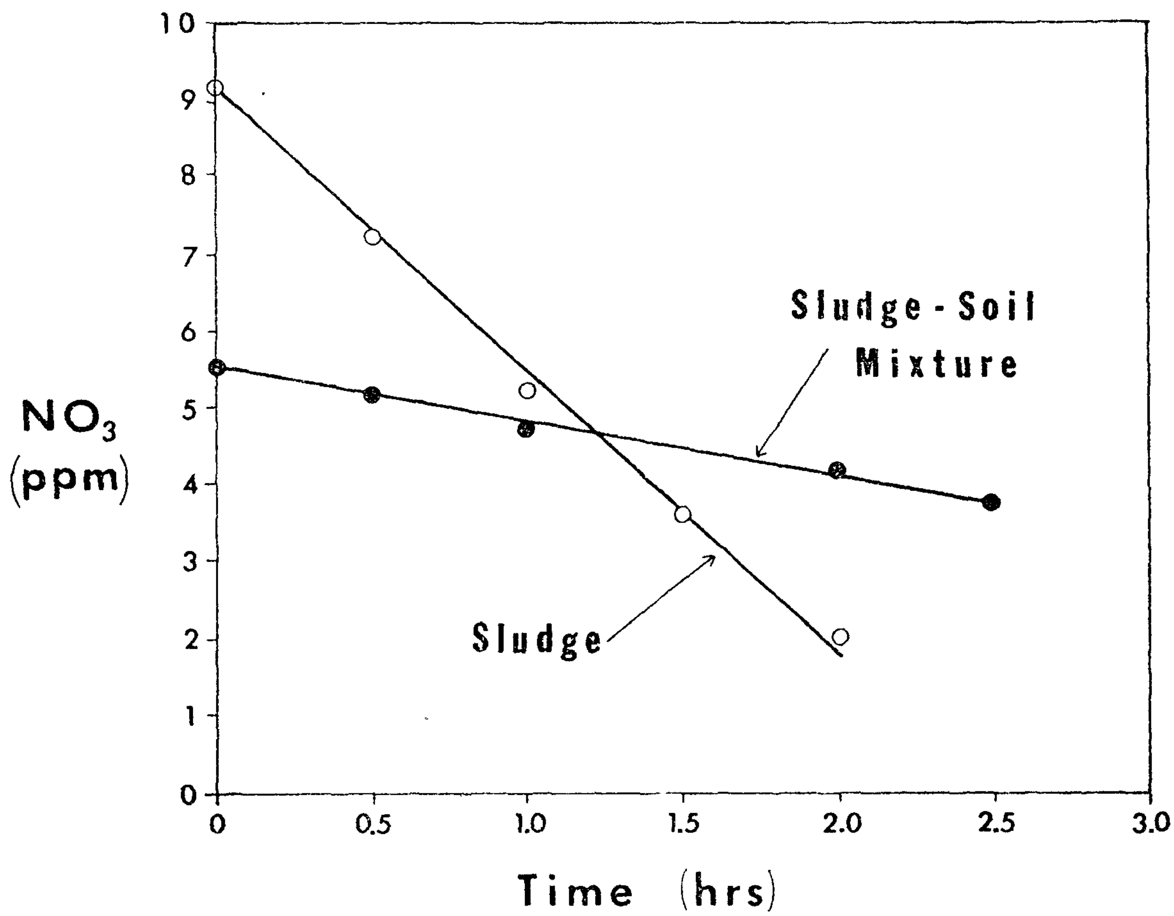


Figure 29. Denitrification of NO_3 added to sludge and a mixture of 50 g of soil.

fate, however, depends greatly on the form in which it exists. For instance, loss by leaching can occur only if a water soluble form, such as ammonia or nitrate nitrogen, is present. Nitrate is very soluble in water in the presence of all common cations (Handbook of Chemistry and Physics, 1955). Ammonia nitrogen is also water soluble, but its solubility is restricted by pH (42). From the equilibrium equation for ammonia in water:

$$NH_3 + H_2O \rightleftharpoons NH_4^+ + OH^- \qquad (1)$$

and from the ion-product of water:

$$H_2O \rightleftharpoons H^+ + OH^- \qquad (2)$$

it can be seen that if the concentration of H^+ is decreased, i.e., the pH is increased, the concentration of OH^- must increase to maintain the equilibrium shown in equation (2), which, in turn, forces a shift in the equilibrium of equation (1) to the left. Sinch NH_3 is a gas, it may be volatilized, thus, resulting in a net decrease in the ammonia concentration in the water.

Nitrogen contained in anaerobically digested sludge is primarily in two forms, ammonia and organic N. This is clearly understandable since any nitrate or nitrite which exists in the digester feed is reduced under the anoxic conditions which exist in the digester. The relative amounts of ammonia and organic N depend among other variables on the sludge age and digester detention time since decomposition of soluble nitrogenous compounds occurs during the acid regression stage of digestion, resulting in the formation of ammonia and other compounds (43).

Considering, then, that sludge contains both ammonia and organic N, it is well to recognize the N transformations which can take place when sludge is applied to soil:

1. Proteolysis is the enzymatic hydrolysis of proteins into peptides and the further hydrolysis of peptides resulting in the release of individual amino acids.

2. The amino acid groups are further decomposed by microbial action into various nitrogen-free organic compounds and ammonia (NH_3). This process is known as ammonification or deamination.

3. The combination of proteolysis and ammonification is commonly known as mineralization - the conversion of organic to inorganic N. The ammonia thus formed can either be utilized by plants and microorganisms or, under favorable conditions, oxidized to nitrates.

4. The oxidation of ammonia to nitrate, which is accomplished in two distinct steps by distinctly different microorganisms, is called nitrification. Nitrosomonas is capable of oxidizing ammonia to nitrite (NO_2^-) and Nitrobacter carries on the oxidation from nitrite to nitrate (NO_3^-). Although a very few other genera of bacteria are capable of performing these oxidations, these two are thought to be of most importance in the process (118).

5. Several microorganisms are able to cause the reverse of nitrification by reducint nitrate to nitrite and further to ammonia. This phenomenon occurs under anoxic conditions (163).

6. Other organisms are capable under anaerobic conditions to transform nitrates to N gas or nitrous oxides. Known as denitrification, this process occurs to a very small extent in well-aerated soils, but can become pronounced in soils saturated with water and containing an abundance of organic matter.

7. Inorganic N can be converted to organic N by plants or by microbial synthesis. This process is collectively known as immobilization.

For more detailed discussions of the above N transformations which can occur in soils and of the relative significance of each, the reader is directed to Alexander (2), Bear (12), Burges (27), and Waksman (163).

From the above information it can be seen that N contained in liquid digested sewage sludge not only can be converted into several different forms, but also has many possible fates when the sludge is applied to soil. Therefore, any attempt to determine how much N will be made available to the crop must be accompanied by an investigation of each of the pathways N can take once the sludge is applied to the soil. It is believed that two of the most important pathways which reduce the amount of N available to the crop are the leaching of nitrate and ammonia to the ground water and the volatilization and subsequent loss of ammonia to the atmosphere.

Scope of the investigation - The purpose of this investigation was to determine the rate of ammonia volatilization from liquid digested sewage sludge in a holding lagoon, and to derive a mathematical model to describe such volatilization as it varies with pH, depth of sludge and mixing. The reasons for conducting the study of ammonia volatilization in this manner were twofold.

Primarily the study was initiated to determine a method of estimating one of the pathways nitrogen in sludge can follow when applied to soil. It was reasoned, however, that almost any facility designed to spread liquid sludge on cropland must, by necessity, include a holding lagoon to accommodate sludge produced between applications or when physical conditions prohibit application. Therefore, a laboratory study using sludge columns of various depths was conducted to simulate conditions existing in sludge lagoons.

Secondly, a mathematical model was developed, which was then programmed in Fortran IV, in order to study the interrelationship of the factors of pH, depth of sludge and mixing as measured by the diffusivity coefficient. Such a study using only laboratory procedures, would have been impossible considering the time available for the investigation.

The laboratory investigation consisted of two main parts: (1) determination of ammonia profiles in sludge and (2) measurement of the ammonia evolved from the surface of sludge. A single 1.5 m column was used to determine ammonia profiles, but five columns with depths ranging from 2.5 cm to 1.5 m were used to demonstrate the effect of sludge depth on ammonia evolution.

Experimental equipment and procedures - Liquid digested sludge from the Champaign-Urbana Sanitary District Treatment Plant in Urbana, Illinois was collected in carboys so that 25 liters was transported to the laboratory for use in the investigation. The treatment at the Urbana Plant consists of split flow to both conventional activated sludge units and trickling filters, followed by anaerobic digestion of the solids, the majority of which come from the activated sludge process. The sludge was taken directly from a sampling valve at the base of one of the secondary digesters.

The average suspended solids content of the sludge was 2.80 percent, and the initial pH and temperature values were 7.5 and 33.5°C. The solids content was determined by the "Residue on Evaporation" method and the pH by the "Glass Electrode Method" as described in Standard Methods for the Examination of Water and Wastewater (1965). The initial ammonia N and organic N concentrations, also determined according to Standard Methods (1965), were 363 mg/1 and 1030 mg/1, respectively. All initial determinations were made on a composite sample from all liquid sampling ports of the 1.5 m sludge column (as described below) taken shortly after the column was filled with sludge. Solids content and pH were determined in triplicate, while ammonia and organic N determinations were made in quadruplicate. Ammonia N determinations were made by distillation into standard boric acid according to Standard Methods (1965), except that the endpoint of the back titration with standard sulfuric acid was determined by drawing a titration curve instead of using an indicator. This titration curve method is believed to give slightly better accuracy. A statistical analysis of the method revealed a coefficient of variation of 2.0 percent (CV = 100 x standard deviation/mean).

Because temperature plays a major role in the evolution of a gas from solution, in the diffusion process and in the biological reactions occurring in sludge, both the temperature of the sludge and the room temperature were monitored. The room temperature remained at 26.4 ± 0.2°C throughout the course of the experiment. Since the temperature of the sludge attained equilibrium (room temperature) after only 30 hours, its effect on the overall experiment was assumed to be negligible.

The column used to contain the sludge and simulate a lagooned situation was fabricated from an 20.3 cm diameter (19 cm inside diameter) plexiglass tube, 1.65 m long with flanges on both ends as shown in Figure 30. A solid bottom cover was bolted to the bottom flanges and a top cover with two access holes was bolted to the top flange. Both top and bottom were sealed with rubber O-rings, and the bottom was reinforced with bathtub caulk to prevent leakage. Sampling ports, consisting of 0.95 cm diameter glass tubing installed through No. 10 rubber stoppers, were positioned as shown in Figure 30. The glass tubes extended about 5 cm into the column to facilitate collection of representative samples. When not in use the sampling ports were closed by clamping the rubber tubing fixed to the outside end of the glass tubing.

Samples were taken periodically from all sampling ports according to the following schedule:

0 hr (composite)	309 hr
118 hr	452 hr
165 hr	527 hr

All samples were analyzed for pH and ammonia nitrogen as described above.

The laboratory studies of ammonia profiles and ammonia evolved were concurrent, therefore, the sludge used was identical to that used for the determination of ammonia profiles as previously described.

Five columns of various lengths, including the 1.5 m column described above were used to simulate sludge lagoons of various depths. The columns were constructed to accommodate sludge depths of 2.5 cm, 15.2 cm, 0.3 m, 0.9 m and 1.5 m with a 15.2 cm free space for air circulation at the top of each. All columns were constructed of 20.3 cm diameter plexiglass with flanges and covers as described above. Each column was equipped with an air flow meter and inlet and outlet tubes positioned in the top access holes as shown in Figure 30. Each of the outlet tubes led to an ammonia absorption unit as illustrated in Figure 31. Vacuum flasks were used for the liquid traps, and standard 125 ml sintered glass air bubblers were used for the ammonia absorbing solution (0.124 N H_2SO_4). Vacuum was applied through a common manifold system connected to the laboratory vacuum line through a pressure regulator.

After each column was filled with fresh sludge and the tops were closed and sealed, 100.0 ml of ammonia absorbing solution (0.124 N H_2SO_4) was added to each of the air bubblers and the air flow regulated to about one l/min. This flow rate was sufficient to renew the air in the free space about every five minutes and still permit over 99 percent absorption efficiency in the gas bubblers.

Measurements of the ammonia concentration were then made at irregular intervals by titrating the absorbing sulfuric acid solution with sodium hydroxide of the appropriate normality (0.100 N or 0.050 N). After each titration fresh sulfuric acid was placed in each of the air bubblers and

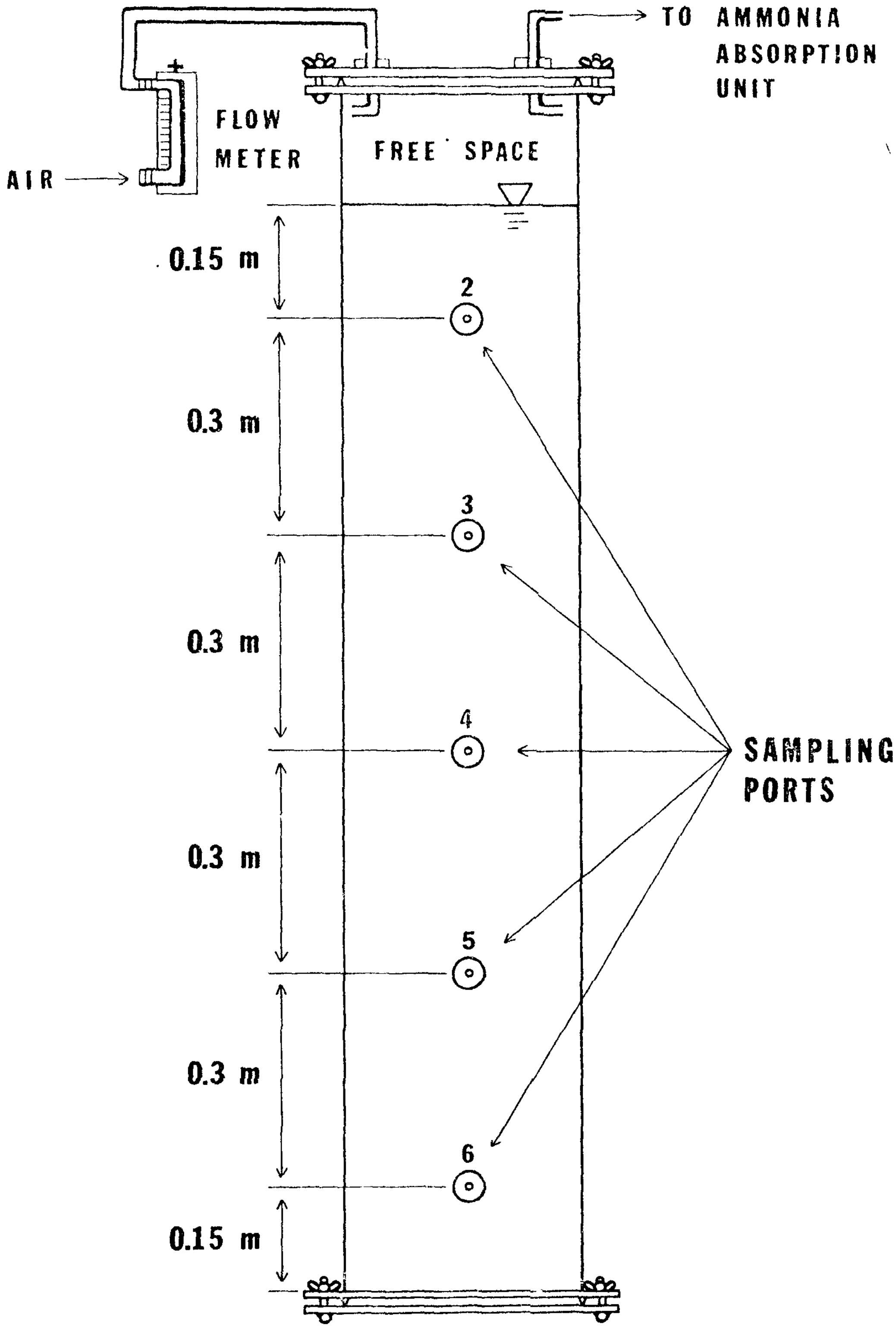


Figure 30. Diagram of apparatus used in determining NH_3 volatilization from sludge.

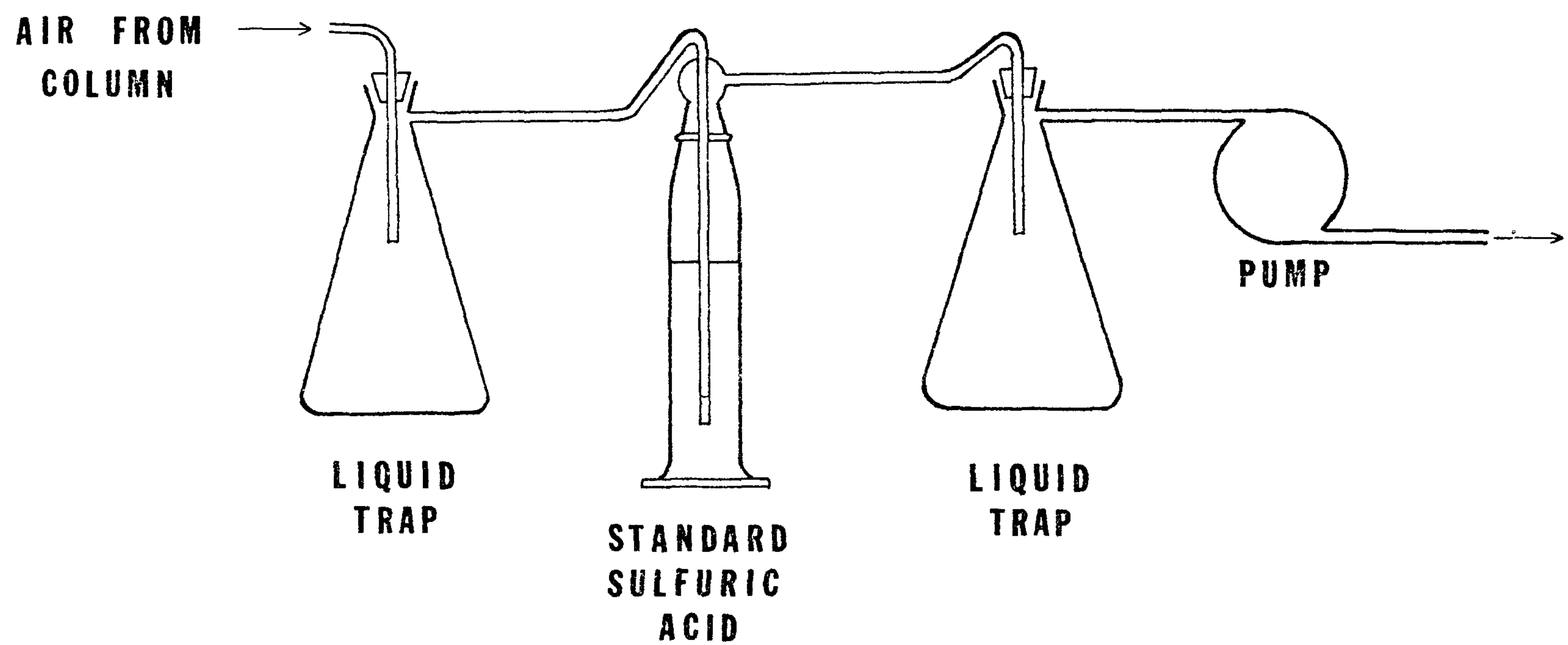


Figure 31. Schematic diagram of apparatus used to determine amount of NH_3 evolved.

the air flow restarted. Sampling times were irregular since all units could not be sampled simultaneously, however, it was attempted to make titrations according to the following schedule:

1 hr	120 hr
3 hr	144 hr
6 hr	168 hr
24 hr	216 hr
48 hr	264 hr
96 hr	312 hr

Before the experiment using sludge was begun, a determination of the ammonia content of the laboratory air was made by setting up an ammonia absorption unit without a sludge column. No appreciable ammonia was collected during a run of 96 hr duration at a flow rate of one l/min; therefore, no ammonia absorbing unit was used to scrub the air going into each column.

The method was further refined by running a statistical analysis on 12 replicate samples of a solution containing a known amount of ammonia. This analysis revealed a very small coefficient of variation (CV) of 0.43 percent.

Results of laboratory investigation - The results of the 22-day study of the ammonia profiles in liquid digested sludge are shown in Figure 32. The experimental points are encircled and curves have been approximated to fit the profiles. Note that the abscissa is only a segment of the whole scale and that the ordinate is inverted and shown as depth.

Since the experiment was run under the most natural conditions possible, it was recognized that some difficulty would be encountered with solids separation. Indeed this was the case. A definite interface between the solid and liquid portion of the sludge was observed through the clear plexiglass column. A record of the interface height during part of the laboratory run is shown in Figure 33. It can be seen that a definite equilibrium was attained with the interface height at about the middle of the 1.5 m column. This equilibrium was maintained throughout the investigation. The effect of this solid-liquid separation can be clearly seen in the profiles of Figure 32 as indicated by the definite change in curvature of the profiles at the 0.75 m level.

Note that, instead of decreasing, the concentration of ammonia N in the sludge actually increased over the period of the experiment. This implies that the rate of ammonia volatilization was exceeded by the rate of deamination under the conditions present in the laboratory run. Note also that the rate of deamination in the lower part of the column appears to have been greater than the rate of deamination in the upper part. This indicates that the sludge solids had a significant effect

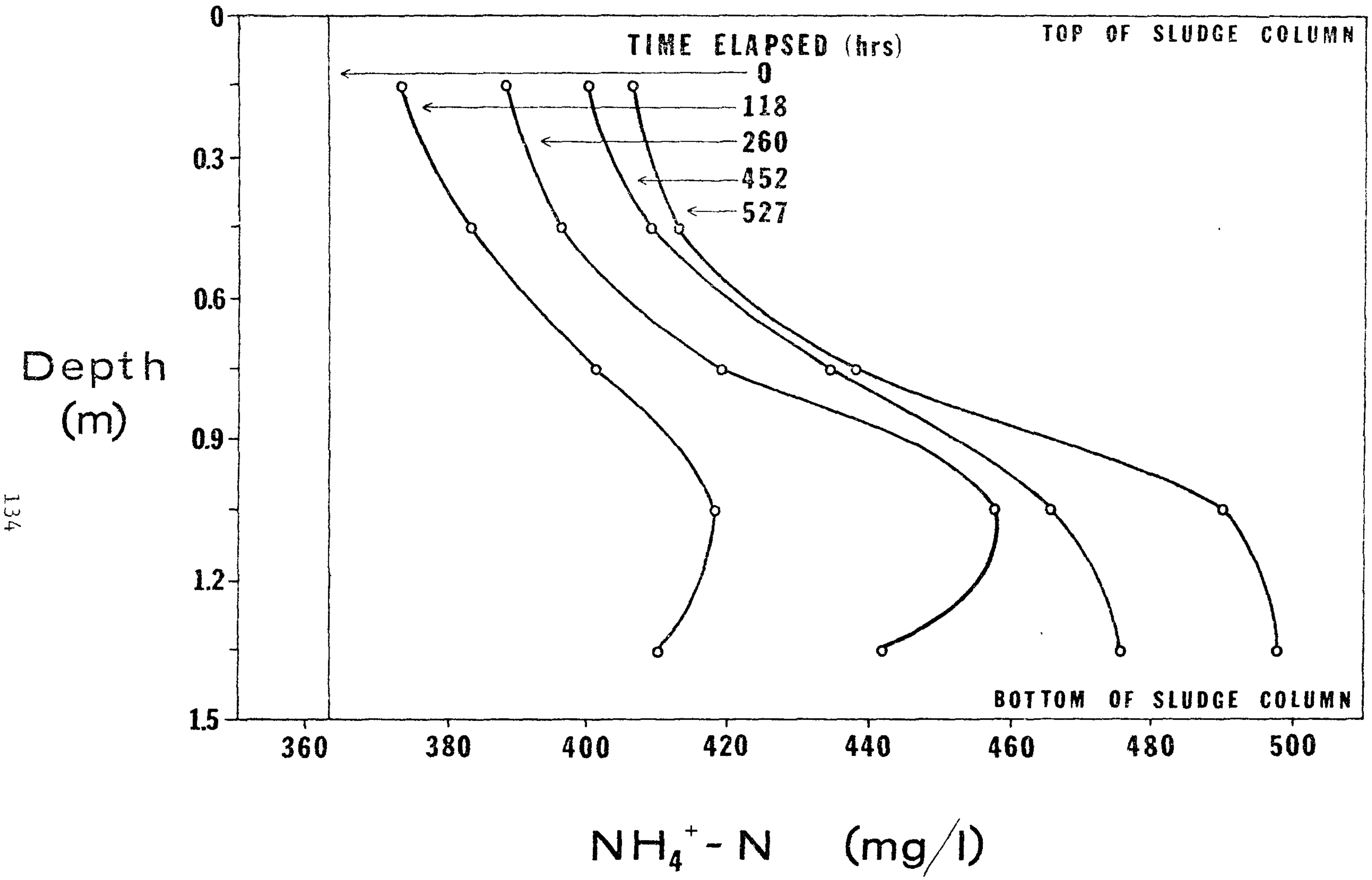


Figure 32. Distribution between 0 and 527 hours of NH_4-N with depth in an undisturbed sludge column.

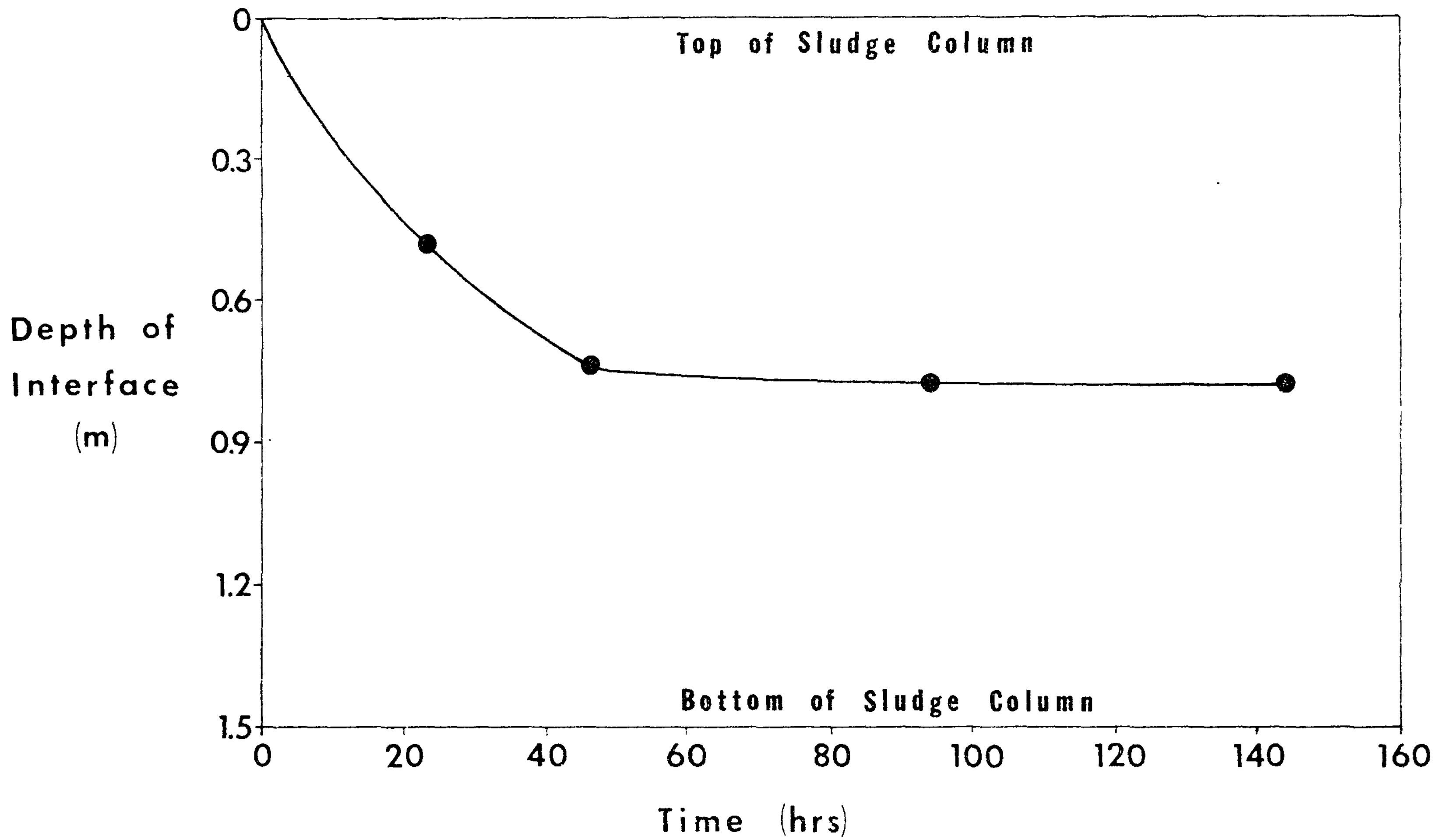


Figure 33. Depth of interface between thick suspension and supernatant as a function of time.

on the deamination process, a fact which is understandable since more ammonia can be formed where a greater organic N source is present. Thus, the deamination process, like all biological processes, depends on the quantity and availability of suitable substrate.

Ammonia volatilization curves for the five columns used in the study are shown in Figure 34. The depth of sludge is indicated by each curve. Note that the initial rate of ammonia evolution was linear with time in all cases investigated. It is obvious, however, that the three shorter columns were depleted of free, readily volatilized ammonia during the coarse of the experiment. In fact, the 2.5 cm became depleted first, then the 15.2 cm column, then the 0.3 m column as expected. Note, also, that the rate of evolution for the two longer columns (approximately 0.8 mg/hr) appears to have been significantly less than the average rate for the three shorter ones (approximately 1.0 mg/hr). This is surmised to be the result of floating scum which formed on the surfaces of both the 0.9 m and 1.5 m sludge columns.

Development of a mathematical model - In the development of the mathematical model it was necessary to make several simplifying assumptions. Because the loss of ammonia from sludge involves not only deamination, a biological process, but also diffusion of ammonia, a physical process, many assumptions were required to simplify the problem. For a description of the assumptions, derivation of the mathematical model, and the Fortran IV program used to achieve rapid analysis of the mathematical model the reader should see the thesis prepared by R. B. Gossett, Dept. of Civil Engineering, University of Illinois.

Factors affecting ammonia volatilization - The effect of mixing on the rate of mass transport and subsequent evolution of ammonia from liquid sludge was investigated by varying the effective diffusivity coefficient, D, as empirically determined for use in the mathematical model from the experimental data. Values of D used in the investigation varied from 9×10^{-4} m^2/hr, that value which best fit the laboratory data, to 6.15×10^{-3} m^2/hr, 1000 times the coefficient of molecular diffusivity of ammonia in water at 20°C (128). All of the values studied are within the realm of slow mixing and were assumed not to cause resuspension of the sludge solids.

Upon first consideration, mixing may be thought to be a major factor in the rate of ammonia evolution; however, the results of the computer run using different values of D are evidence to the contrary. Table 39 is a summary of the results showing percent total N evolved from the 1.5 m column as it varies with time and diffusivity coefficient, D. As determined experimentally the initial concentration of ammonia N, soluble organic N, and insoluble organic N were 365, 260 and 770, respectively, all expressed as mg/l of N. These values were used in comparing the results of the model with the laboratory results. The pH

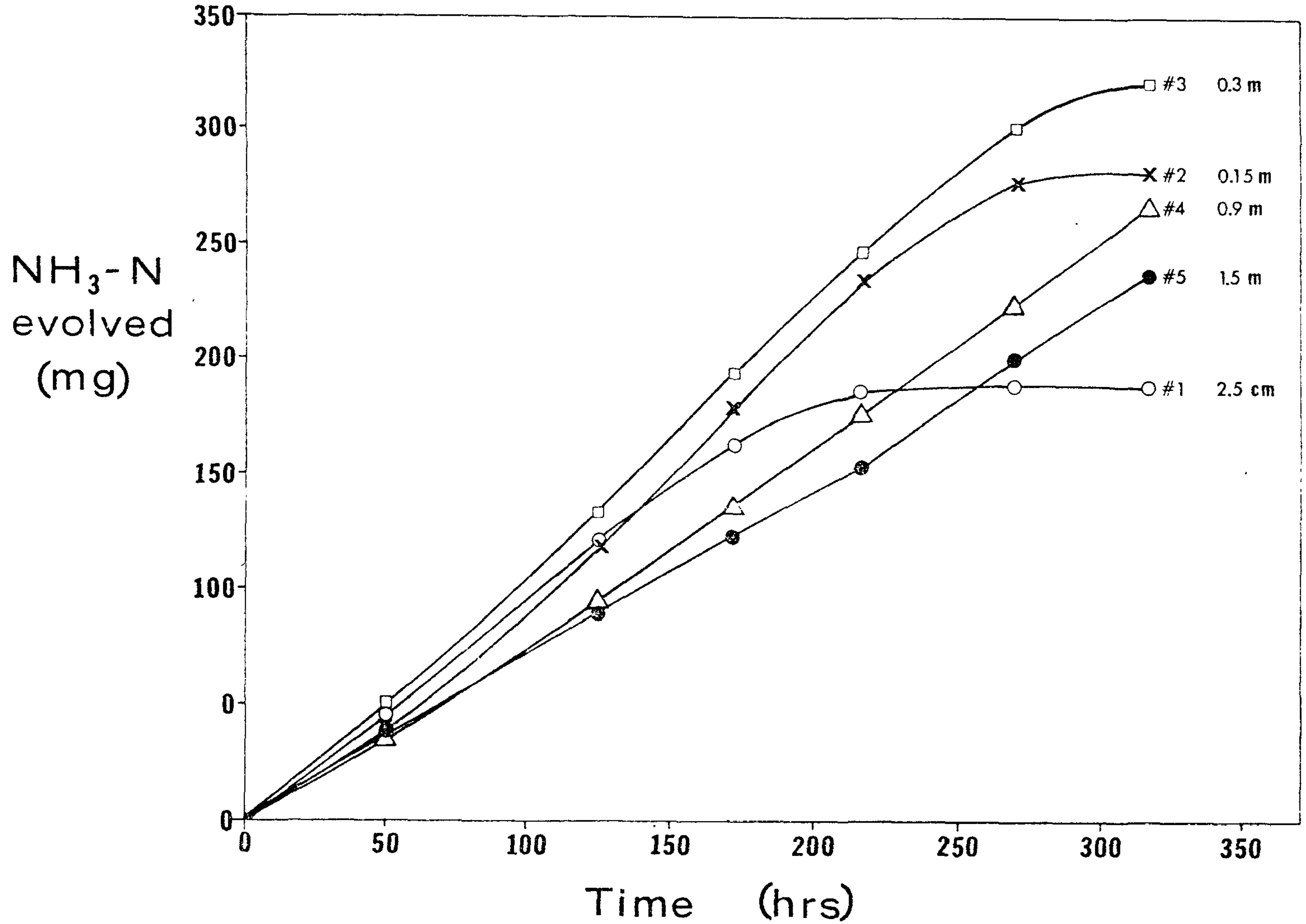


Figure 34. Volatilization of NH_3-N with time from sludge columns of different depth.

value of 7.62 and other rate constant values used in the mathematical model where those determined from a best fit of laboratory data.

Table 39. Percent total N evolved as NH_3-N from a 1.5 m sludge column by varying the effective diffusivity coefficient, D, to demonstrate the affect of mixing on ammonia evolution.

	Values of D (10^{-3} m^2/hr)				
Time (hr)	0.9	1.8	3.6	5.4	6.15
168	0.42	0.42	0.42	0.43	0.43
336	0.85	0.86	0.87	0.87	0.87
672	1.76	1.79	1.81	1.82	1.82
1344	3.73	3.81	3.86	3.88	3.88
2016	5.81	5.95	6.02	6.05	6.05
2688	7.94	8.13	8.23	8.26	8.27
3360	10.07	10.30	10.43	10.47	10.48
4032	12.16	12.44	12.59	12.64	12.65
8064	23.43	23.92	24.18	24.27	24.28

Notice that the percent total N evolved as ammonia varies only slightly with any change in D, even when a holding time of nearly a year is considered. It is recognized that sludge probably would not be stored in a lagoon for this period of time, however, a one year detention time is used to emphasize the ineffectiveness of slow mixing on ammonia loss by volatilization. It must be noted that the mathematical model developed is not meant to be applied to the study of violent or rapid mixing, therefore, modification of the model would have to be made before the effects of such phenomena could be investigated.

The effect of sludge depth on ammonia loss was studied by varying total depth in the model. Values of depth used varied from 2.5 cm to 2.4 m. The latter depth is a practical limit for sludge lagoon depth, and the former value simulated sludge applied to land with the assumption that no evaporation or percolation of liquid occurs. Although this assumption is unrealistic over long periods of time, the use of a 2.5 cm depth helps to illustrate the great effect of depth, or more appropriately surface area to sludge volume ratio, on the loss of ammonia by volatilization. Table 40 shows the effect of sludge depth on total N loss by ammonia volatilization. It can be seen that the loss of ammonia is relatively unaffected by depth from 0.15 to 2.4 m in the first 1344 hr (about 2 mo). The 2.5 cm and 7.5 cm depths differ from the rest only because of the small amounts of ammonia originally present in the sludge.

Table 40. Grams of NH_3-N evolved /m^2 as a function of sludge column depth.

	Depth of Sludge (m)						
Time (hr)	0.025	0.075	0.15	0.3	0.6	1.2	2.4
168	4.97	6.11	6.37	6.59	6.63	6.63	6.61
336	7.76	11.36	12.46	13.22	13.48	13.51	13.49
672	10.52	19.76	23.73	26.49	27.68	27.99	27.98
1344	12.78	30.88	42.77	52.09	57.13	59.10	59.34
2016	13.99	37.49	57.50	75.49	86.62	91.72	92.79
2688	14.79	41.58	68.60	96.14	115.11	124.78	127.33
3360	15.31	44.18	76.83	113.89	141.89	157.44	162.33
4032	15.67	45.87	82.83	129.00	166.78	189.22	197.22
8064	16.31	48.87	95.86	177.89	275.78	354.67	395.78

Although total N loss (gm NH_3-N/m^2) increases with depth, it is very interesting to note the effect of expressing the results as a percent of total N originally present in the column of sludge. Table 41 demonstrates this point. Notice that the greatest percent loss, as expected, occurs early in the lower depths, showing that mass transport of ammonia through the bulk liquid is the predominant factor in controlling the rate of ammonia loss under the given conditions.

The hydrogen ion concentration (expressed as pH, the negative logarithm of the hydrogen ion concentration) effects the equilibrium between ammonia, NH_3, and ammonium ion, NH_4^+, in solution. If the theoretical equation for ammonia equilibrium in water is considered:

$$NH_3 + H^+ \rightleftharpoons NH_4^+ \quad ,$$

it can be seen that an increase in the hydrogen ion concentration (a decrease in pH) will force the equilibrium to the right, leaving less NH_3

Table 41. Percent total nitrogen evolved as NH_3-N as a function of sludge column depth.

	Depth of Sludge (m)						
Time (hr)	0.025	0.075	0.15	0.3	0.6	1.2	2.4
168	18.8	7.7	4.0	2.1	1.0	0.5	0.3
336	29.3	14.3	7.8	4.2	2.1	1.1	0.5
672	39.7	24.9	14.9	8.3	4.4	2.2	1.1
1344	48.3	38.9	26.9	16.4	9.0	4.7	2.3
2016	52.9	47.2	36.2	23.8	13.6	7.2	3.7
2688	55.9	52.3	43.2	30.3	18.1	9.8	5.0
3360	57.8	55.6	48.4	35.9	22.3	12.4	6.4
4032	59.2	57.7	52.1	40.6	26.3	14.9	7.8
8064	61.6	61.5	60.3	56.0	43.4	27.9	15.6

and forming more NH_4^+. On the other hand, a decrease in the hydrogen ion concentration (increase in pH) will have the opposite effect, resulting in an increase in NH_3. Because the rate of volatilization of NH_3 depends on the magnitude of NH_3 concentration in the surface film, an increase in pH will tend to increase the rate of ammonia volatilization by making more NH_3 available at the surface film.

In developing the mathematical model it was assumed that NH_3 and NH_4 diffuse to the surface film at the same rate. If, however, NH_3 diffuses more rapidly or a gross increase in pH causes ammonia gas formation in the bulk liquid, the rate of ammonia volatilization would be further increased by an increase in pH.

In this study pH was varied from 6.0 to 11.0, values which were assumed to be practical and economically feasible limits for lagooned sludges with pH controlled. Some sludges may naturally have a pH below 7.0 but probably not below 6.0. On the other extreme, a pH of 11.0 can practically be achieved by lime addition. Table 42 shows the computer results of the pH study.

It can be seen from Table 42 that an increase in pH to at least 9.0 can be very beneficial in increasing the ammonia-volatilization rate. Increasing the pH to higher values yields diminishing returns, therefore, the economics of base addition must be examined more closely if raising the pH to higher values is considered.

Table 42. Percent total nitrogen evolved from a 1.5 m column of sludge at different pH values.

	pH					
Time (hr)	6.00	7.00	8.00	9.00	10.00	11.00
168	0.01	0.10	0.93	4.29	6.83	7.23
336	0.02	0.21	1.86	7.70	11.35	11.86
672	0.05	0.45	3.78	13.71	18.79	19.44
1344	0.10	0.97	7.77	24.12	30.64	31.41
2016	0.16	1.54	11.81	32.67	39.50	40.24
2688	0.22	2.14	15.76	39.51	46.00	46.65
3360	0.29	2.76	19.54	44.86	50.70	51.24
4032	0.35	3.38	23.11	48.97	54.05	54.49
8064	0.76	7.10	39.55	59.16	60.98	61.09

Since the concentrations of ammonia and organic N in the digested sludge obtained from the Urbana Treatment Plant may be somewhat lower than those from sewage treatment plants receiving more industrial waste, the effect of higher N content (NH_4^+-N = 800 mg/l, soluble org-N = 350 mg/l and insoluble org-N = 1050 mg/l) was investigated. This eliminated the possibility of biased results based on only one set of N concentrations. Table 43 shows a comparison using the constants which fit the laboratory data. It is apparent from Table 43 that a higher initial N content in the liquid sludge promotes the loss of ammonia by volatilization. This effect is surmised to be the result of a larger driving force in the deamination process, resulting in increased NH_4^+-N concentration gradients and, thus, in a higher rate of NH_4^+-N diffusion rate within the liquid sludge. The data in Table 43 indicates, however, that the relative effect of higher initial N concentrations on the total N evolved by ammonia volatilizacion is minimal over a long period of time.

Table 43. Percent total nitrogen evolved from a 1.5 m column of sludge at two different initial nitrogen concentrations.

	Initial Nitrogen Concentrations	
Time (hr)	Low Nitrogen*	High Nitrogen**
168	0.42	0.54
336	0.85	1.10
672	1.76	2.23
1344	3.73	4.58
2016	5.81	6.99
2688	7.94	9.41
3360	10.07	11.79
4032	12.16	14.12
8064	23.43	26.53

* NH_4^+-N = 365 mg/l, soluble org.-N = 260 mg/l, and insoluble org.-N = 770 mg/l.

** NH_4^+-N = 800 mg/l, soluble org.-N = 350 mg/l, and insoluble org.-N = 1050 mg/l.

Conclusions - The following conclusions can be made based on the results of this study:

1. Under the laboratory conditions encountered in this investigation (pH 7.6, room temperature of 26.4°C, depth of 1.5 m, and without artificial mixing), the rate of deamination in liquid digested sludge exceeded the rate of ammonia volatilization resulting in an ammonia N accumulation in the sludge even though the total N content decreased slightly.

2. The rate of ammonia volatilization from liquid digested sewage sludge was determined under laboratory conditions and a mathematical model was derived to describe N loss by ammonia volatilization as it varies with mixing, depth of sludge, pH, and initial concentrations of ammonia and organic N. Using the mathematical model developed in this investigation, along with its inherent assumptions, it was determined that:

a. Mixing of the sludge, within the narrow range of effective diffusivity coefficients studied, had little effect on the rate of ammonia loss by volatilization. This

a. (cont) conclusion cannot be generalized to include violent mixing, i.e. surface renewal and surface area increase, since it was decided that the mathematical model developed could not be used to apply to these specialized and extreme cases. The purpose of this study was not to investigate ammonia stripping, but to determine incidental ammonia volatilization prior to and during liquid digested sludge application on land.

b. The N lost by ammonia volatilization per unit surface area varied less than expected as the depth of sludge was varied from 2.5 cm to 2.4 m, however, the effect of sludge depth on the percent of the total N initially present in the sludge which was volatilized as ammonia over a given period of time is quite pronounced. As expected, the greatest percent N loss occurred when the surface area to volume ratio was greatest, i.e. at the smallest sludge depth.

c. The one parameter that most affects the rate of ammonia volatilization from liquid digested sewage sludge is pH. Increasing the pH of the sludge from 7.0 to 9.0 increased the amount of ammonia lost (as percent of total N initially present) by a factor of 40 for a 168 hr (7 da) holding time, and by a factor of 8 for an 8064 hr (336 da) holding time. Increasing the pH incrementally above 9.0 resulted in diminishing increases in the amount of ammonia evolved.

d. Under all conditions studied, except when the pH was increased to 9.0 and above, an ammonia accumulation took place in the sludge.

e. The effect of initial concentrations of ammonia and organic N (within reasonable limits that might be found in liquid digested sewage sludges) on the percentage of N lost by ammonia volatilization was negligible.

Stability Constants of Metal-Polyelectrolyte Complexes Occurring Naturally in Soils and Sewage Sludge

Introduction - Many investigators have shown that weak acid polyelectrolytes occurring naturally in soils, sediments, organic wastes, and river and drainage waters form complexes with metal ions of the transition series. These natural polyelectrolytes, often referred to as humic substances, are involved in the weathering of rocks and minerals, and they are believed to be responsible for the migration and enrichment of mineral substances in sedimentary rocks and certain mineral deposits. The trace elements found in municipal wastes and soils may occur largely in association with complex organic polyanions. Furthermore, the availability of micronutrients to plants and microorganisms is greatly affected by complexation reactions.

The solid component of digested sewage sludge consists of a mixture of organic matter and mineral material in about equal proportions. The heavy metals in liquid digested sewage sludge appear to be strongly bound to the organic matter. This interaction between heavy metals and humic-like polymeric substances may have a profound effect on the mobility and toxicity of metal ions when sewage sludge is applied to agricultural soils.

Complex formation between a chelating agent and a metal ion alters the properties of the metal ion, including solubility, oxidation state, and free-energy level of the half reaction. In biological systems, nutrient uptake and energy transformations are controlled by the electron free-energy, E, of the oxidation-reduction reaction, which is given by the Nernst equation:

$$E = E^{o} - \frac{kT}{n} \ln \frac{[R]}{[0]} \quad (1)$$

where E^{o} is the standard free-energy and [R] and [0] are concentrations of the reduced and the oxidized forms of the element. In the presence of a complexing agent, the electron free-energy level of the reaction is changed; thus, nutrient uptake of microbial processes are modified.

The main thermodynamic characteristic of a metal-organic matter complex is its stability constant, K, which is related to the change in free-energy, ΔF, accompanying complex formation.

$$\Delta F = -RT \ln K \quad (2)$$

Stability constants would be most useful in predicting solubility and movement of metal ions in sludge-treated soils and their availability to plants and microorganisms.

Several attempts have been made to determine stability constants of soil organic matter-metal complexes, but the accuracy of these measurements in soils is suspect. No work seems to have been done with sewage sludge.

The naturally occurring polyanions used in this study were from a number of sources, including soils, a North Dakota lignite, and digested sewage sludge. Because the soil humic acids were available in significant quantities the work involving methodology was done with the soil materials rather than with sewage sludge. Along this line, it should be emphasized that the main objective of the present study was to develop methods for determining stability constants which might ultimately be applied to sludge-treated soils.

Special attention was given to the ion exchange equilibrium and potentiometric titration methods. A modified mathematical model based on Schubert's ion exchange technique was developed which was free of certain errors and assumptions made by previous workers. The method was tested and applied for measuring stability constants of complexes between Zn^{+2}, Cu^{+2} and Mn^{+2} and soil humic and fulvic acids, as well as similar type substances from sewage sludge.

Materials and methods - The sludge, obtained from the Calumet treatment plant of the Metropolitan Sanitary District of Greater Chicago, is a black slurry usually having a solid content of 2 to 4 percent by weight. About half of the solids are organic materials, the remainder consisting of mineral elements including heavy metals which are strongly combined with the organic components.

Two approaches were used to extract humic-like substances from the sludge: (1) elimination of metal ions from the solids with buffered citric acid to leave organic constituents behind, and (2) extraction of humic-like substances with alkaline solutions.

The procedure of Jenkins and Cooper (76) with slight modification was applied to extract strongly bound metal ions from the sludge. In a typical experiment, one g sample of untreated, freeze-dried sludge, or sludge pretreated with 0.3 N HF:0.1 N HCl, was suspended in 75 ml of 2 percent citric acid-ammonium citrate buffer at pH values of 2.0, 3.0 and 4.0 and the mixtures shaken for two hours. Each sample was then centrifuged and the supernatant analyzed for Ni^{+2}, Pb^{+2}, Mn^{+2}, and Cr^{+2}.

Results given in Figures 35, 36, 37, and 38 show that, except for Pb^{+2}, from 2 to 5 successive treatments reduced the concentrations of metal ions in the extracts to near zero ppm. With the exception of Pb^{+2}, pH had little effect on the amount released. Extracts from the HF;HCl-treated sludge were considerable lower in metal ions.

The concentration of Ni^{+2} in the extracts of pH 2.0 was slightly higher than at pH 3.0 and 4.0, as can be seen by inspection of Figures 35 and 39. With Pb^{+2}, however, a slight increase was observed at pH 4.0, with little difference between buffers of pH 2.0 and 3.0 (Figures 36 and 39). Removal of Mn^{+2} followed a trend similar to Ni^{+2} and Pb^{+2} (Figure 37). Comparison of the concentration of Mn^{+2} in the first extract shows that amount removed decreased with an increase in pH. The extraction of Cr^{+2} (Figure 38) was practically complete with a single treatment, with pH having little effect on removal.

Comparison of the results with those of Jenkins and Cooper (76) revealed that shaking the sludge with acid solution was more effective in extracting metal ions than successive percolation. Fewer than five extractions removed most of the soluble metal ions while 16 treatments were required with the percolation method. Jenkins and Cooper (76) reported that no more than 57% of Cu^{+2}, 73% of Ni^{+2} and 75% of Zn^{+2} could be removed by buffered acids.

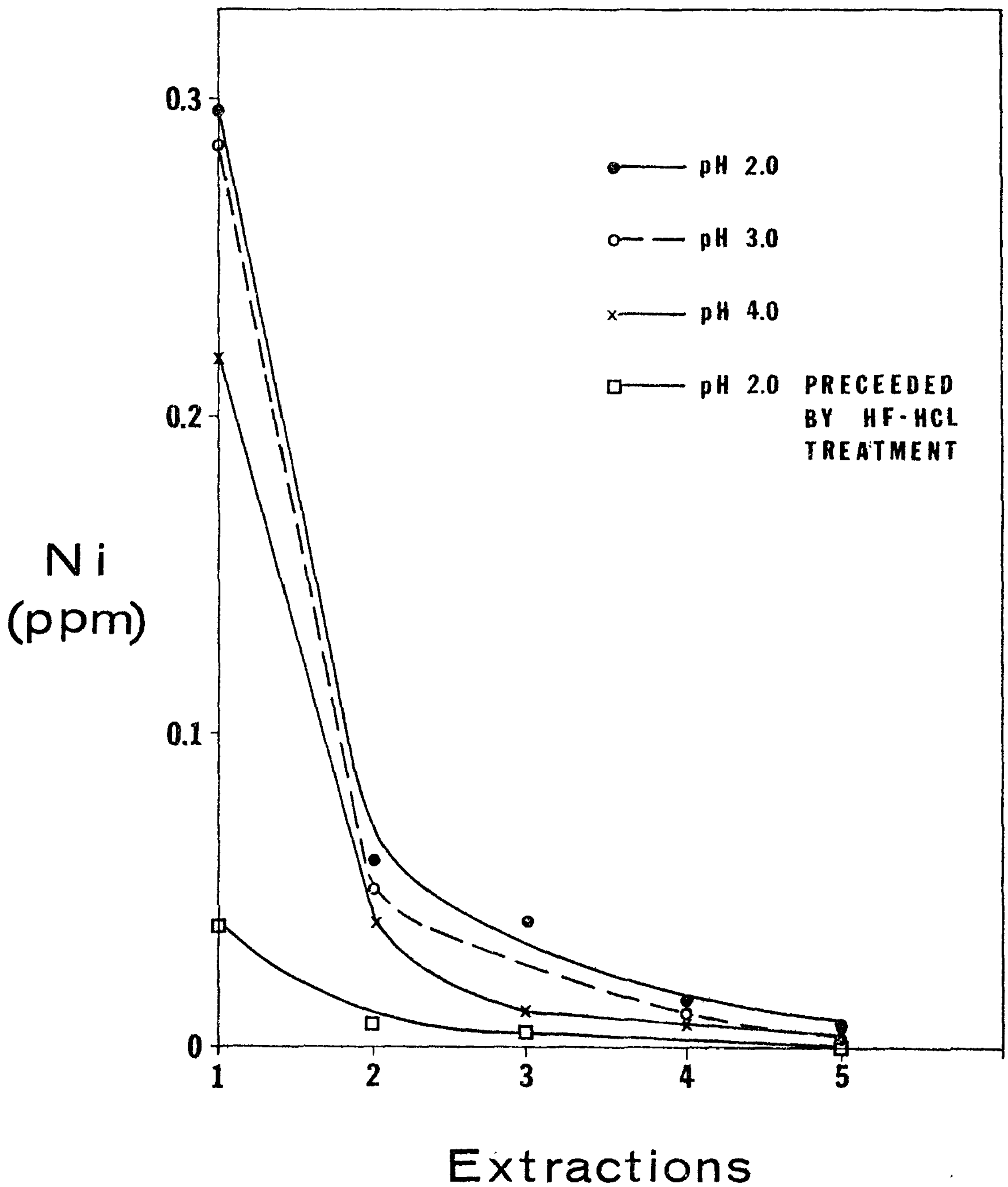


Figure 35. Concentration of Ni^{+2} in successive buffered citric acid extracts of sludge solids. A one-gram sample of freeze-dried sludge solids was extracted with 75 ml of the acid solutions.

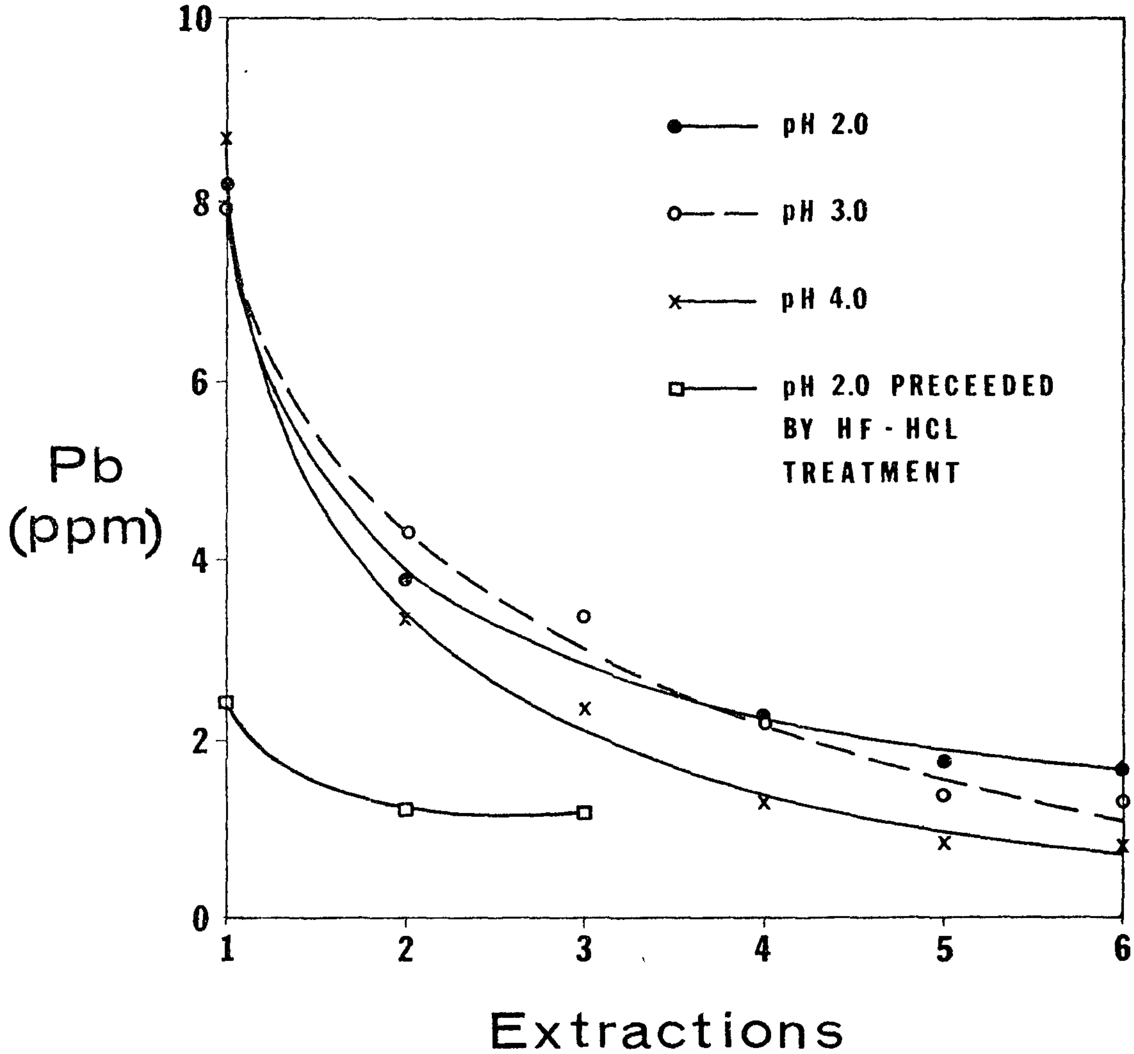


Figure 36. Concentration of Pb^{+2} in successive buffered citric acid extracts of sludge solids. A one-gram sample of freeze-dried sludge solids was extracted with 75 ml of the acid solutions.

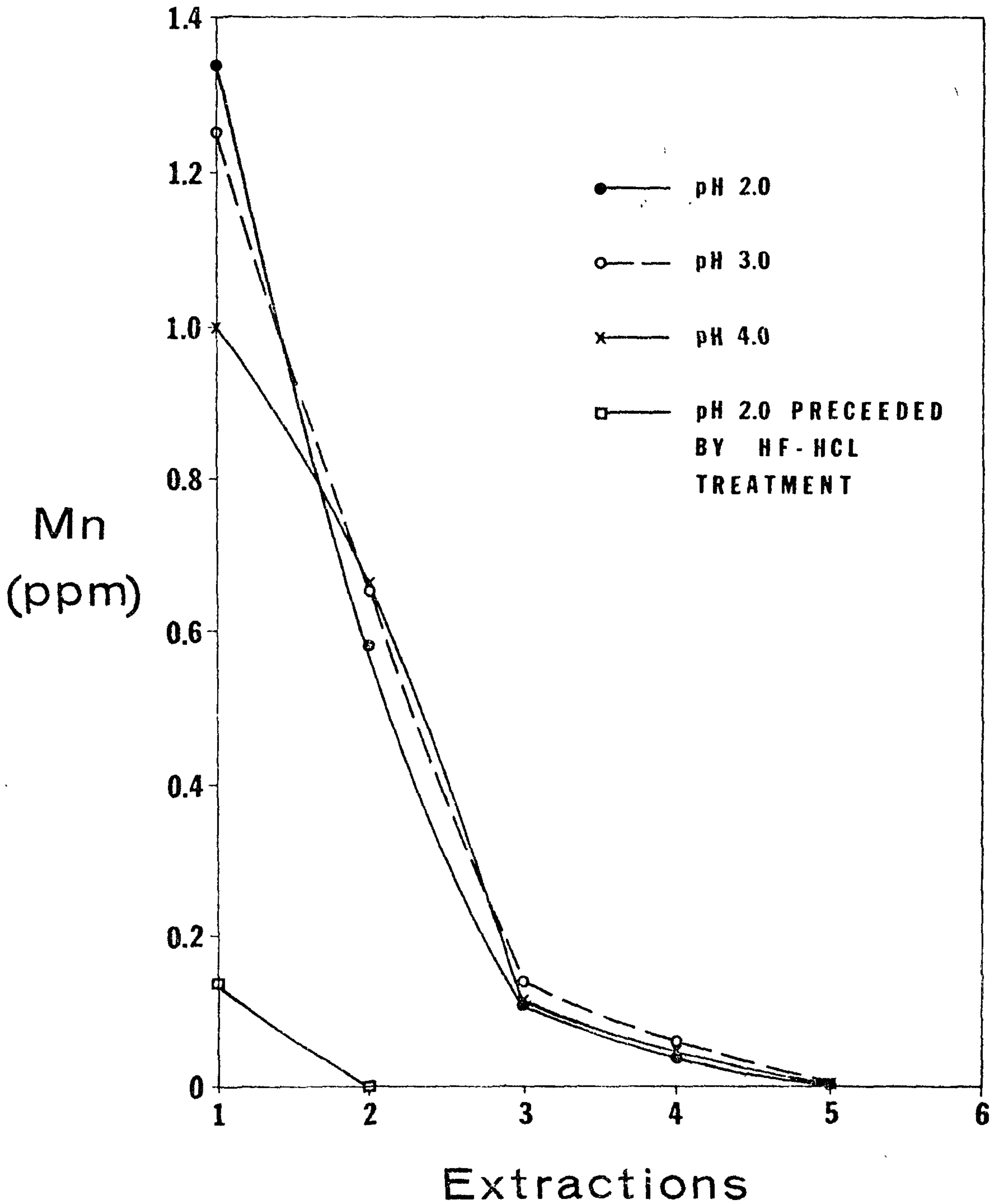


Figure 37. Concentration of Mn^{+2} in successive buffered citric acid extracts of sludge solids. A one-gram sample of freeze-dried sludge solids was extracted with 75 ml of the acid solutions.

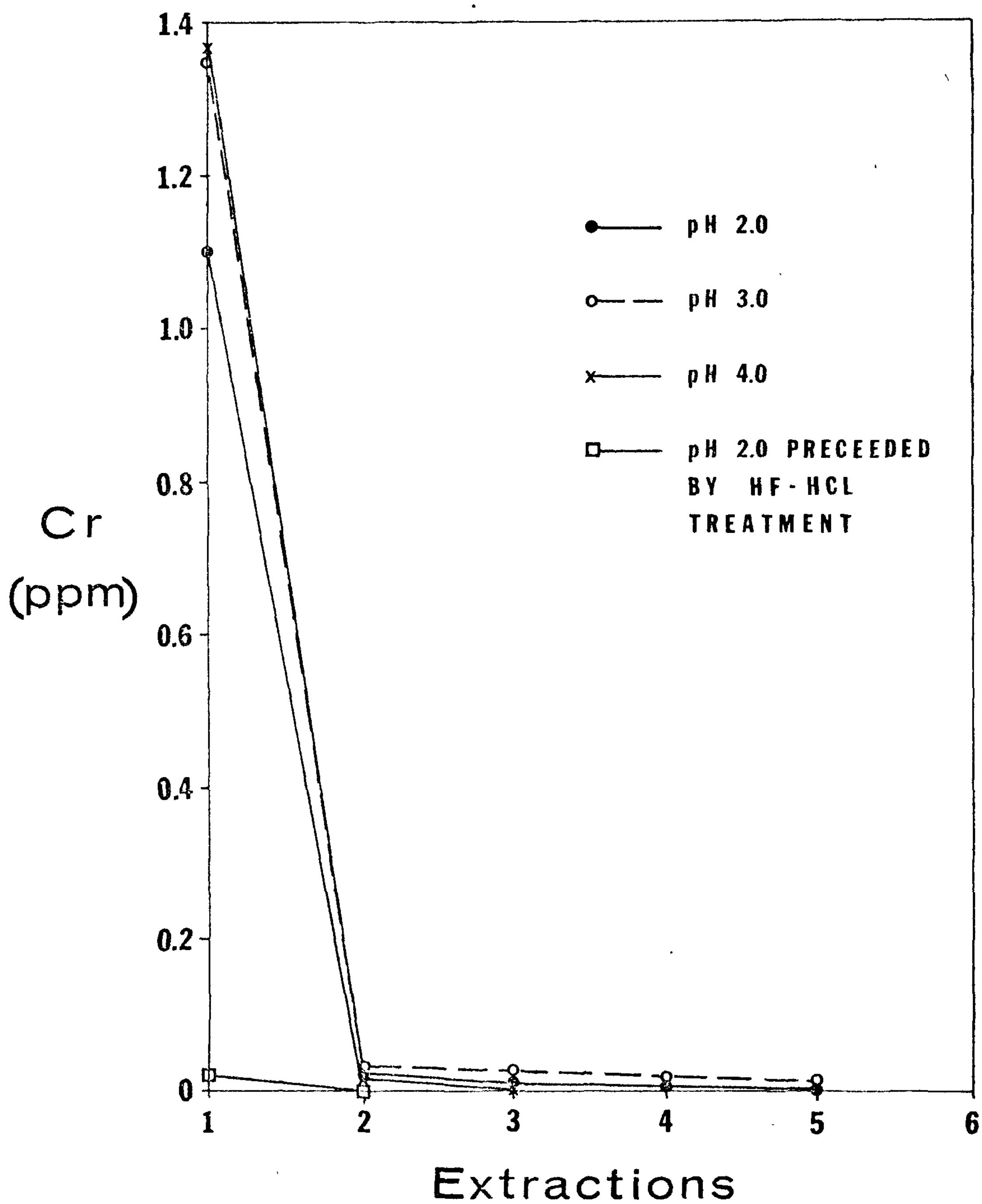


Figure 38. Concentration of Cr^{+2} in successive buffered citric acid extracts of sludge solids. A one-gram sample of freeze-dried sludge solids was extracted with 75 ml of the acid solutions.

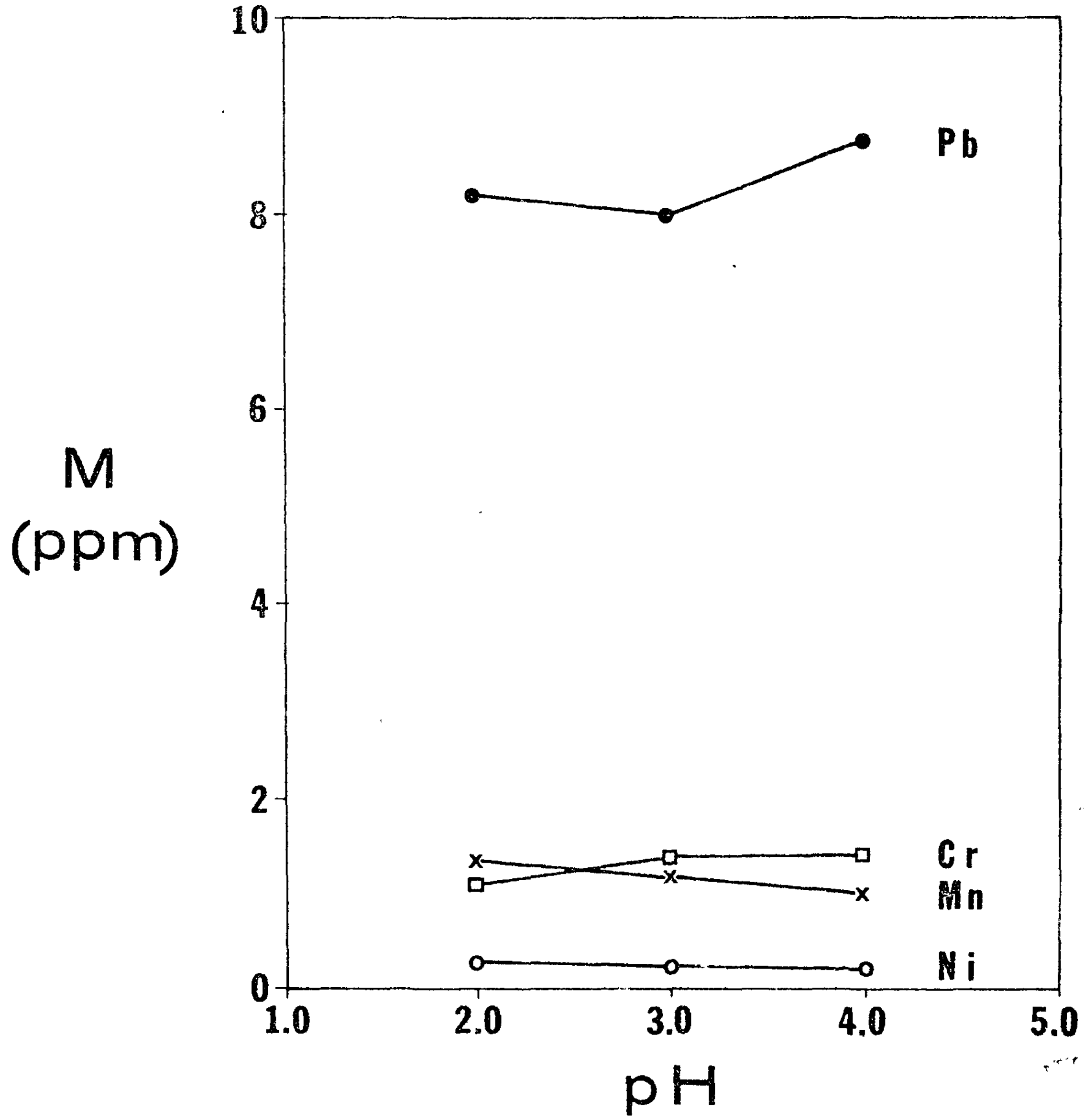


Figure 39. Effect of pH on total amounts of metal ions extracted from one-gram sample of sludge solids with five successive 75-ml treatments.

Butler (29) found that successive treatment of crude soil humic acids with 0.3 N HF:0.1 N HCl was effective in removing mineral matter. It seemed reasonable to subject sludge solids to such treatment for eliminating metal ions. Table 44 shows that the ash content of a sludge sample was reduced from 50 percent to 11 percent by 30 successive extractions with the HF:HCl solution. No further reduction was obtained by 10 successive treatments with 6 N HCl. Therefore, it was concluded that complete removal of inorganic components from the sludge solids was impractical by treatment with mineral acids.

Table 44. Elimination of mineral matter from sludge solids using mineral acid solutions.

No. of successive treatments	Reagent	% Ash
0	-	59.0
6	0.3 N HF:0.1 N HCl	19.09
17	0.3 N HF:0.1 N HCl	17.2
30	0.3 N HF:0.1 N HCl	11.0
40	6 N HCl[a]	11.0

[a] First 30 times by 0.3 N HF:0.1 N HCl followed by 10 treatments with 6 N HCl

Humic and fulvic acids are normally recovered from soil by extraction with caustic alkali solutions or $Na_4P_2O_7$ (52)(88)(135) and it seemed reasonable to assume that these reagents could also be used to dissolve the complex polyanions associated with sludge solids. Accordingly, one-gram portions of freeze-dried, powdered sludge were suspended in 75 ml of 0.05, 0.10, 0.25, 0.5 N NaOH solution and in 0.15 N $Na_4P_2O_7$ solution. The mixtures were shaken for one hour after which the insoluble residues were removed by centrifugation and a portion of the supernatant liquids were analyzed for carbon by Mebius' modification of Tinsley procedure (103). A second portion was used for the determination of ash content. In this case, 0.1 N HCl was added to precipitate the humic acids whose ash content was determined by combustion at 550°C for 18 hours.

The results, given in Table 45, show that the percent organic matter extracted increased with concentration of NaOH (from 27.3 percent to 38.0 percent). A slightly lesser amount was extracted with $Na_4P_2O_7$ (22.7 percent).

Table 45. Extraction of organic materials from sludge solids with alkaline solution.

Base	Concentration $\underline{N}$	Organic matter extracted	Ash
		- - - - - - - - % - - - - - - - -	
$Na_4P_2O_7$	0.15	22.7	9.52
NaOH	0.05	27.3	7.09
NaOH	0.10	29.5	6.67
NaOH	0.25	31.7	6.51
NaOH	0.50	38.0	6.59

Inspection of Table 45 shows that sludge humic acids obtained by extraction had rather high ash contents (6.5 to 9.5 percent). Accordingly, a combination of acid and base treatment was applied. In addition, organic solvents were employed for removal of rats, waxes, and resins. The fractionation scheme adopted for recovery of humic acids from the original sewage sludge suspension is given in the flow diagram shown in Figure 40.

In a typical experiment, 200 ml digested sewage sludge was allowed to settle. The solids (about 6 g) were recovered by centrifugation and shaken three times with 250 ml 0.3 $\underline{N}$ HF:0.1 $\underline{N}$ HCl and centrifuged. The residue was suspended in 250 ml of 0.15 $\underline{N}$ $Na_4P_2O_7$, after which the soluble portion was recovered by centrifugation and acidified with 0.1 $\underline{N}$ HCl. The humic acid precipitate was recovered by centrifugation and freeze-dried. Then fats, waxes, and resins were removed by successive extraction with ether, benzene, and chloroform (24 hours for each solvent). The residual material was then shaken with 75 ml of 0.1 $\underline{N}$ HCl and insoluble residue was recovered by centrifugation and was called "crude humic acid". Only 15 percent of the "crude humic acid" was soluble in 0.15 $\underline{N}$ $Na_4P_2O_7$, the rest being soluble in increasing concentrations of NaOH (0.1, 0.25, 0.5, 1.0, and 2.0 $\underline{N}$). Each solution was acidified, centrifuged, and the precipitate washed with the HF:HCl solution as discussed before. The humic acid soluble in 0.15 $\underline{N}$ $Na_4P_2O_7$ contained 1.4 percent mineral matter.

Removal of mineral matter from "crude humic acid" was also accomplished using EDTA.

Sludge humic acids varied in color from brownish and greenish black to black. Except for the $Na_4P_2O_7$-extracted preparation, they were only slightly soluble in solutions of pH below 10, the EDTA-extracted fraction being the least soluble. Their potentiometric titration curves

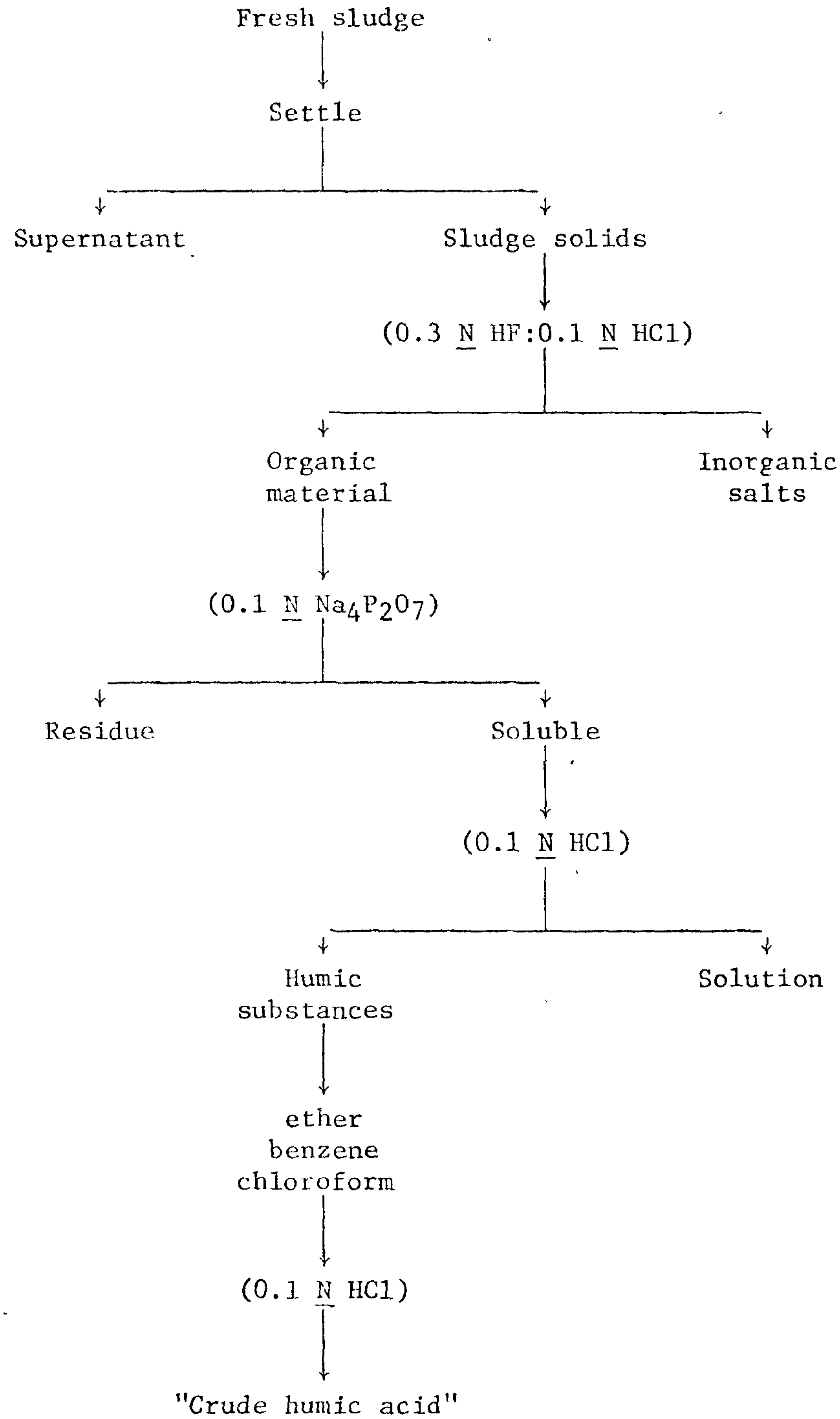


Figure 40. Scheme for Extracting Humic-Like Materials from Sewage Sludge.

(Figures 41, 42 and 43), like those of humic acids from other source materials, showed a single inflection point. Acid dissociation constants calculated from these curves varied from 3.75 to 5.25, somewhat in the same range as values for other humic acids (Table 46). These values are typical of those shown for carboxyl groups, which normally lie between 4 and 5 (152). Titratable acidity of the sludge humic substances, as reported in Table 46, were somewhat lower than those of humic and fulvic acids. However, the $Na_4P_2O_7$-fraction had a total acidity close to that of the soil humic acids of 330 meq/100 g. This may explain the higher solubility of the $Na_4P_2O_7$-soluble humic acid of sludge as compared with the alkali-soluble fractions.

Table 46. The titratable acidity and pK_a in 0.1 N KCl of some naturally occurring polyelectrolytes in soils and digested sewage sludge.

Humic acid	Titratable acidity	pK_a
	- - meq/100 g - -	
Brunizem	378	4.10
Peat	375	4.25
Leonardite	392	4.20
Synthetic*	410	3.55
Sludge (0.15 N $Na_4P_2O_7$)	330	4.10
Sludge (0.10 N NaOH)	119	5.25
Sludge (EDTA)	191	3.75

* a glucose-glycine condensation product.

The sludge humic acids were freeze-dried, ground and their infrared (IR) spectra prepared by KBr pellets following the procedure described by Goh (55). The IR spectra of the various humic acids given in Figure 44 showed only slight variations and were characterized by main absorption bands at 3300, 2910, 1650, and 1530 and a minor one at 1220 cm^{-1}. A sharp absorption band at 2110 cm^{-1} was displayed by $Na_4P_2O_7$-extracted preparation which was removed by hydrolysis with 6 N HCl (Figure 45, Trace A). This band was attributed to C≡N stretching of cyanide. The fraction treated with EDTA showed a sharp absorption band at 2910 and a strong one at 1030 cm^{-1} which suggests that this fraction may contain carbohydrates.

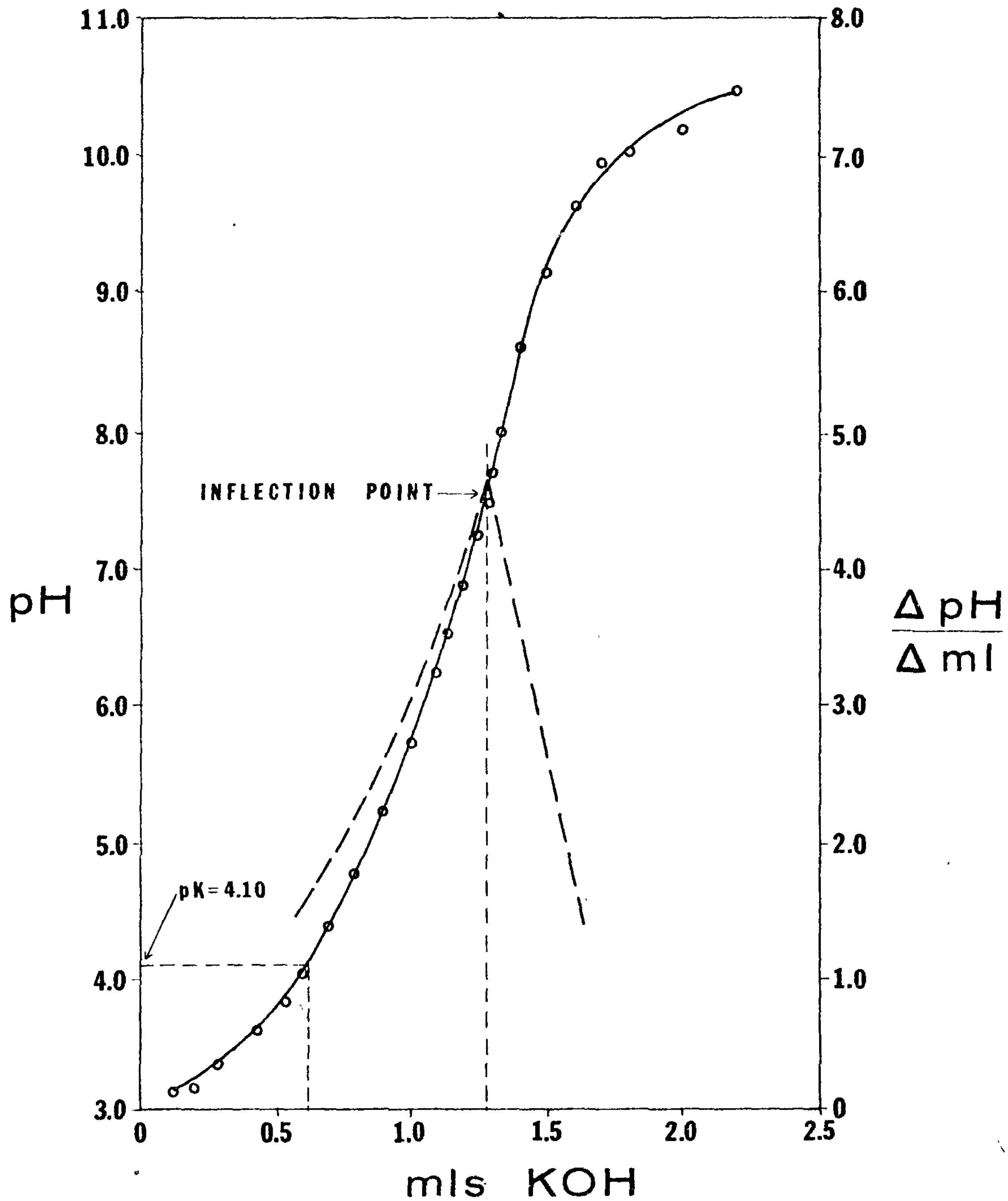


Figure 41. Titration curve in 0.1 N KCl of 25 mg of humic acid obtained by $Na_4P_2O_7$ extraction of sludge. Titration carried out using 0.085 N KOH, standard calomel and glass electrodes.

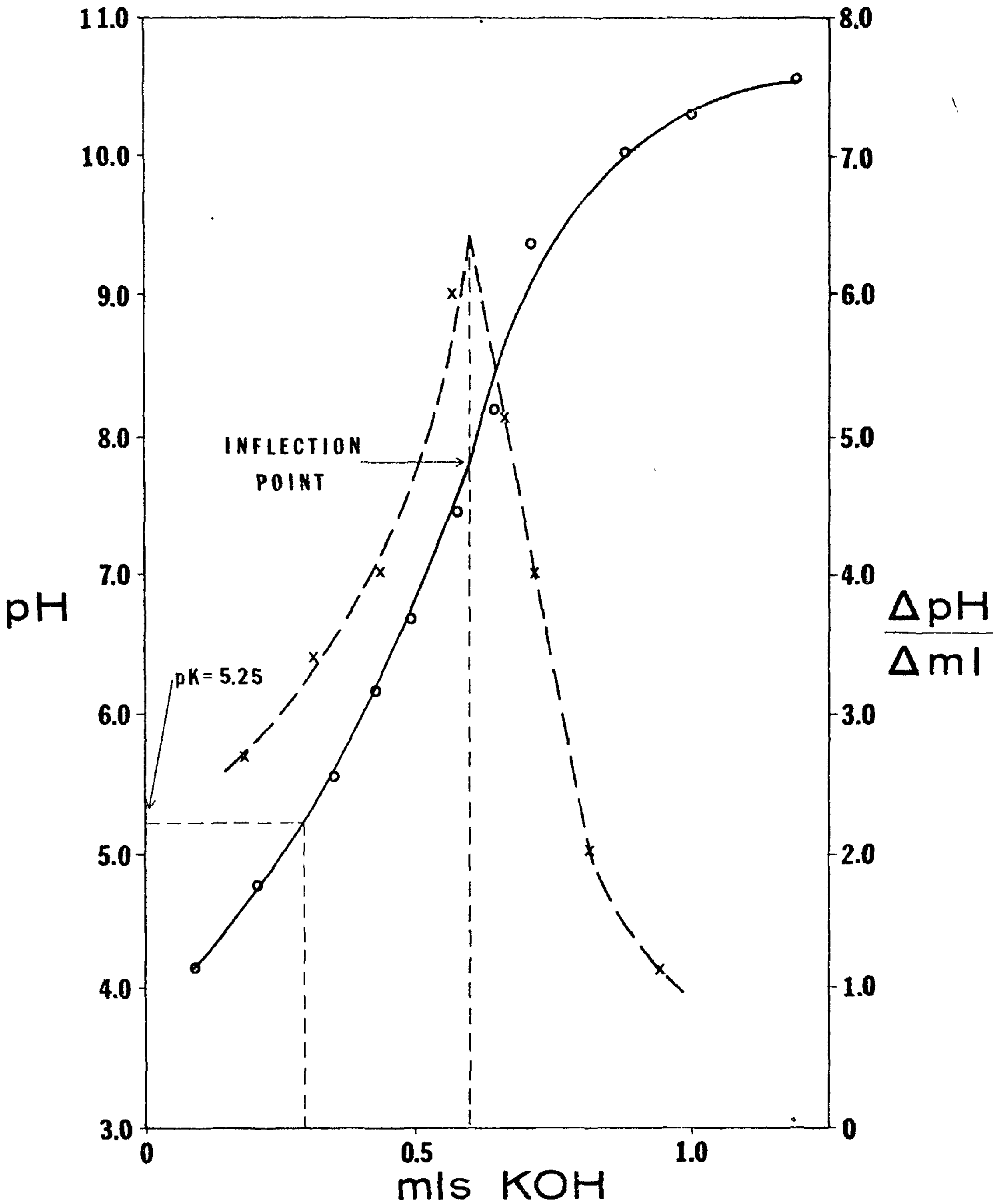


Figure 42. Titration curve in 0.1 N KCl of 25 mg of humic acid obtained by NaOH extraction of sludge. Titration carried out using 0.085 N KOH, standard calomel and glass electrodes.

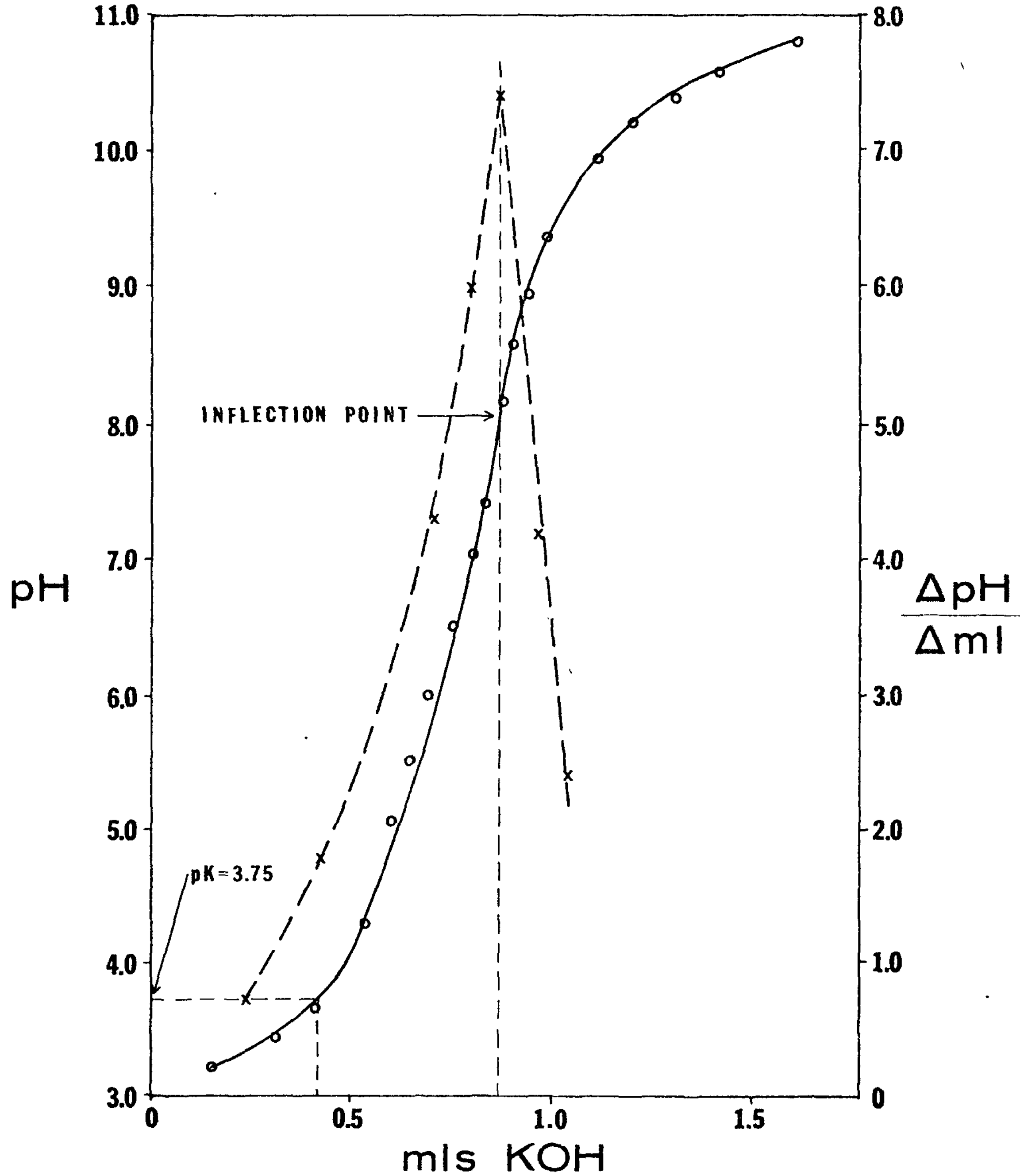


Figure 43. Titration in 0.1 N KCl of 25 mg of humic acid extracted from sludge with EDTA. Titration carried out using 0.085 N KOH, standard calomel and glass electrodes.

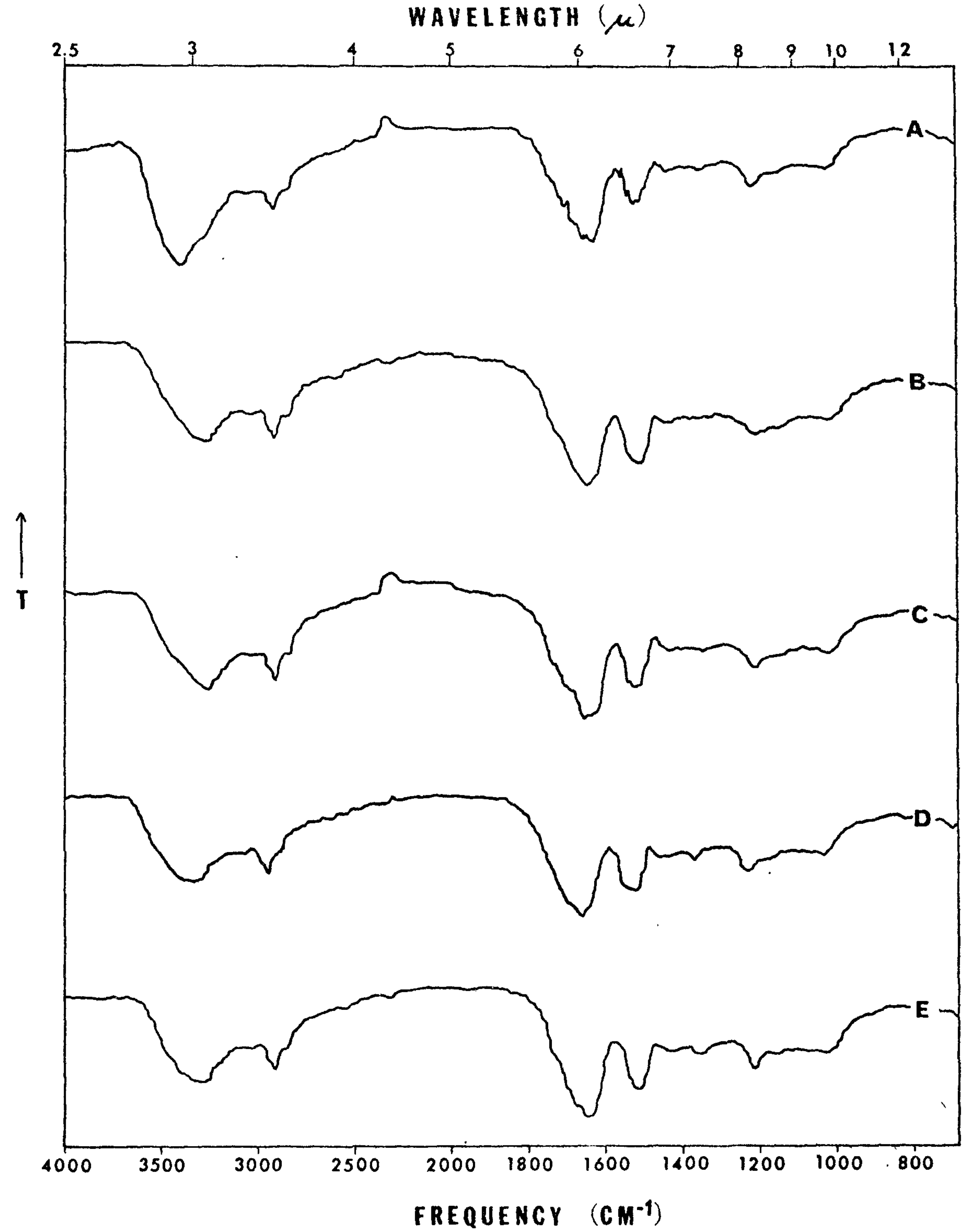


Figure 44. Infrared spectra of humic acids extracted from sludge with different concentrations of NaOH. Concentrations of alkali correspond to A, 0.1 N; B, 0.25 N; C, 0.5 N; D, 1.0 N; E, 2.0 N.

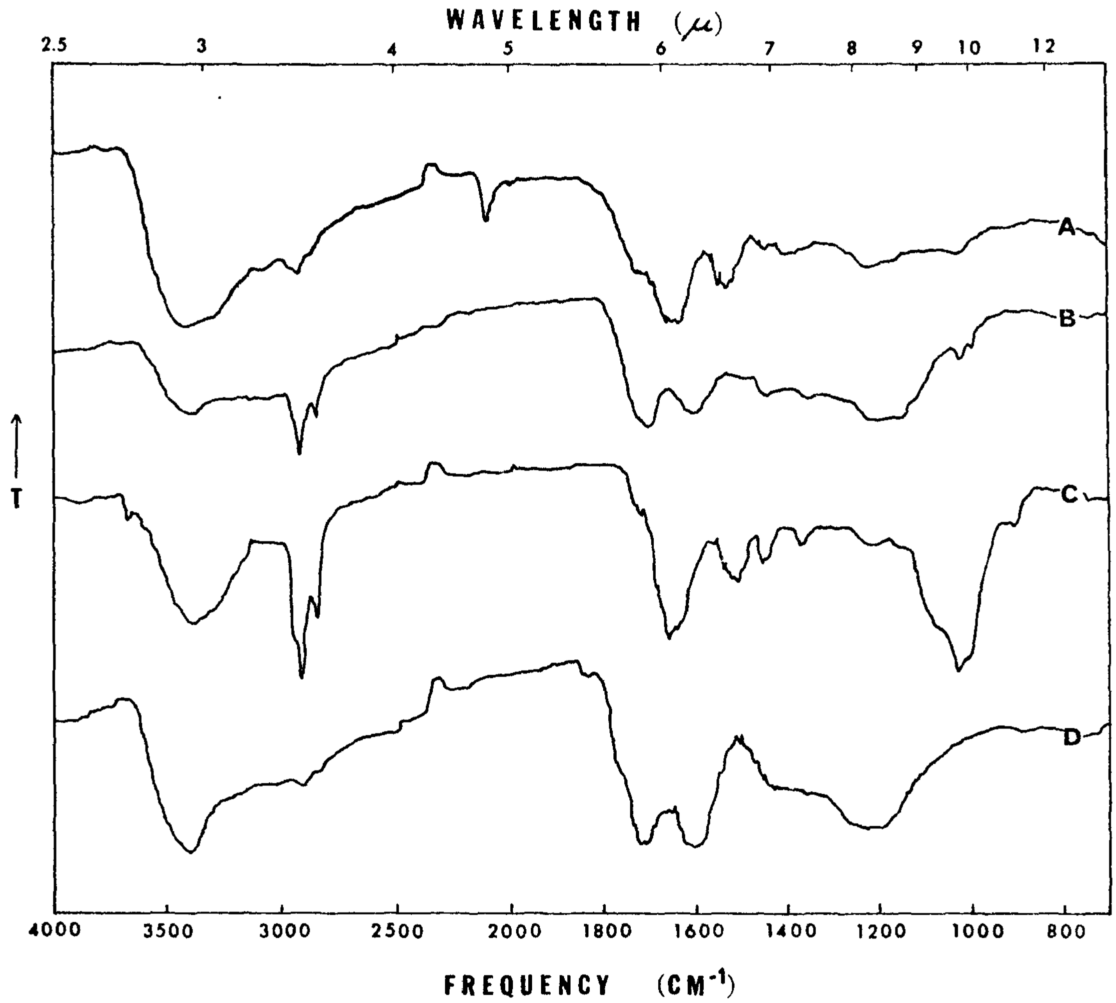


Figure 45. Infrared spectra of humic acid extracted from sludge with $Na_4P_2O_7$ (A), then hydrolyzed with 6 N HCl (B), and humic acid extracted from Leonardite with EDTA.

The IR spectra of sludge preparations and soil humic acids were similar in that both displayed absorption bands at 2900 and 3400 cm^{-1} due to aliphatic C-H stretching and H-bonded OH groups, but differed in that the former absorbed at 1650 and 1530, while the latter absorbed at 1720 and 1600 cm^{-1}. The spectra of sludge preparation closely resembled that reported by Stevenson and Goh (148) for lake humic acid where the absorption bands at 1650, 1530, and 1220 cm^{-1} were related to the protein content of the material. Accordingly, they were assigned to the amide I, II, and III bands of peptides. Thus, the 1650 cm^{-1} may be attributed to C-0 and C-N stretching (Amide I); the 1540 cm^{-1} band to mixed vibration of N-H in-plane bending and C-N stretching (Amide II); and the 1240 cm^{-1} absorption to mixed vibration of OCN and NH modes (Amide III). The lake humic acid was extracted from a sediment believed to consist entirely of algal materials and may be rich in peptides or proteins (21). Similarly, the siudge humic materials are derived from bacterial remains.

Hydrolysis of the $Na_4P_2O_7$-preparation with 6 N HCl resulted in loss of the stretching at 1660 and 1550 cm^{-1} due to removal of protein. The spectrum of the hydrolyzed sample closely resembled those of the soil humic acids. However, C-H stretching at 2900 cm^{-1} was stronger in the acid hydrolyzed sludge humic acid.

Mathematical model - A mathematical model based on Schubert's ion exchange equilibrium technique was developed for measuring stability constants of metal-polyelectrolyte complexes naturally occurring in soils and digested sewage sludge. The model was free of certain assumptions and errors inherent in the ion exchange technique as applied previously to natural polymer complexes.

The procedure was also suitable for determining values for $\underline{a}$, the number of metal ions per mole of complex or complexing sites, and $\underline{b}$, the number of ligands or complexing sites per mole where $\underline{a} \neq 1$ or is unknown.

The equation relation M_aCh_b (complex molecule), M_f (free metal ion), and Ch_f (free ligand) is obtained from the definition of the stability constant, K,

$$(M_aCh_b) = K(M_f)^{\underline{a}} (Ch_f)^{\underline{b}}$$

The concentration of complexed metal ion, M_c, can be expressed as:

$$(M_c) = \underline{a}(M_aCh_b)$$

which by substituting into the first equation yields

$$(M_c) = \underline{a}K(M_f)^{\underline{a}} (Ch_f)^{\underline{b}} \quad .$$

The logarithm of both sides yields

$$\log (M_c) = \log \underline{a} + \log K + \underline{a} \log (M_f) + \underline{b} \log (Ch_f)$$

A graphical approach was used to solve the latter equation. Essential steps are as follows:

1. Estimates were made for total metal ions in the solution phase, (M_f) or $(M_f + M_c)$, and on the resin, M_R, for various levels of applied metal, M, in the absence and presence of chelating agent, Ch.
2. Plots were obtained of M_R vs. $(M_f + M_c)$ from which (M_f) and (M_c) were obtained at a constant value of (Ch).
3. Value of $\underline{a}$ was obtained from the slope of the line obtained by plotting $\log (M_c)$ vs. $\log (M_f)$.
4. Steps 1 and 2 were repeated at several concentrations of (Ch), and (M_c) was obtained as a function of (Ch), at a constant (M_f). A plot of $\log (M_c)$ vs. $\log$ (Ch) gave $\underline{b}$ as the slope.
5. Values for log K were obtained from the equation of the intercept obtained in 3 $[I_a = \log K + \log \underline{a} + \underline{b} \log (Ch_f)]$ or 4 $[I_b = \log K + \log \underline{a} + \bar{a} \log (M_f)]$.

For more detailed information regarding the derivation of the mathematical model and its utilization in determining the stability constants for various metal-polyelectrolyte complexes, the reader should see the thesis prepared by M. Sobhan-Ardakani, Dept. of Agronomy, University of Illinois.

The procedure was applied for determining stability constants with Zn^{+2} for humic acids from two different soils, lignite, peat, and digested sewage sludge. In accordance with step 1 of the procedure, variable amounts of $ZnCl_2$ were added to 25 ml volumetric flask containing 0.1 to 0.5 g K-saturated Amerlite IR-120 cation exchange resin and specific volumes of humic acid stock solution previously prepared in 0.1 N KCl at pH 6.5. Before adjusting the volume to 25 ml, 2 ml of ^{65}Zn solution having an activity of 17,500 counts/minute per ml was added to the flask. Then a 1-ml aliquot of supernatant solution was used to measure the Zn concentration $(Zn_f + Zn_c)$ in the solution phase. The amount of Zn adsorbed on the resin, Zn_R, was determined by difference. From the plots of Zr vs. $(Z_f + Zn_c)$, as directed by step 2 of the procedure, concentration levels of Z_f and Zn_c were obtained at a constant concentration level of humic acid (Ch). The concentration levels of humic acids are expressed as "normality" or total quantity of potential acidic hydrogen (COOH plus acidic OH groups) or more specifically on the basis of potential complexing sites.

The data for the distribution of different species of Zn^{+2} in the ion exchange equilibrium system with the five humic acids are presented in Table 47. These data were used to make plots of log (Zn_c) vs. log (Zn_f) as directed by step 3 to obtain values of a (slope). Values for a from the slope of the plots presented in Figure 46 for the different samples range from 0.898 to 0.964. Furthermore, plots of log (Z_c) vs. log (Ch_f) made from the data obtained from three levels of humic acids yielded values of b (slope) ranging from 0.96 to 1.06. Within experimental error the value of b can be assumed to be unity meaning that the Zn-humic acid complex contained one molecule of humic acid.

Table 47. Distribution of Zn^{+2} species in the ion exchange equilibrium system in the presence of different humic acids and 0.1 N KCl, pH 6.5.

	Concentration		Zn_R	Zn_f	Zn_c
Humic acid	mg/l	N x 10^3	μmoles	μmoles/l	μmoles/l
Peat	200	1.15	0.252	9.14	17.14
			0.759	15.30	27.69
			2.431	30.60	55.53
			5.580	61.20	82.31
Harpster	200	0.93	0.209	6.89	21.42
			0.576	12.24	38.09
			2.137	27.54	70.38
			4.809	53.55	120.89
Brunizem	200	1.06	0.424	10.71	9.18
			1.000	16.83	15.37
			2.794	36.72	35.16
			6.137	68.08	53.55
Leonardite (hydrolyzed)	200	1.20	0.252	9.18	17.44
			0.645	15.30	32.13
			2.362	30.60	58.29
			5.385	58.14	93.17
Sludge ($Na_4P_2O_7$)	200	0.99	0.382	9.95	11.48
			0.954	16.83	18.36
			2.862	33.66	35.16
			5.882	64.26	67.32

With the values of a and b determined, all the information as required in step 5 for calculating the stability constants for the five Zn-humic acid was available. Using the derived values, the stability constants

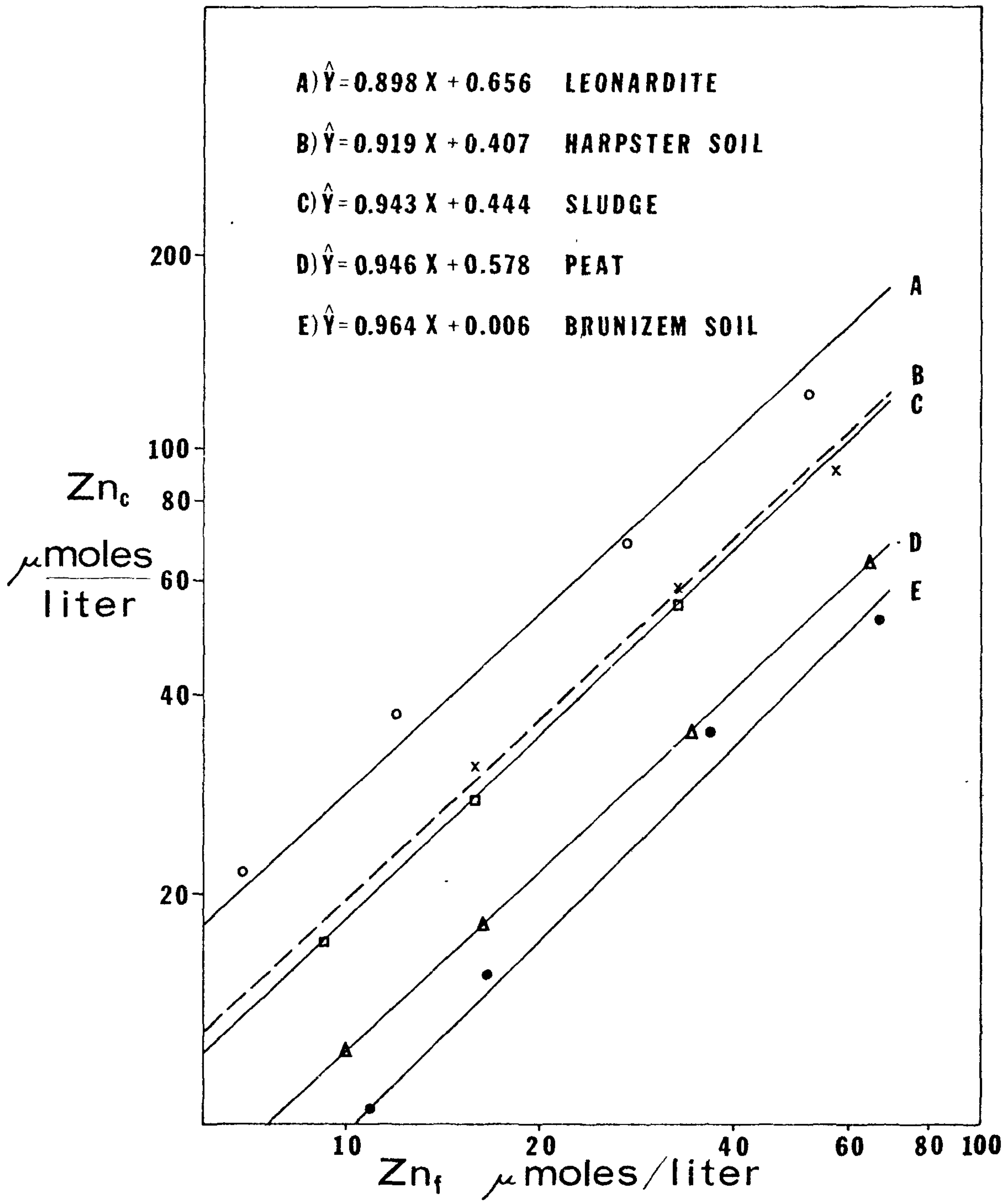


Figure 46. Graphical solution of the equation log (M_c) = log $\underline{a}$ + log K + $\underline{a}$ log (M_f) + $\underline{b}$ log (Ch_f) for five Zn(II)-humic acids from the data given in Table 47.

for the five different Zn-humic acid complexes are given in Table 48. Values of log K for the humic acid preparations varied from 2.98 for Zn^{+2}-Brunizem humic acid to 3.85 for Zn^{+2}-Leonardite humic acid. Considering the heterogeneous nature of humic acids, i.e. differences in molecular weights, functional group contents, and probably arrangements of complexing sites, differences in formation constants would be expected.

Table 48. Stability constants of Zn^{+2}-humic acid complexes at pH 6.5 and μ = 0.1 N KCl measured by ion exchange equilibrium.

Source of humic acid	Log K
Leonardite	3.85(3.22)[a]
Harpster soil	3.51
Sludge (0.15 N $Na_5P_2O_7$)	3.48
Rifle peat	3.31
Brunizem soil	2.98

[a] value in parentheses refers to a hydrolyzed sample

Potentiometric titration - The method of potentiometric titration was also adapted for determining stability constants of complexes of Cu^{+2} with naturally occurring polyelectrolytes. A modification of Bjerrum's approach was used in which an iterative procedure (Secant Method) was used to solve an exponential equation relating acid dissociation constant to hydrogen ion concentration and dissociated Ch, permitting calculation of stability constant. The University of Illinois' IBM 360/75 computer was employed for this purpose, using the Fortran IV language. Values for log K varied from 3.82 to 4.28.

Conclusion - The methods presented here for characterizing the complex reactions between metal ions and humic substances may be useful in predicting the fate of heavy metals applied as constituents of "stabilized" municipal sludges.

Digested Sludge Dewatering on Soils

Introduction - The rate at which digested sludge dewaters after application on crop land is one parameter which is needed to determine possible application frequencies and loading rates.

The rate of digested sludge drying as a function of convective and radiative heat transfer has been reported by Quon and Ward (124) and Quon and Tamblyn (125). By varying temperature, relative humidity and flow rates of air over a broad range of values, they found that when sludge temperatures were low and the air humidity was high, the rate at which digested sludge dried by convective heat transfer was only about one-half of evaporation from a free water surface. However, when sludge temperatures were high and air humidity was low, the rate of convective drying of digested sludge approached the rate of evaporation from a free-water surface. When evaporation was produced as a result of only radiant energy incident on the surface, the rates of evaporation from a digested sludge surface and a free-water surface were found to be essentially equal. At an intensity of 1.0 cal. per sq cm per min the evaporation rate was 0.9 x 10^{-3} gm per sq cm per min. One-half of the incident energy on the sludge surface was expended as latent heat of vaporization. When drainage or infiltration of digested sludge water into sand contributed to the sludge dewatering process, the evaporation rate from the sludge surface as a result of radiative heat transfer was depressed by 22 percent.

They found that digested sludge dried at a constant rate until its moisture content approached 70 to 90 percent of total weight. Where-ever the rate of evaporation decreases in the range from 70 to 90 percent moisture, it has been referred to as a critical moisture content for sludge dewatering.

The present study was undertaken to determine how digested sludge dewaters on soils. Special attention was given to determining what effect the antecedent moisture content of soils and solids content of sludges has on the dewatering rate of digested sludge applied on crop land. At the same time, a rather cursory examination of the chemical properties of soil water samples collected by means of an evacuated porous ceramic cup apparatus was made.

Experimental apparatus and procedure - The digested sludge used in the study was obtained from the Metropolitan Sanitary District of Greater Chicago's Calumet sewage treatment plant. Characteristics of the digested sludge are given in Table 49. More complete information with regard to chemical and physical properties of digested sludge from the Calumet treatment plant has been presented by Hinesly and Sosewitz (69).

Plexiglass cylinders, 14.4 cm in diameter and 48 cm long, were used to determine the rate of infiltration of distilled, tap and sludge water into Blount silt loam and Plainfield sand soils, both low in organic matter content. In the bottom of the cylinders, washed gravel was placed to occupy a depth of 2 cm. Level with the bottom of the gravel, small plastic tubes were installed through the walls of the cylinders.

Table 49. Digested sludge characteristics.

Sample Number	pH	Conductivity mmhos/cm	NH_4-N ppm	Total Solids %
1	7.35	3.87	626.9	2.41
2	7.54	3.58	535.5	2.52
3	7.71	3.30	574.7	2.44
4	7.40	3.78	604.6	1.74
5	7.10	4.83	862.8	5.53
6	7.20	4.60	715.6	4.40
7	7.12	4.77	595.6	4.99
8	7.61	4.15	491.5	3.10
9	7.43	3.62	522.9	2.72

The small tubes were used to convey effluent to sample collection containers. Soil was compacted to its original density for a depth of 36 cm over the gravel in the cylinders. All soil columns employed for constant head infiltration rates were saturated with tap water and allowed to drain three days before the studies were initiated. For the determinations of infiltration rates, water or sludge was maintained at a constant depth of 5 cm above the soil surface.

After the infiltration studies were concluded, three of the plexiglass cylinders were reused to investigate changes of soil pH and Eh (redox potential) when sludge loading rates were varied. Six equally spaced holes were drilled through the cylinder walls at depths of 8, 18 and 28 cm below the surface of the Blount silt loam soil column. The holes, fitted with rubber stoppers, provided an access for removing small core or plug samples from the soil columns at the above respective depths each week for six weeks after the beginning of sludge applications. Sludge loading rates for the three columns were 1.25 cm per week, 2.5 cm at two week intervals, and a constant sludge depth of 5 cm above the soil surface. The pH and Eh determinations were made from suspensions of 5 g of soil sample and 15 g of boiled, distilled water immediately after each sample was extracted from a soil column.

Plexiglass cylinders of the same dimensions and construction were used to study sludge dewatering by drainage and evaporation on the surface of Blount silt loam and Plainfield sand. After digested sludge containing 3.1 percent total solids was applied on the surface of the soil columns, the depth of the sludge surface was measured as a function of time. As dewatering proceeded the sludge surface eventually decreased to a level where its height above the soil changed very little. At the point where decreases in sludge depths were small with time, small plug samples of the sludge were taken for moisture determinations. At the beginning of a study, the moisture content of the sludge was calculated from decreasing surface level values, but after the solids became concentrated enough to present a somewhat stable surface level moisture contents were determined gravimetrically. The rate of sludge dewatering was then calculated as cm/min from the various moisture content determinations so that the units of measurement would be consistent with the earlier recorded liquid sludge depths.

Small glass pans, 6.0 cm in diameter and 4.0 cm high, were used for determining the convective evaporation rate of water and sludge. Evaporative losses were determined by weighing the pans three times each day. The depth of sludge or water in the pans ranged from 0.5 cm to 3.5 cm.

All of the above studies were conducted in an air-conditioned laboratory where changes in air temperatures and relative humidity were small. Temperatures ranged from 23 to 25°C and relative humidities from 30 to 37 percent.

During the latter part of the summer of 1968, 32 infiltration and sludge dewatering studies were conducted on a Blount silt loam soil located on the NE Agronomy Research Center, near Elwood, Illinois. The field studies were made by applying sludge or water in metal cylinders which were 51 cm in diameter, 60 cm long and pressed into the soil to a depth of 28 cm. Sludge was applied to depths of 1.25, 2.5, 5, 7.5 and 10 cm in the metal cylinders. When water was applied, its depth was always 5 cm per application.

The infiltration rates of sludge or water were calculated from measurements of sludge or water surface levels with time and the soil surface area enclosed by the metal cylinders.

Tensiometers were installed at depths of 7.5 and 33.5 cm below the soil surface inside some of the infiltration cylinders. Also, porous ceramic cups were installed usually at two depths below the soil surface inside some of the infiltration cylinders. To obtain soil solution samples, the porous ceramic cups were evacuated by means of a 50 to 60 cm hanging mercury column. Nitrate concentrations, pH, conductivity, and redox potential values were determined from the soil solution samples.

Results and analysis - The change in infiltration rates or hydraulic conductivity with time, when laboratory prepared columns of soil had a constant 5 cm depth of digested sludge containing 2.7 percent solids maintained on the surface, is shown in Figure 47. The change in infiltration rate for tap and distilled water on Blount silt loam is shown in the same figure. At first the infiltration rate of digested sludge on sand was much greater than on the silt loam soil, but after about three days

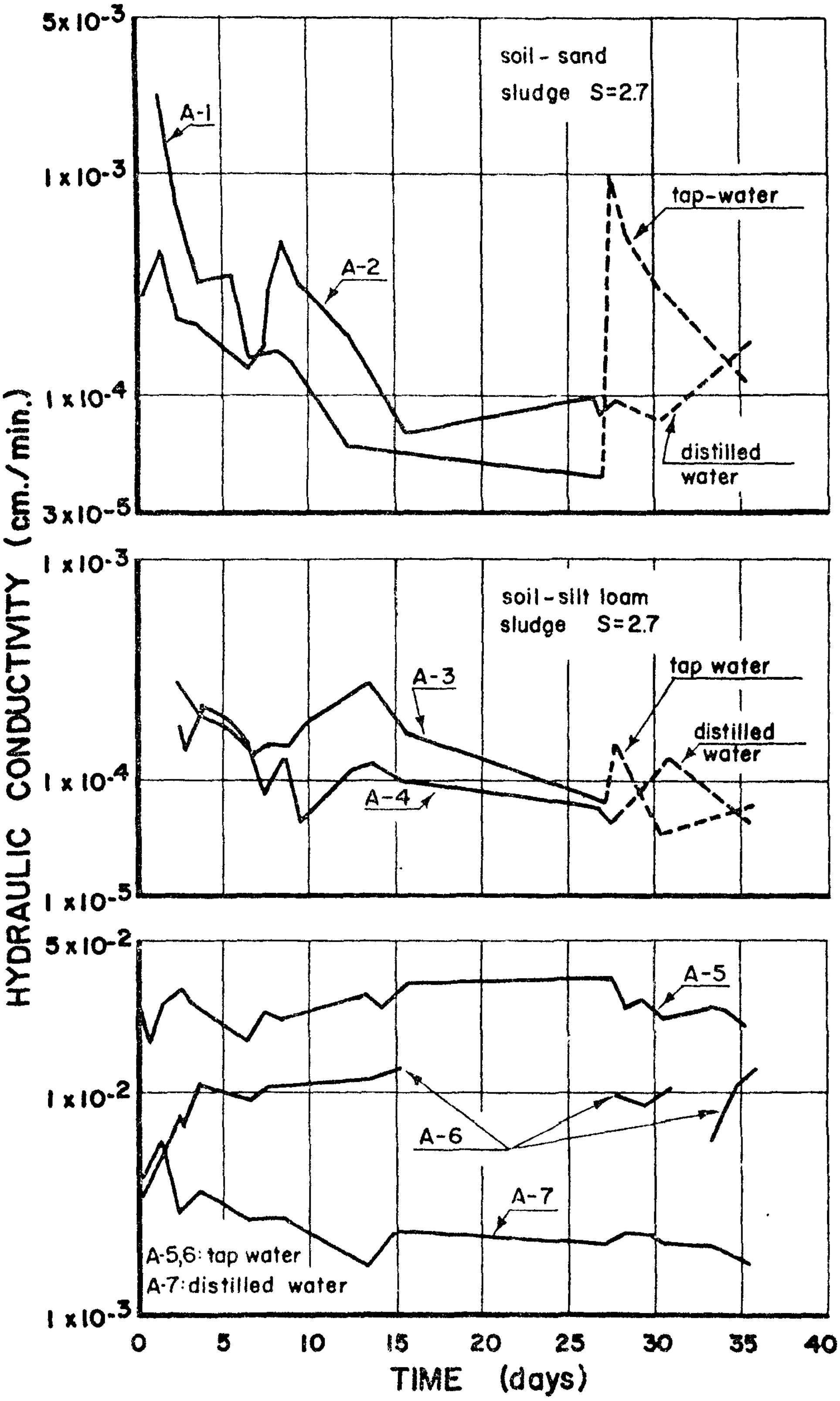


Figure 47. Hydraulic conductivity values of Plainfield sand A-1,2 and Blount silt loam A-3,4,5,6,7 as determined with digested sludge and water.

the difference between rates for the two soils was small. It appears that after about three days the infiltration rate of digested sludge is determined by the solids accumulating on the soil surfaces. When digested sludge was carefully replaced on the 27th day with either tap or distilled water on either surface of the two different soil types the infiltration rate was temporarily increased. The temporary increase was probably due to the unavoidable disturbance of the solids that had accumulated on the soil surfaces.

When distilled water was used to determine the hydraulic conductivity of Blount silt loam, the more or less constant value found after about 10 days was considerably less than that found with tap water. The hydraulic conductivity of Plainfield sand was about 10 times greater than for Blount silt loam and varied very little with regard to whether tap or distilled water was used.

Infiltration of water from digested sludge into Blount silt loam and Plainfield sand was only 1/15 and 1/400 of that in equal periods of time with applications of tap water.

Figure 48 shows the electrical conductivities and nitrate concentrations of effluent collected from the soil columns during the period in which the hydraulic conductivity studies were conducted. After one to two weeks the electrical conductivities of effluent from sand columns approached that of the applied sludge of about 3 mmhos/cm. On the other hand, electrical conductivities of effluent from Blount silt loam soil columns slowly increased during the duration of the study to only about 1 mmhos/cm.

Although the applied digested sludge did not contain NO_3-N, the nitrate concentration in effluent samples from columns of Plainfield sand approached values of 9 to 10 ppm (Figure 48) in about two weeks after the hydraulic conductivity study was initiated. Nitrate concentrations in the effluent of Blount silt loam at first decreased but later increased with time. However, nitrate concentrations in the effluent from the silt loam soil were only about one-half of that found in effluent samples from columns of sand.

Electrical conductivity and nitrate concentration values for effluent samples from Blount silt loam soil columns subjected to constant levels of surface water are presented in Figure 49. Nitrate concentrations decreased to less than 0.5 ppm during the first 10 days, regardless of whether tap or distilled water was used. On the other hand, conductivity values of effluent varied according to the kind of water used and approached that of the applied water.

When digested sludge is applied on the surface of soils, dewatering proceeds as a result of evaporation of water from the sludge surface and infiltration of water through the soil surface. Therefore, it was

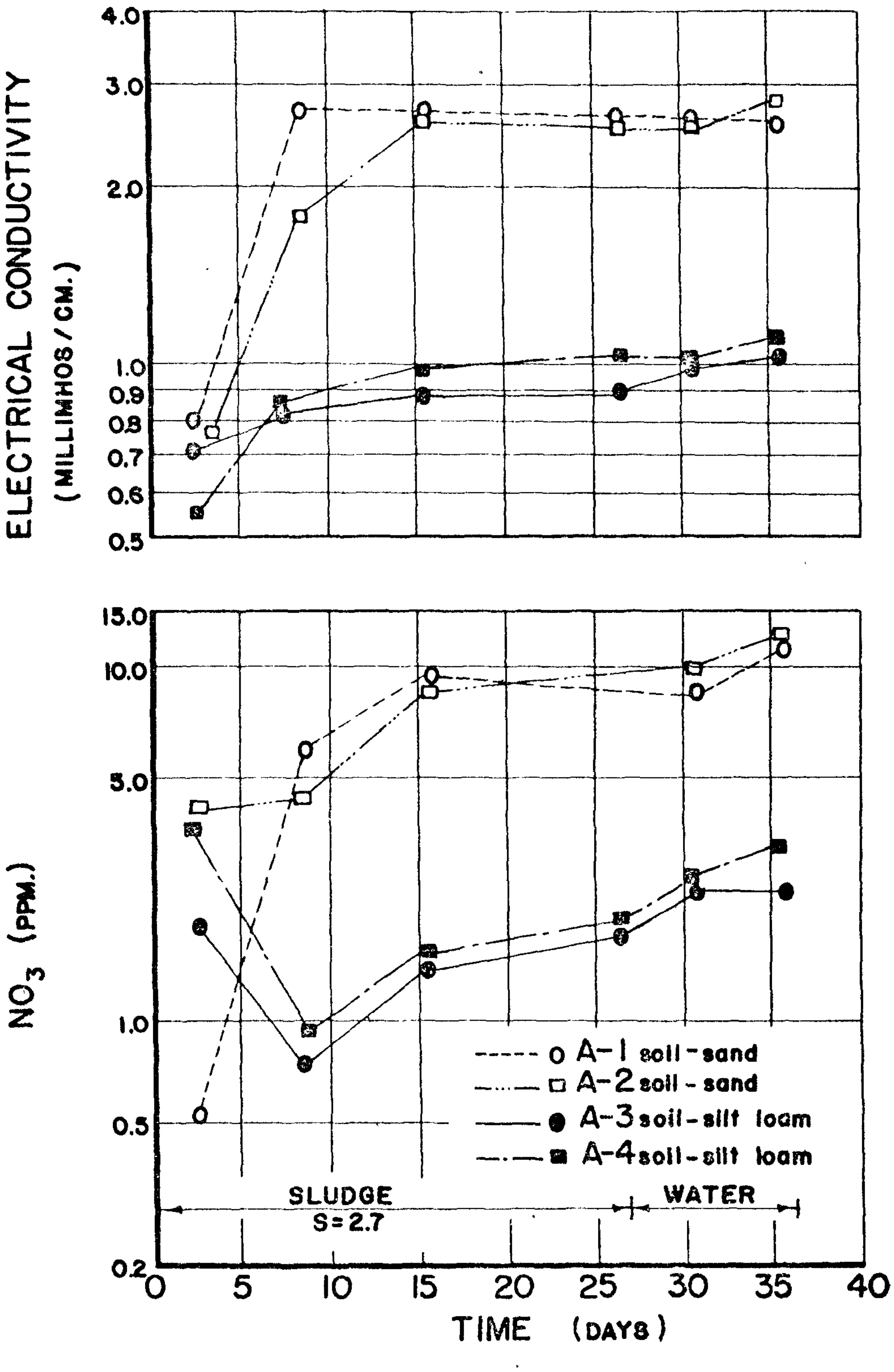


Figure 48. Electrical conductivity and nitrate concentration changes in effluent from Plainfield sand and Blount silt loam soil columns treated with digested sludge.

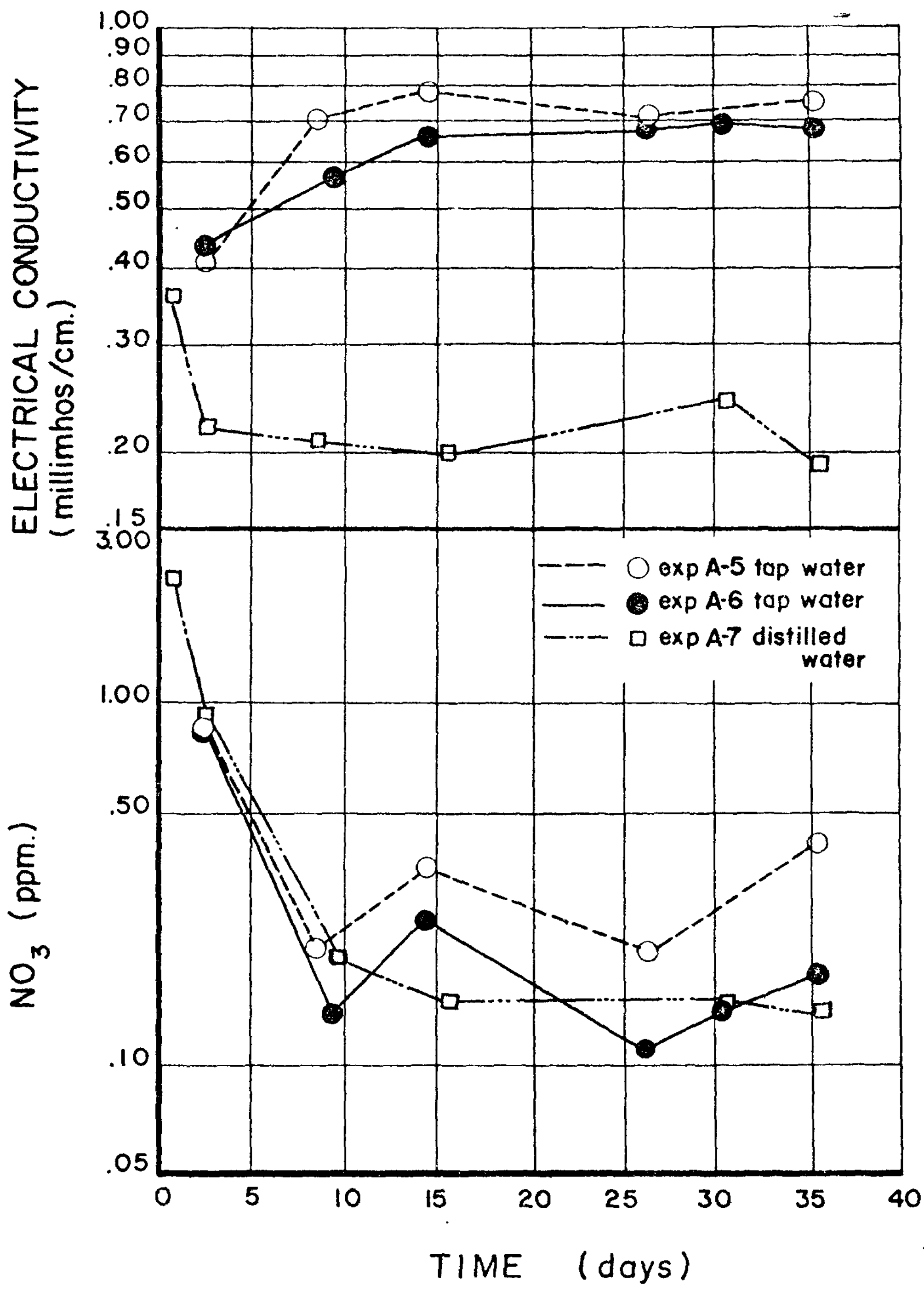


Figure 49. Electrical conductivity and nitrate concentration changes in effluent from Blount silt loam soil columns treated with water.

expected that antecedent soil moisture conditions would influence the rate of sludge dewatering by its effect on the infiltration rate.

The decrease in sludge surface levels with time after applications of 1.25, 2.5, and 5.8 cm of digested sludge on the surface of columns of Plainfield sand having different initial moisture contents is shown in Figure 50. From the results shown in Figure 50, it may be concluded that antecedent moisture does not affect the dewatering (evaporation and infiltration) of sludge on sands. When dewatering rates are calculated and plotted against time, as displayed in Figure 51, it may be seen that the initial dewatering rate is about the same (3×10^{-2} cm/min) for all loading rates. Initially the dewatering rate of sludge on sand is about 200 times greater than dewatering by evaporation alone. With the lowest loading rate of 1.25 cm, the dewatering rate of digested sludge on sand decreased to that of evaporation alone in about 800 minutes. When the loading rate was 5.8 cm about 9500 minutes were required for the dewatering rate on sand to be reduced to that expected by evaporation alone.

The change in sludge surface level with time after an application of 2.5 cm on columns of Blount silt loam soil at four different initial moisture contents is exhibited in Figure 52. From Figure 52 it is evident that the rate at which sludge dewaters on fine textured soil decreases as the initial soil moisture content is increased. The decreasing levels of water (E-5) and sludge (E-1,2) with time of evaporation from pans were plotted as a part of Figure 52 for the sake of easy comparison with the sludge dewatering data obtained from soil columns. By comparing evaporation plus infiltration, the significant contribution of infiltration to the dewatering of sludge at the several initial soil moisture contents is seen more clearly.

At soil moisture content of 34.5 percent, the sludge dewatering rate as a function of time on Blount silt loam is shown in Figure 53 to be only slightly greater than would be expected by evaporation alone. The lower the initial soil moisture content at the time of sludge application on fine textured soils the larger the contribution infiltration makes to the dewatering process. When the initial soil moisture content is low, the dewatering rate of sludge drops to an exceedingly low level in a short time as a result of the more rapid removal of water by capillary absorption into the soil.

When the dewatering rate of sludge on a silt loam soil with a high initial moisture content is plotted against the moisture content of the sludge, as in Figure 54, it appears that the dewatering rate is greater than the evaporation rate (2×10^{-4} cm/min) as long as the moisture content of sludge is greater than about 80 percent. Thus, infiltration contributes to sludge dewatering on initially moist soils as long as

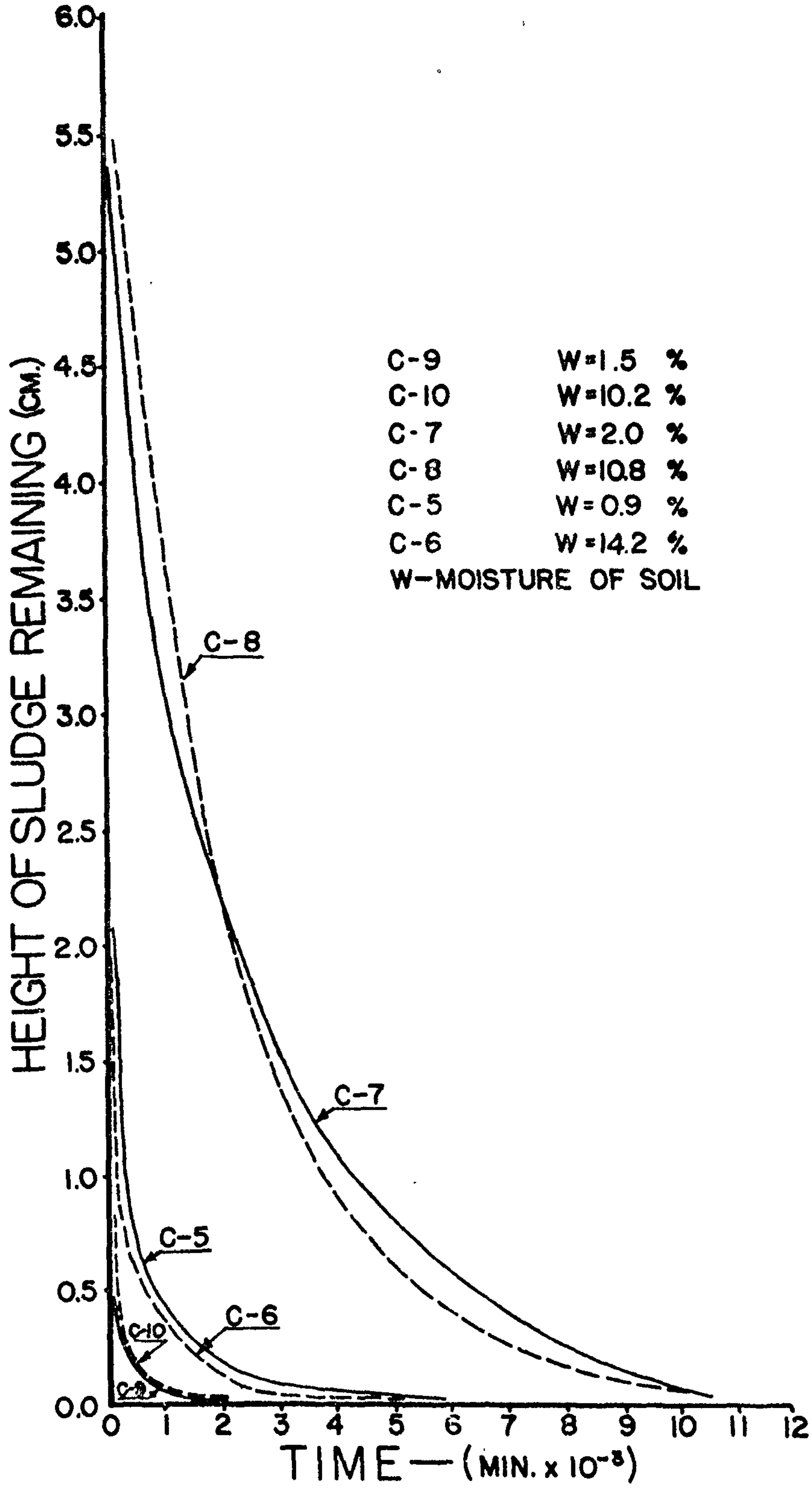


Figure 50. Decrease of digested sludge surface levels with time for several loading rates of digested sludge on Plainfield sand soil columns at different initial moisture contents.

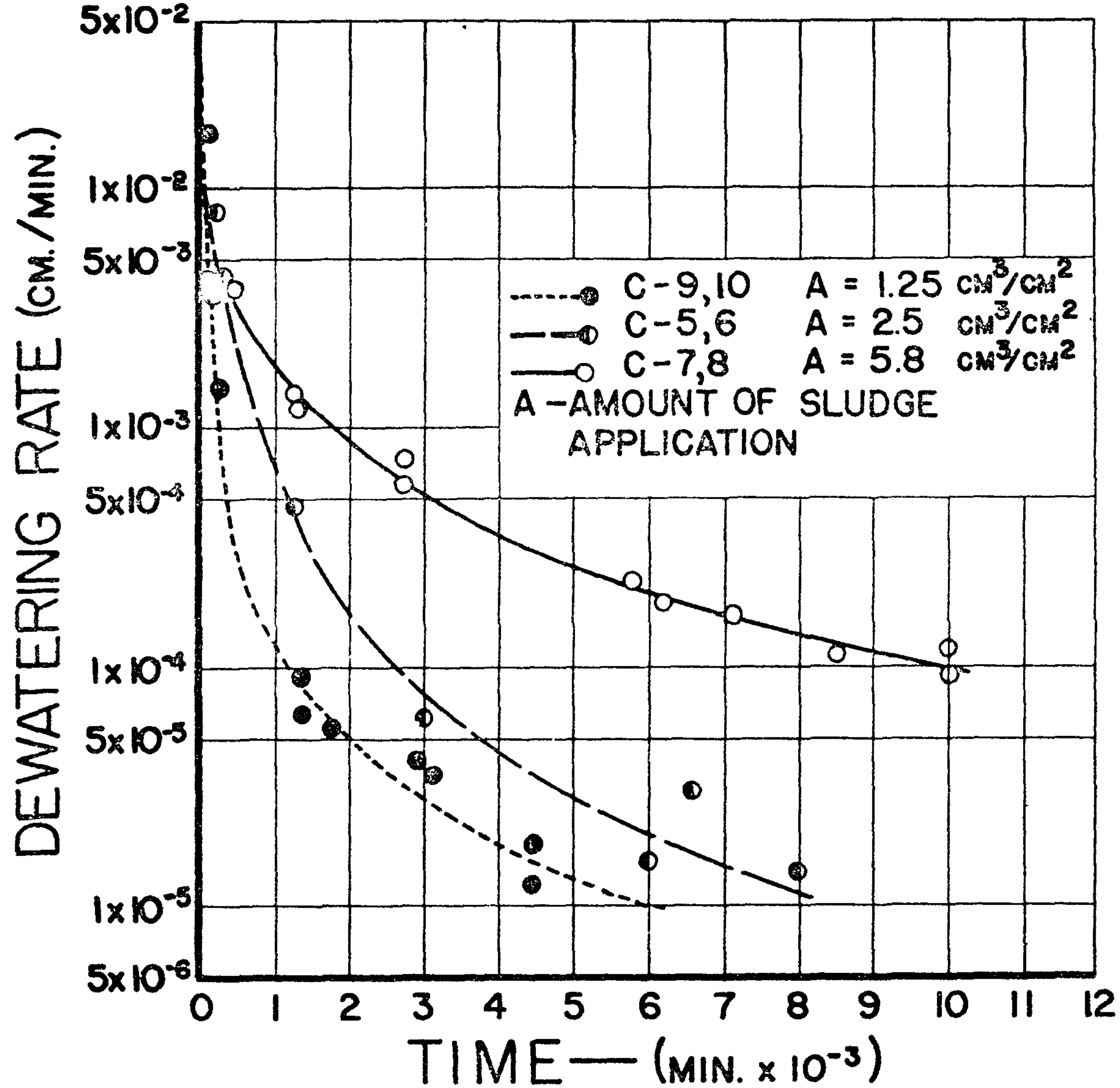


Figure 51. Changes in dewatering rate with time after digested sludge is applied at three different loading rates on columns of Plainfield sand.

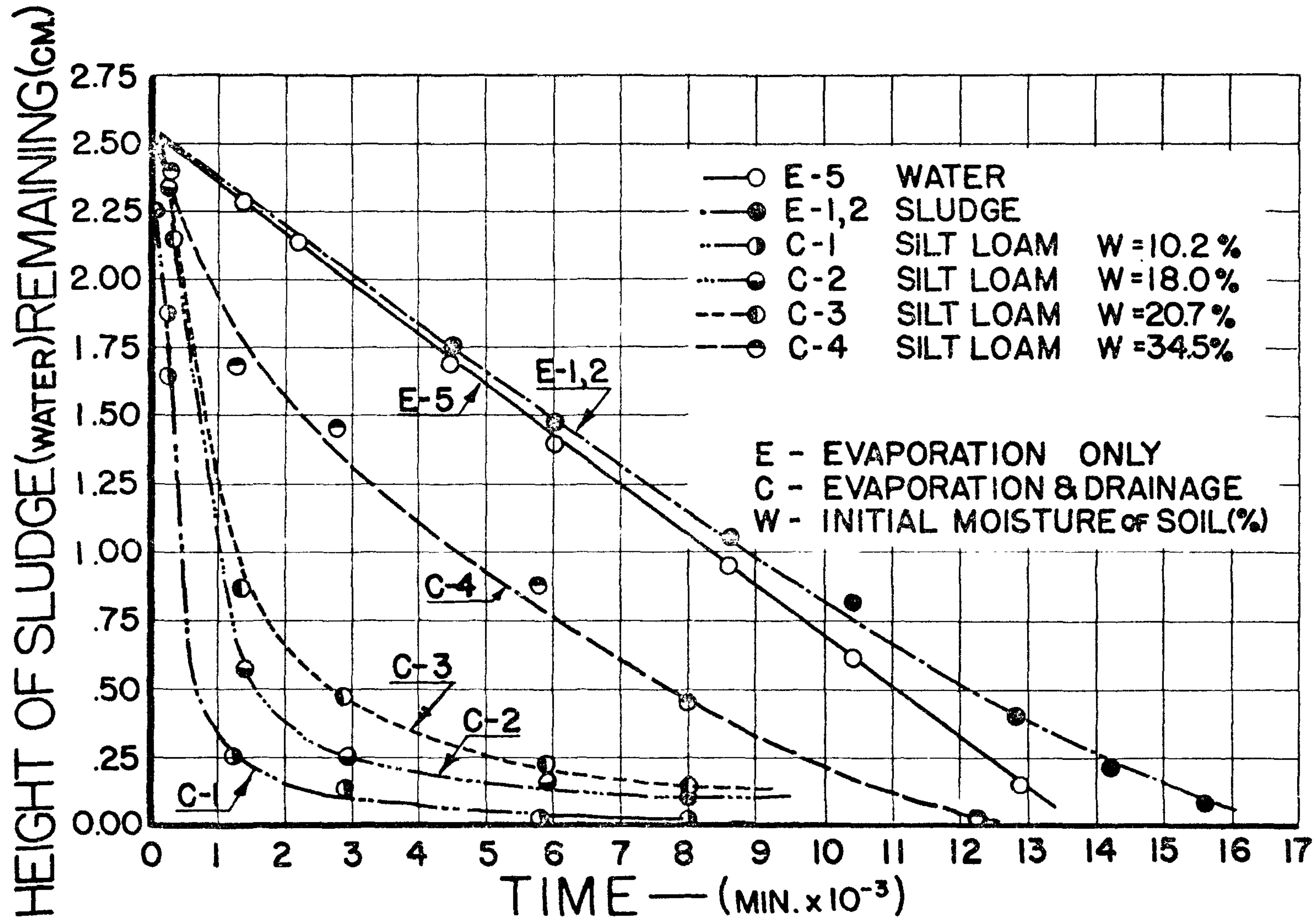


Figure 52. Decrease of height of the surface of digested sludge above an arbitrary datum with time on Blount silt loam soil columns at different initial moisture contents. Sludge and water in the absence of soil included for comparison.

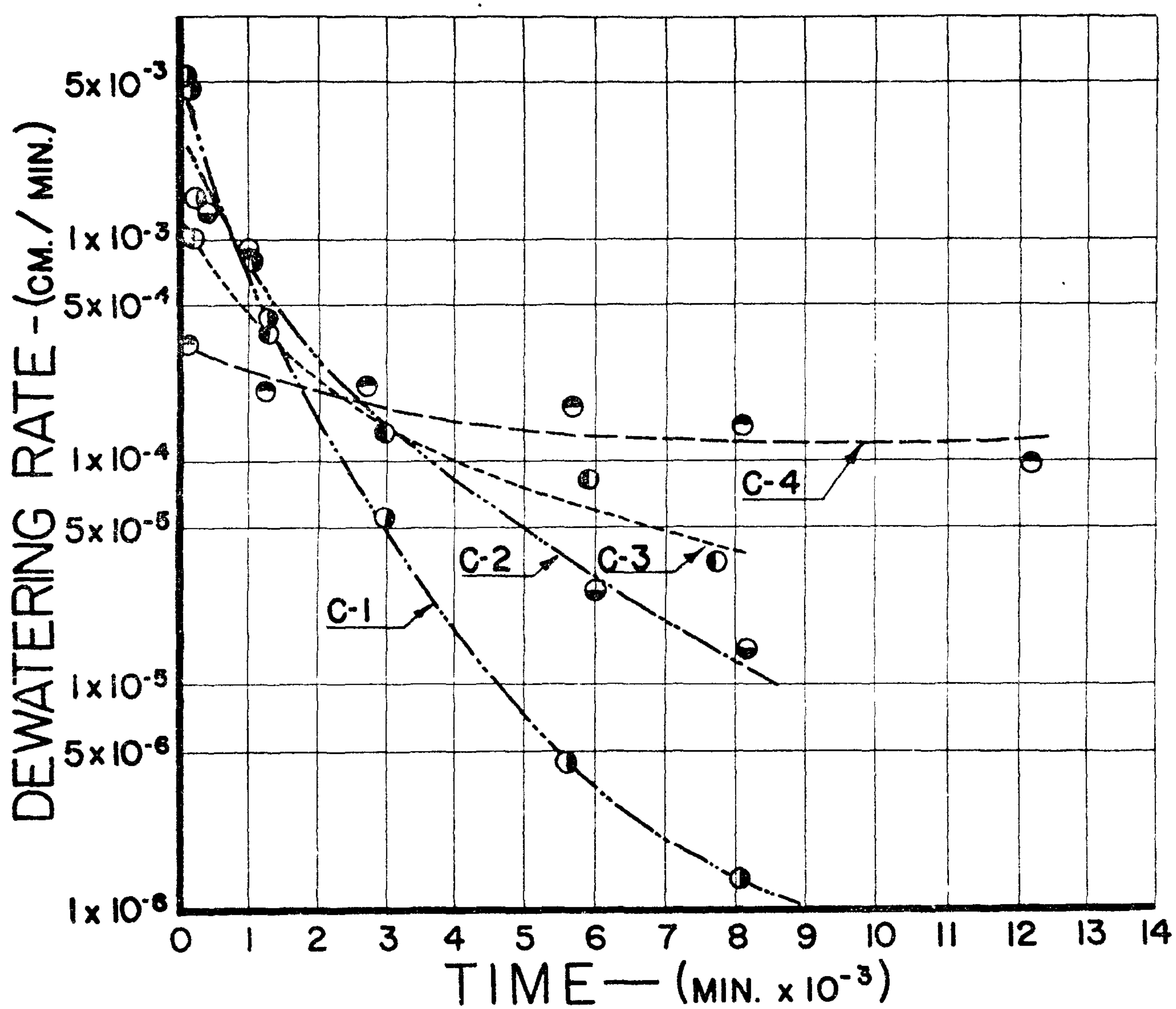


Figure 53. Dewatering rate changes of digested sludge with time on Blount silt loam soil columns at different moisture contents.

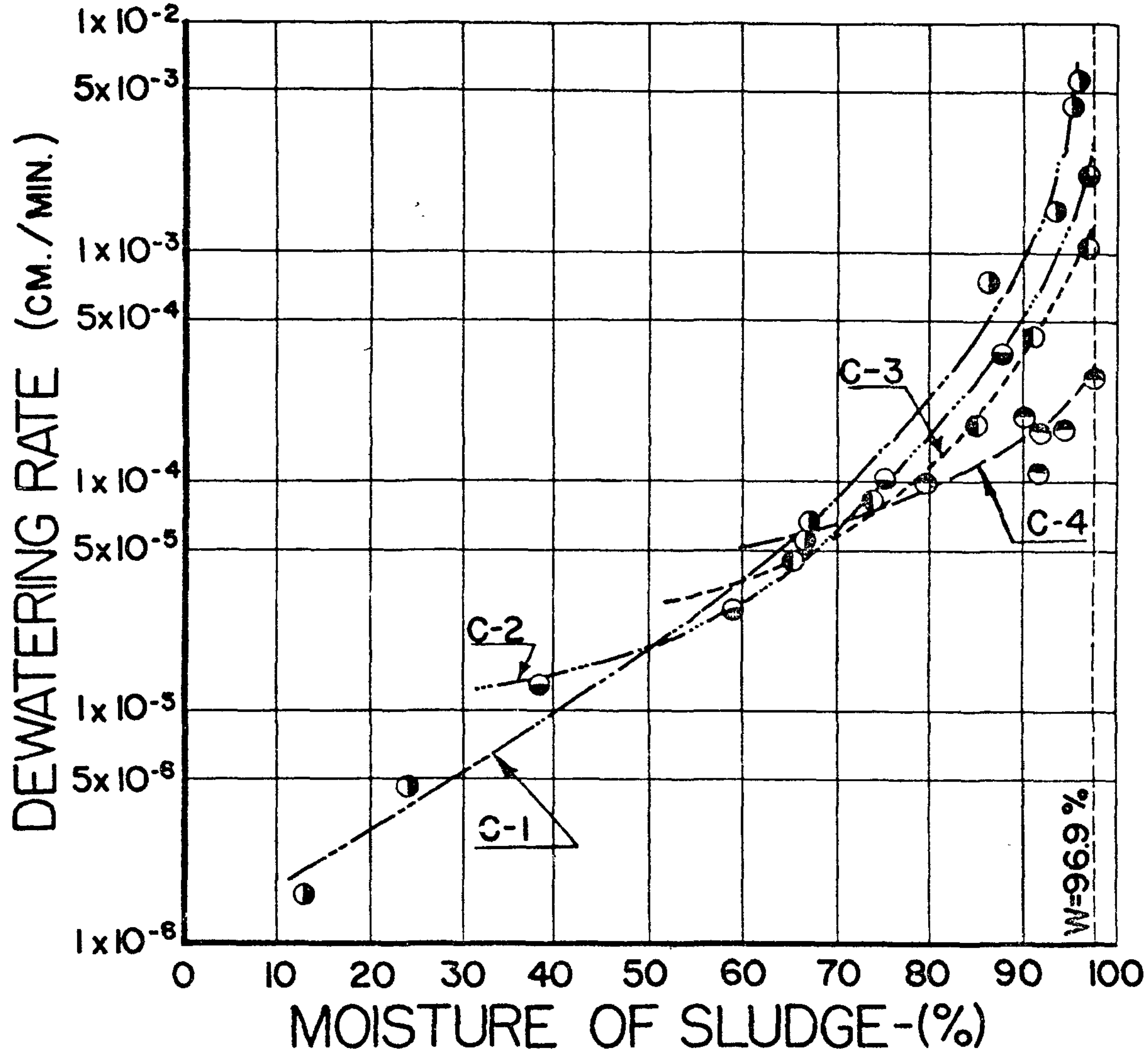


Figure 54. Dewatering rate changes of digested sludge as a function of decreasing sludge moisture contents on Blount silt loam soil columns at different initial moisture contents.

the sludge moisture content is greater than 80 percent. Once the moisture content of sludge has decreased to a value lower than 80 percent, for the most part further dewatering appears to be by evaporation alone. With smaller initial soil moisture contents it appears that a greater quantity of sludge water is absorbed by the soil and the point at which further moisture losses are due to evaporation alone occur at lower sludge moisture contents. It may be seen from Figure 54 that after the point has been reached where further sludge dewatering appears to be due to evaporation alone, the decrease in evaporation is proportional to the decrease in initial soil moisture content.

The sludge and water surface levels in pans as a function of time under the convective evaporational conditions previously discussed, is shown in Figure 55. The decreasing sludge surface level by evaporation with time is a rectilinear relationship until the moisture content of sludge has been reduced to 80 to 85 percent. As shown by experiments E-1,2 and E-4,5 (Figure 55) where evaporative losses of sludge and water, respectively, were determined over a period of 10 days, the evaporation of water from sludge was only slightly less than that from a free water surface during the constant rate period. The constant rate period was independent of sludge depth as determined under the stated laboratory conditions of temperature, relative humidity, air movement, etc. But, at sludge moisture contents of 80 to 85 percent, the decreasing rate of evaporation was definitely ordered with respect to sludge depths. The decrease in sludge drying rate at the critical moisture content was more pronounced for the greater initial sludge depths. As shown in Figure 56, the evaporation rate for both water and sludge approaches 2×10^{-4} cm/min during the constant rate period. However, at the critical moisture content, the evaporation rate falls rather rapidly to about 1×10^{-4} cm/min at a sludge moisture content of about 10 percent. After the sludge moisture content was reduced to about 10 percent by weight, a further decrease in water content with time was small.

Much of the data obtained from the 32 field observations, which included a wide range of sequential sludge and water applications to obtain a variety of antecedent soil moisture and surface conditions, is summarized in Figures 57 and 58.

The relationship between the initial soil moisture content and the time required for a definite volume of sludge liquid to infiltrate Blount silt loam soil is shown in Figure 57. The points plotted in Figure 57 were derived as an average of values from several observations of sludge water infiltration rates where maximum values at the beginning of infiltration were excluded. The time needed for the infiltration of 0.5, 1.25, 2.50 and 5.0 cm of digested sludge water into the soil with various initial moisture contents is based on data obtained from observations made with digested sludge containing from 2.4 to 2.5 percent solids. As expected, the higher the antecedent soil moisture the more extended was the period of time required for a given infiltration volume.

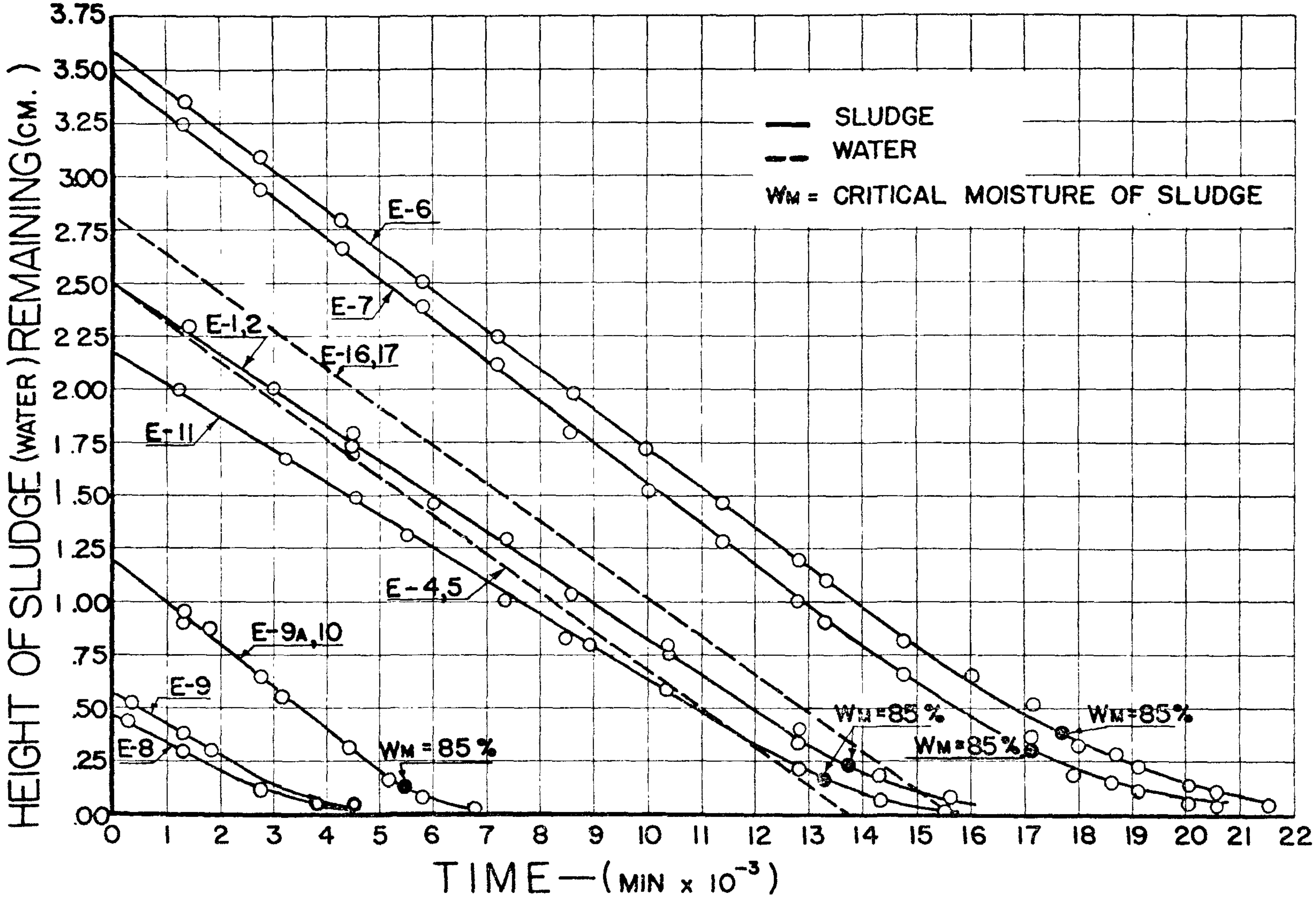


Figure 55. Changes in digested sludge and water surface levels in pans with time of exposure to constant conditions for convective evaporation.

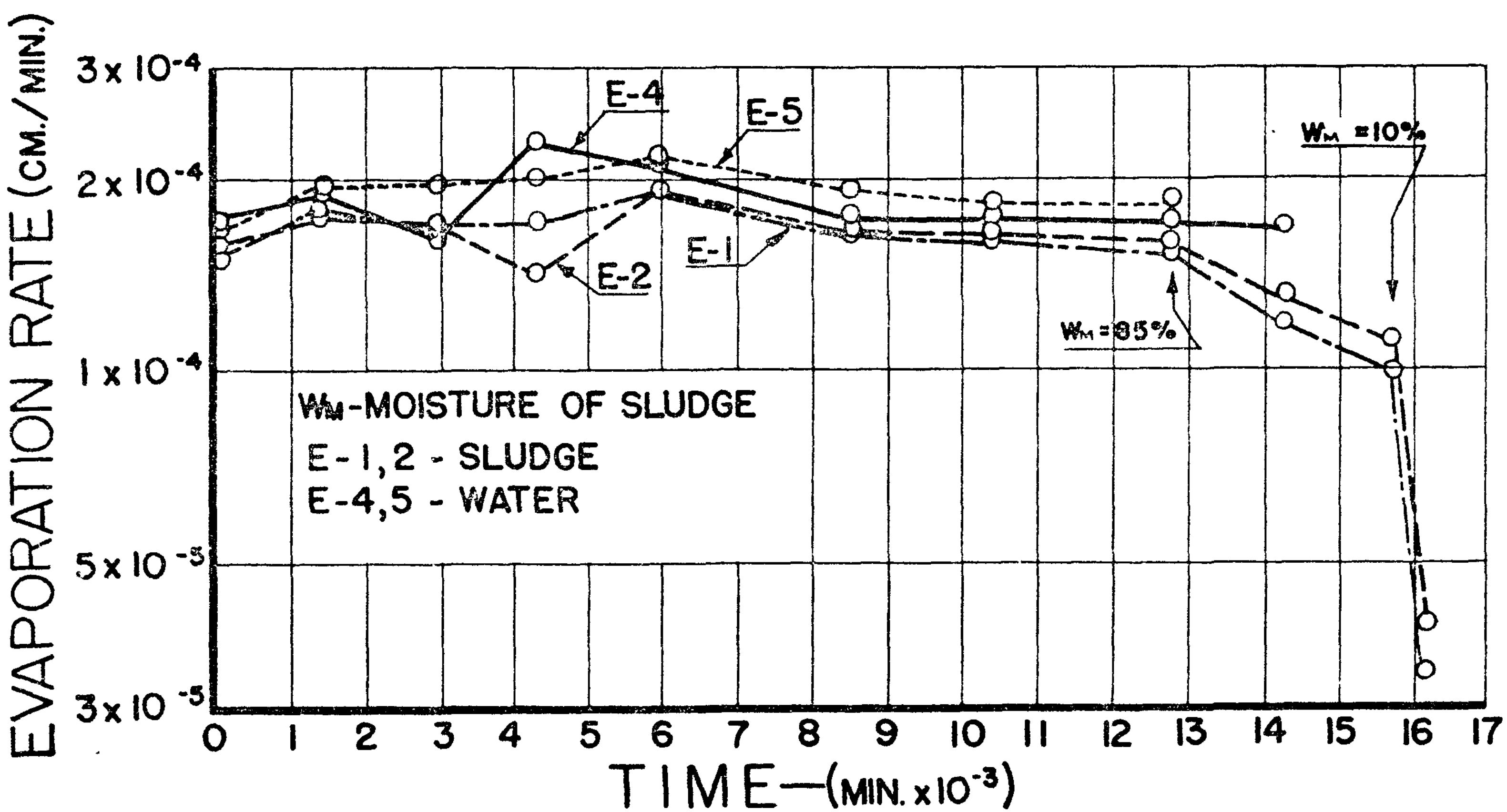


Figure 56. Changes in convective evaporation rate of digested sludge and water in pans with time.

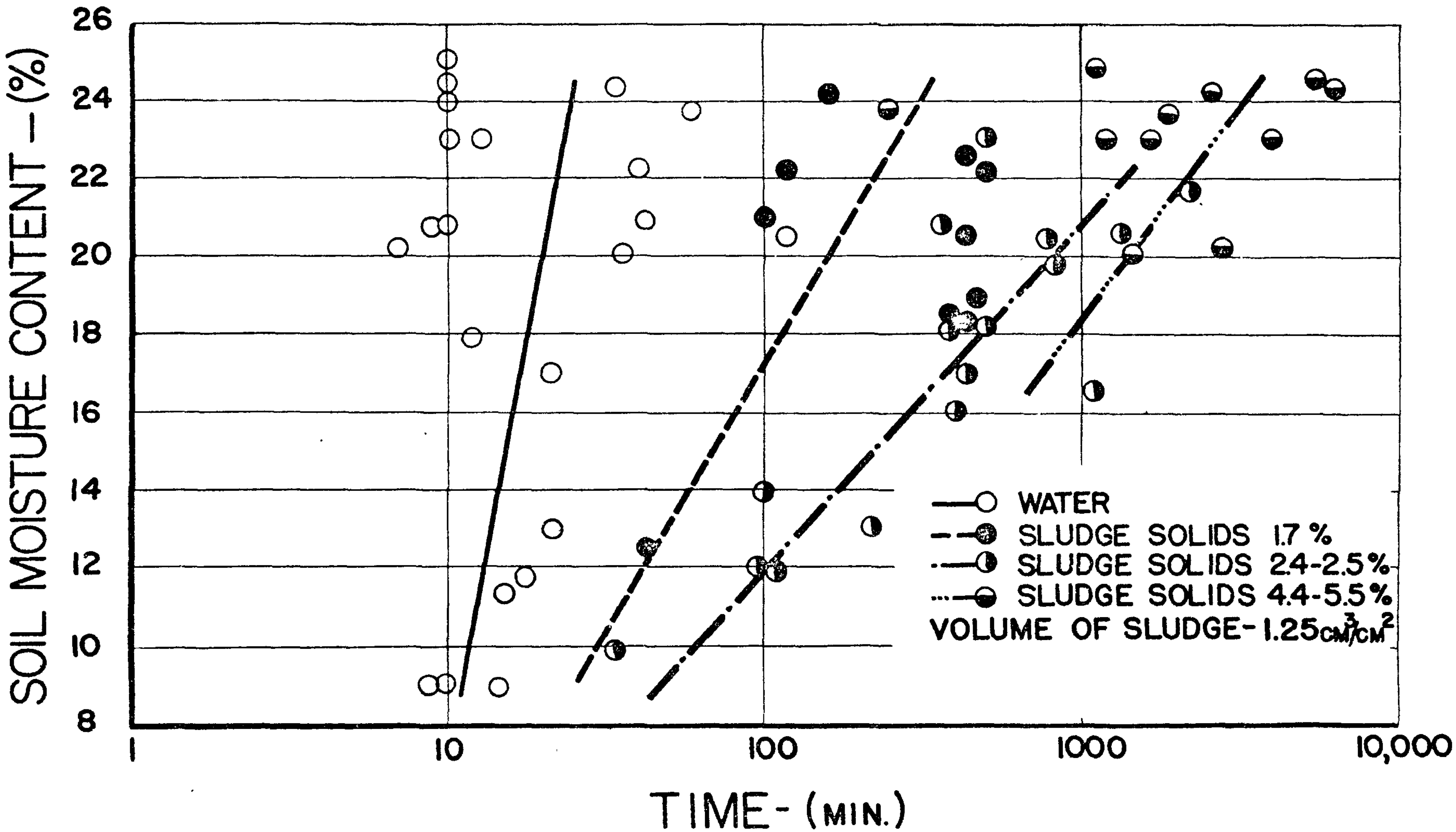


Figure 57. Time required for a given application of digested sludge to infiltrate the surface of Blount silt loam soil at different initial moisture contents.

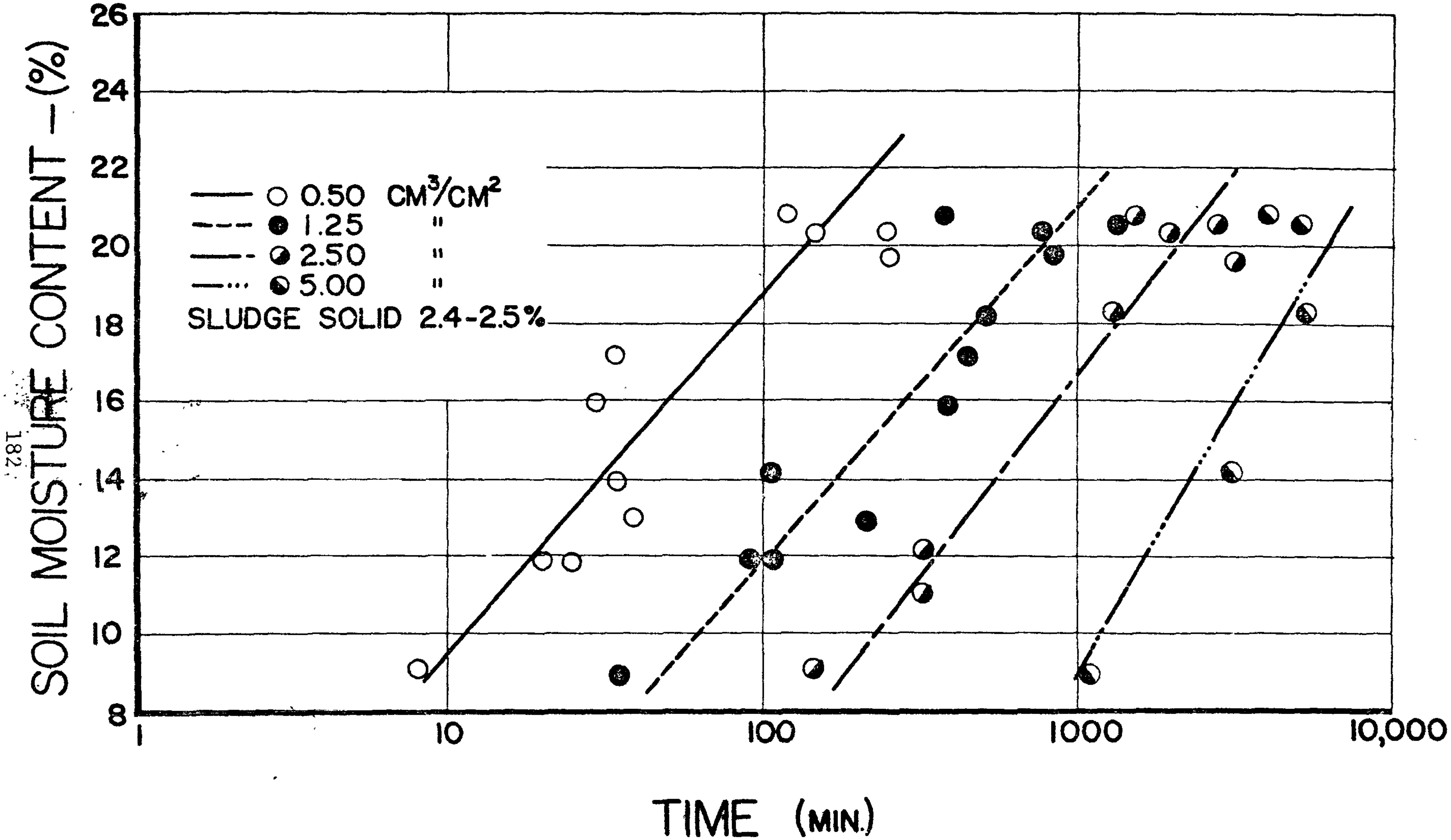


Figure 58. Time required for 1.25 cm of digested sludge liquid containing different amounts of total solids to infiltrate Blount silt loam soil.

The values plotted in Figure 58 are also average values and were obtained from the data of several infiltration studies where water and sludge containing various percentages of total solids were used. Specifically, the relationship between the initial soil moisture content and the time needed for the infiltration of 1.25 cm of water or sludge liquid is shown in Figure 58. Sludge solid contents ranged from 1.7 to 5.5 percent and were found to have a definite influence on the infiltration rate of sludge water. Although the decrease in the infiltration rate of sludge water as its solids content increased was not as great on soils at low as contrasted to high moisture contents, the solids exert a considerable influence on the infiltration of sludge liquid into fine textured soils at all initial moisture contents. The greater the sludge solids content, the smaller was the contribution of infiltration to the sludge dewatering process.

As stated above, the compilation of data to produce Figures 57 and 58 was obtained by extracting information from individual studies, a few of which will be briefly discussed here.

The results obtained after two consecutive sludge applications of 7.5 cm and 5 cm are presented in Figure 59. The change in sludge surface level was measured three times daily during the more rapid infiltration period and at least once a day thereafter. Sludge evaporation was measured in a nearby metal cylinder of the same size and installation as that used for the sludge dewatering study except that the bottom was sealed watertight with a sheet of polyethylene. The cumulative volume was obtained by correcting the measured sludge surface level for moisture losses by evaporation and gains by rainfall. During the first part of the dewatering study, the humidity was especially high and evaporation was small but during the last 15 days the evaporation rate was much higher as a result of lower humidity. The greatest infiltration rate occurred immediately after sludge application and continued at a rather high rate only during the first day, after which it gradually declined. The tensiometer data bear out that fact that the rapid period of infiltration lasts for only a short period of time. Soil water suction became fairly stable about two days after a sludge application. Except for a longer lag period, the response of the deeper tensiometer to changing moisture conditions at the surface did not differ much from the shallow tensiometer. After 4 to 7 days following rather high application rates of sludge, the beginning of soil moisture decrease was reflected by increasing higher suction values, although water was still being supplied to the soil from the sludge. The tensiometers at both the shallow and deeper depths responded rapidly to increases in infiltration of water as a result of rainfall. Even though the sludge cake was removed before the second sludge application was made (Figure 59) this was not necessary for the reestablishment of the infiltration capacity, as shown by other studies.

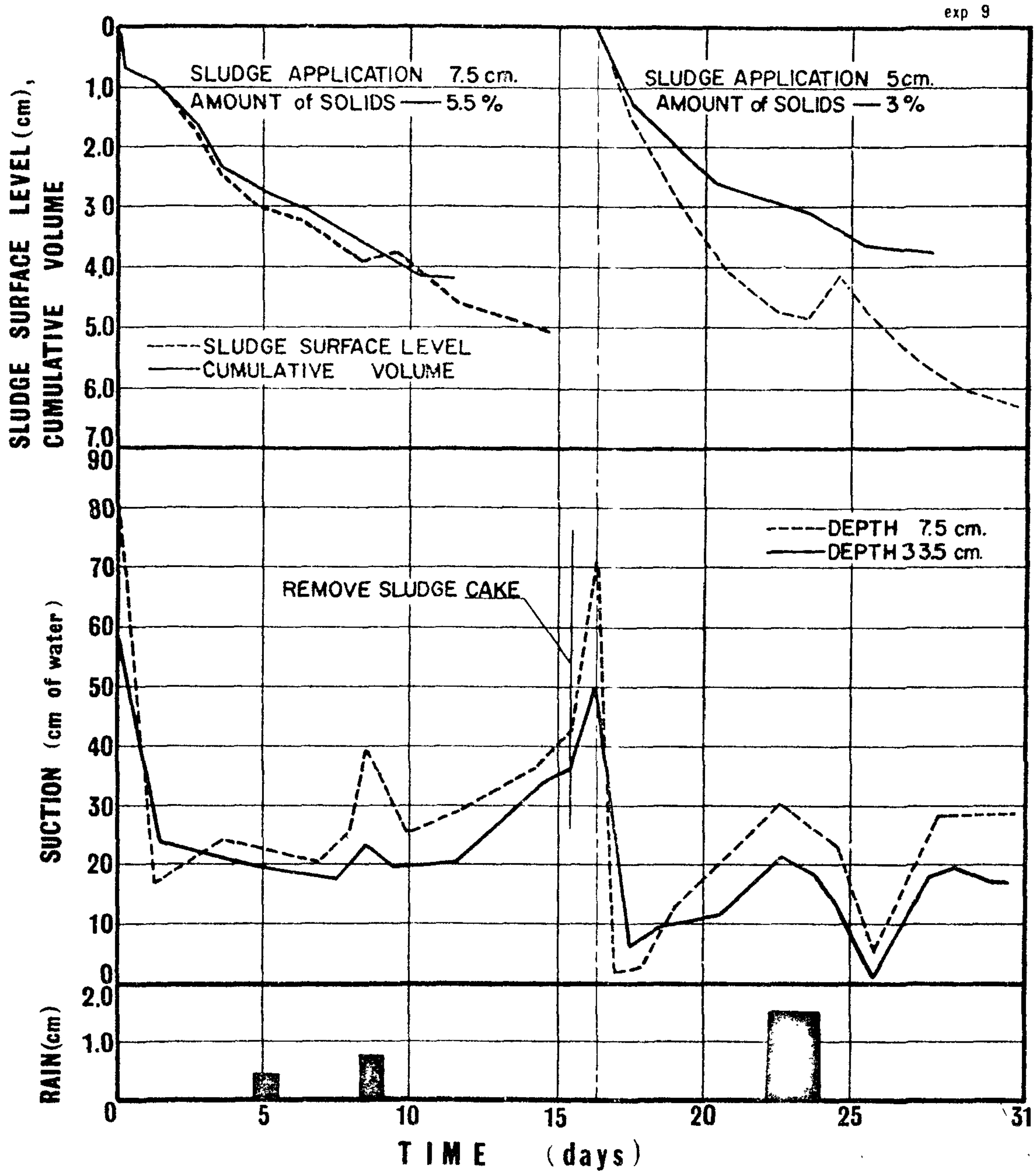


Figure 59. Changes in digested sludge surface levels, by sludge dewatering, by infiltration alone, and soil moisture suctions at two depths with time after sludge applications and rainfall.

In Figures 60, 61, 62, and 63 infiltration data are presented from a few of the 32 field observations in which initial soil moisture (W) determinations were made and where sludge application rates (A), and the solids content (S) of the sludge were varied. From Figures 60 and 61, it may be seen that the infiltration rate of sludge decreases with time in the same way with each succeeding sludge application. If enough time elapsed between sludge applications to permit drying, the infiltration capacity was restored almost to its original value. Also, successive sludge applications do not affect the infiltration rate for water if sludge is permitted to dry between applications. If water is applied before the sludge cake has dried, the infiltration rate is somewhat lower but still greater than when new sludge is added.

The changes of infiltration rates when large amounts of sludge are applied are shown in Figures 62 and 63. When successive 5 cm sludge applications were made, each time before the preceding sludge had completely dewatered, the infiltration rate increased for a short period after an application and then quickly decreased to a more or less constantly declining rate (Figure 62). When one of the cylinders was supplied only once with a 20 cm sludge application (Figure 63), the infiltration rate was observed to decrease continuously, except when rainfall caused short time increases.

Some changes in electrical conductivity values and nitrate concentration levels in soil solution, as successive application of sludge and water were made during the field infiltration rate studies, are presented in Figure 64. The samples were collected from several depths by means of porous ceramic cups buried below the soil surface which was enclosed by the infiltration cylinders. Toward the end of a 70-day period of observations and when total sludge applications exceeded 30 cm, the electrical conductivities of the soil solution were generally in the range of 2 to 3 mmhos/cm. Also, for the higher sludge applications, nitrate contents of the soil solution were in the range of 100 to 300 ppm. The increase in nitrate concentrations indicates that even at the highest sludge loading rates the soil remained aerobic. The fact that soil solution redox potentials did not change with sludge applications adds substantiative evidence that anaerobic conditions in the soil were not produced as a result of sludge loading rates. The loading rates included one where a total of 50 cm was applied within a 70-day period. For comparison with the data presented in Figure 64, electrical conductivity values and nitrate concentrations for soil solution samples collected from various depths where only water was applied on the surface are shown in Figure 65. Where only water was applied, electrical conductivity values were usually less than 1.0 mmhos/cm and nitrate concentrations seldom exceeded 5 ppm.

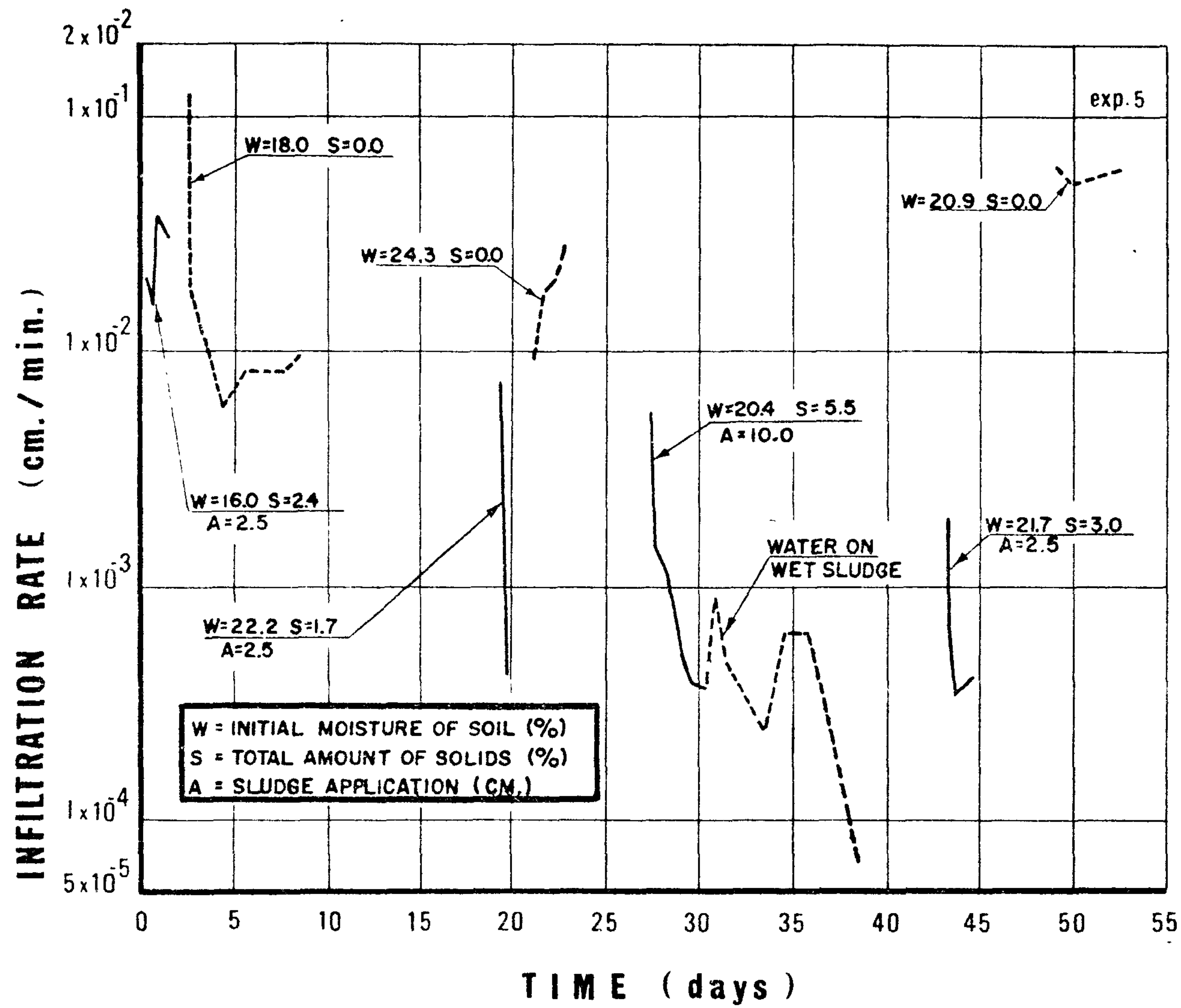


Figure 60. Changes of infiltration rates with time after water and digested sludge applications on Blount silt loam soil, where previous application was permitted to dry.

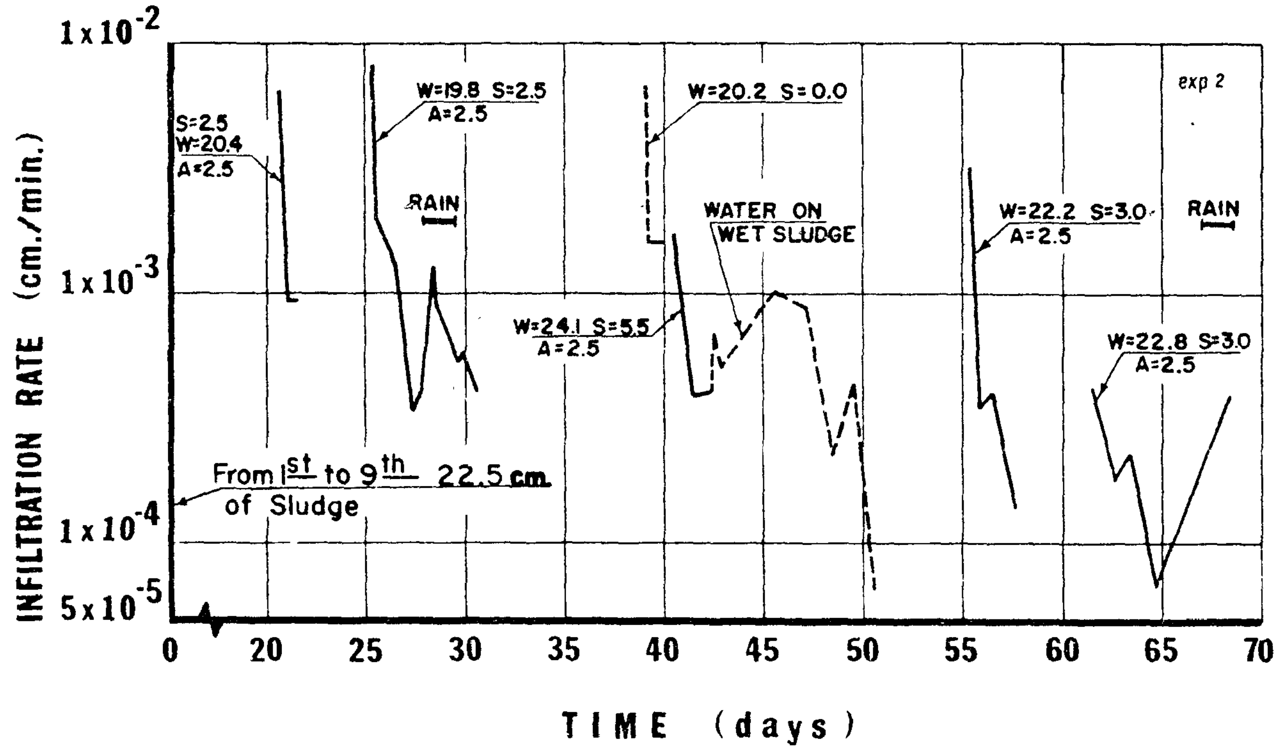


Figure 61. Changes of infiltration rates with time after water and digested sludge applications on Blount silt loam soil which had received an initial high application of sludge and was allowed to dry between each succeeding application.

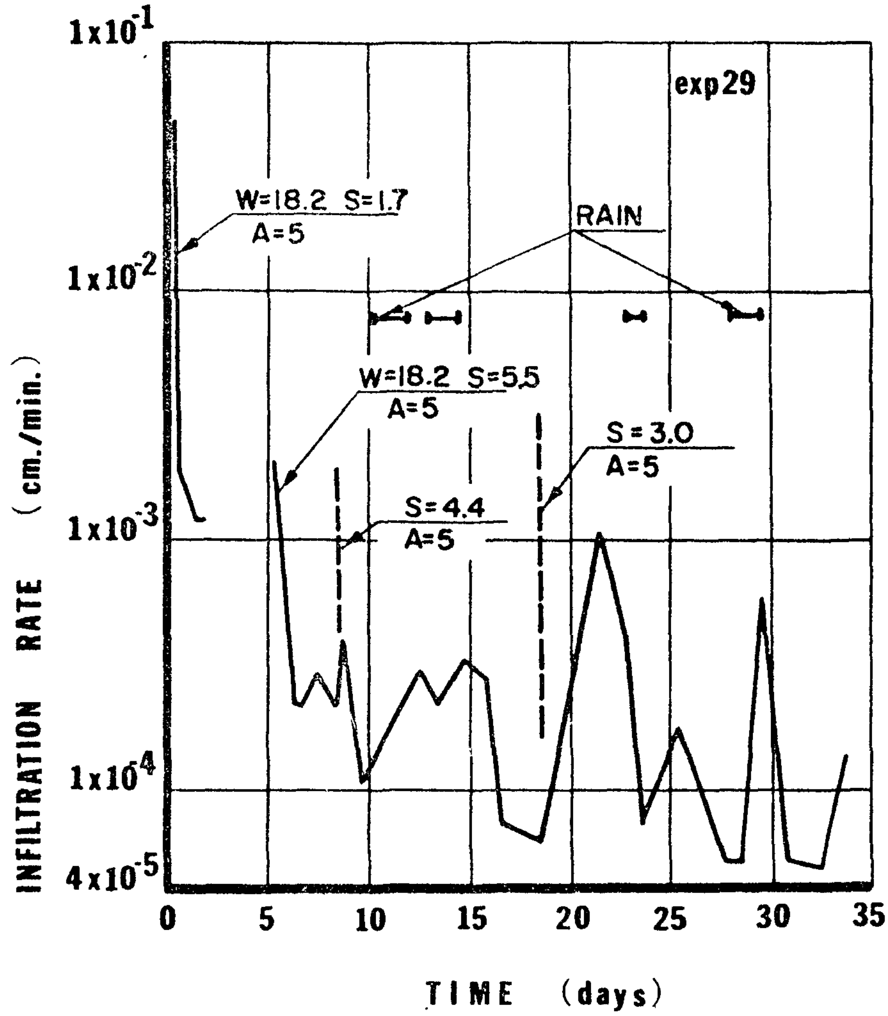


Figure 62. Changes of infiltration rates with time after applying digested sludge on Blount silt loam soil where the sludge residues of a previous treatment were not permitted to dry.

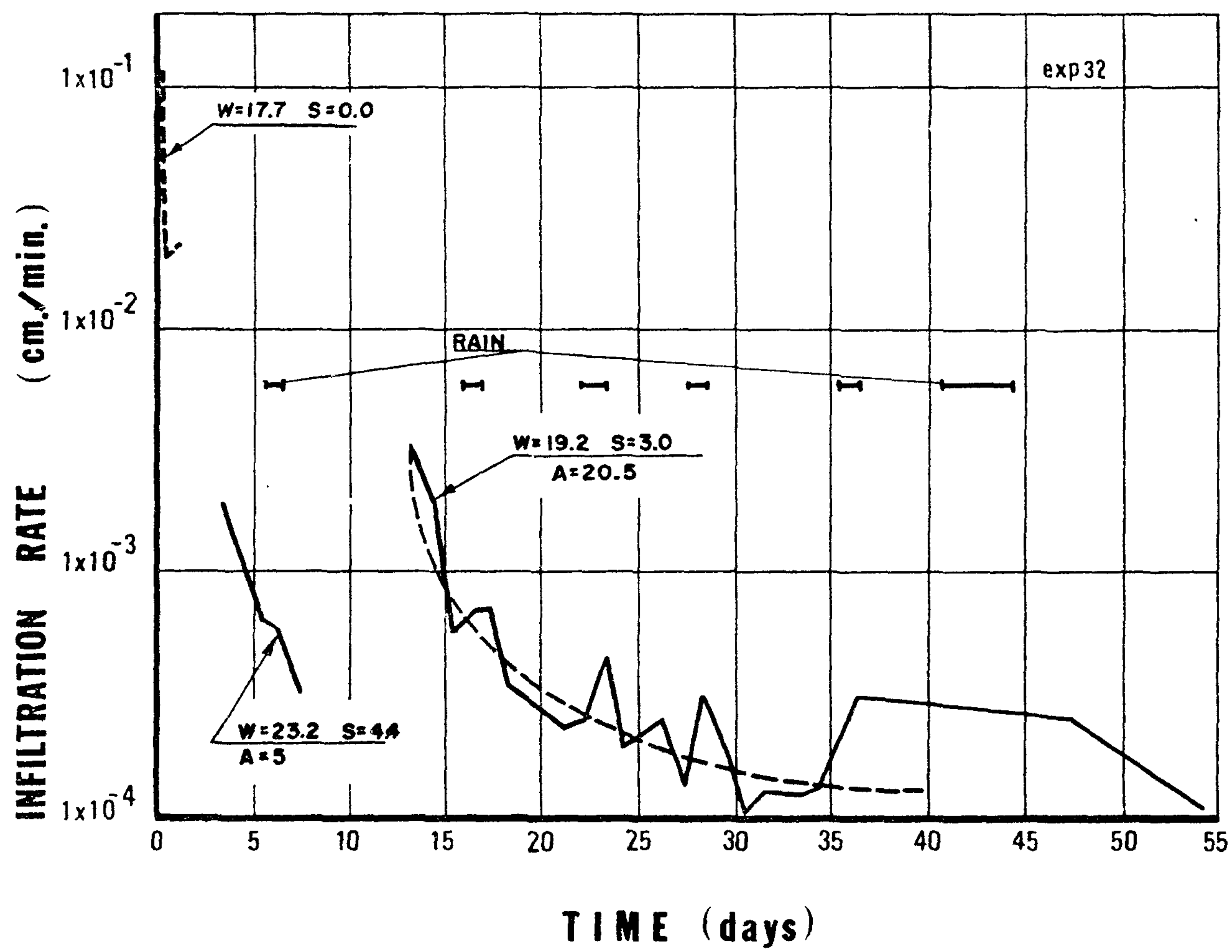


Figure 63. Changes of infiltration rates with time after a large (20 cm) digested sludge application on Blount silt loam soil.

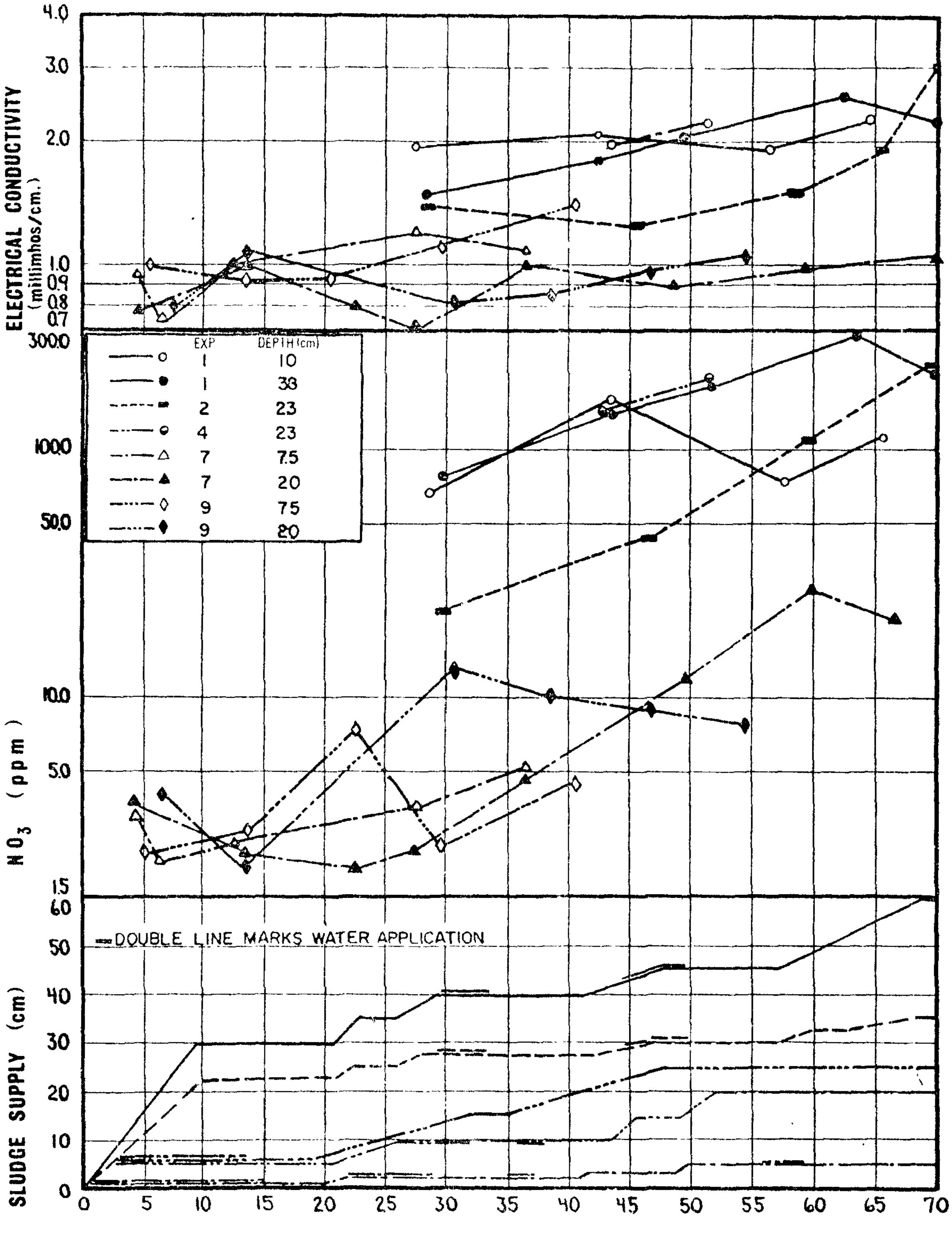


Figure 64. Changes in electrical conductivity values and NO_3 concentrations on the soil solution with time after periodic sludge and water applications on Blount silt loam soil.

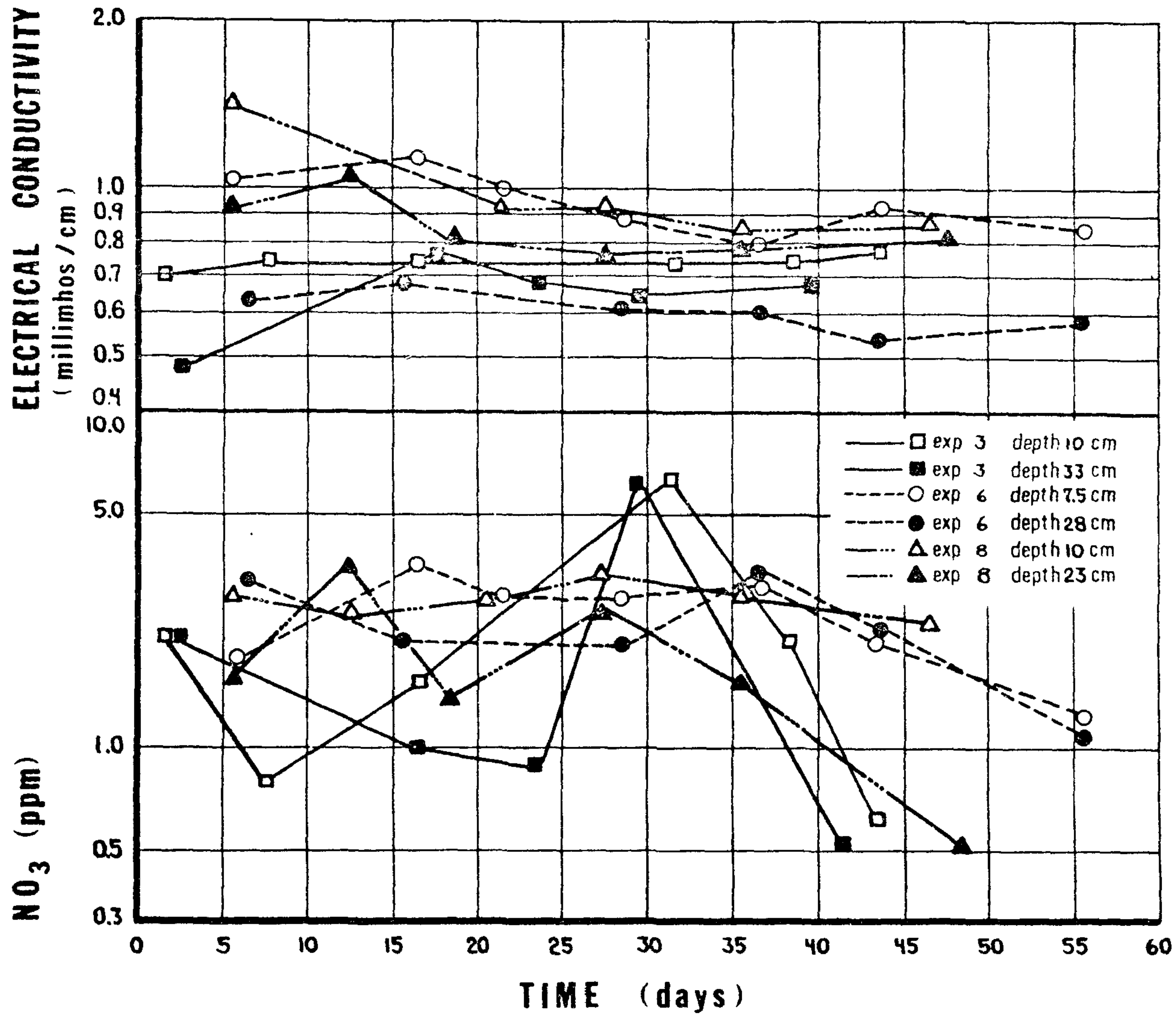


Figure 65. Changes in electrical conductivity values and NO_3 concentrations of soil solutions of Blount silt loam taken from several depths with time.

Summary and conclusions - Factors determining the dewatering rate of digested sludge on soils were investigated under laboratory and field conditions. Soil columns of Blount silt loam and Plainfield sand were used in laboratory sludge dewatering studies. Metal infiltration rings were used for field studies.

Nitrate concentrations, electrical conductivity, and pH and Eh values of effluent and soil water samples were determined. Effluent from soil columns and soil solution samples were collected throughout the period in which the factors influencing the dewatering of sludge on soils were investigated.

When sludge was first applied on soils, dewatering of the sludge was fairly constant and the rate depended on infiltration of water into soils and water losses by evaporation. When the water content of sludge was reduced to about 80 percent by weight, by dewatering on initially moist soils, further drying of the sludge was to a large extent by evaporation alone. Where antecedent soil moisture was low, infiltration contributed to the sludge dewatering process when sludge moisture contents were considerably less than 80 percent by weight. Also on soils where antecedent moisture contents were low and at sludge moisture contents where further dewatering on soils was by evaporation only, the evaporation rate was smaller than from pans.

The rate of infiltration of sludge liquid depended not only on the initial soil moisture content, but also on the solids content of the sludge. The higher the soil moisture and sludge solids contents, the lower was the rate of infiltration. However, antecedent moisture conditions affected the rate of sludge infiltration less on sandy than on fine textured soils.

Initially the infiltration rate of sludge liquid into sand was greater than into silt loam soils. After a few days of successive applications of sludge in the absence of complete drying, the rate was about the same regardless of soil type. It appears that after a period of time the infiltration rate is determined by the sludge cake and not by the soil surface. The soils were unsaturated with respect to moisture and their capacity to transmit moisture was always greater than the infiltration rate determined or controlled by the sludge cake.

When successive sludge applications were made at time intervals such that the sludge cake was not allowed to dry, infiltration rates decreased to very low levels. But if the sludge cake was allowed to dry, the initial infiltration capacity was more or less completely recovered.

Under laboratory conditions, evaporative losses of water from digested sludge in pans were not detected when the moisture content of sludge was reduced to about 8 to 10 percent of the dry weight.

The changes in soil pH and redox potentials were small following various rates and frequencies of sludge applications.

Nitrate nitrogen concentrations continued to increase in the soil with successive sludge applications both in laboratory and field studies. Soil solution nitrate concentrations ranged between 100 and 300 ppm where a total of 50 cm of sludge was applied in 70 days during field studies.

From nitrate concentrations and redox potential measurements, it appeared that anaerobic conditions were seldom, if ever, produced in the soil by exceedingly high sludge loading rates.

Soil conductivity values were increased from an average value of about 1 mmhos/cm where only water applications were made to about 2.5 mmhos/cm where a total of 50 cm of sludge was applied in 70 days. From limited data, it appears that soluble salts will be leached to deeper soil depths in a humic region. During exceptionally dry summers, the salt buildup in the surface of a soil like Blount silt loam, with continuous periodic sludge applications, will exceed that which was found with the total 50 cm application and may cause a plant moisture stress severe enough to reduce crop yields.

SECTION VI

MICROBIOLOGICAL STUDIES

Influence of Soil Moisture on Fecal Coliform Survival

Introduction - A laboratory study of the possible bacterial contamination of the environment resulting from land application of liquid sludge is the subject of a master's thesis prepared by James Schwing, Department of Civil Engineering, University of Illinois. Specifically, the purpose of the study was to evaluate the effect of the moisture content of Plainfield sand on survival of fecal coliforms.

The study was preceded by a review of literature on survival of pathogenic organisms in waste treatment processes including anaerobic digestion and on their survival and movement in soils. Conventional treatment processes achieve significant reductions in the number of intestinal bacteria and appreciable numbers of the organisms are transported to the anaerobic digestion process in sludge. The environment of the digester is unfavorable to pathogenic organisms; however, complete removal cannot be anticipated and land disposal systems must be operated with consideration to the effect of the organisms in the environment. Bacteria can be effectively removed from water leaching through soil - particularly after a build-up of solids occurs in the interstices of the soil. In addition, intestinal bacteria may die off with time in the unfavorable soil environment. Some of the factors which influence the rate of die off include temperature, the level of organic material in the soil, pH, and the moisture content of the soil.

Based on the review of literature, the most critical period for passage of pathogens through the Plainfield sand would be immediately after initiation of a land disposal project. After sludge solids have accumulated in the soil voids and microbiological activity has increased in the soil, removal of pathogens by straining would become more effective and the extent of the bacterial front would retreat to a position closer to the surface of the soil. Long-term changes in the soil structure due to sludge applications would be expected to change the response of pathogens to the soil environment. The increase of organic material in the soil might make the environment more favorable for competitory and predatory soil microorganisms because of the greater retention of moisture and availability of food.

Materials and method - The soil used in these experiments was conditioned by adding a total of 7.6 cm of digested sludge to a 0.6 m deep Plainfield sand column over a period of nine days. The sludge was added 2.54 cm at a time every three days, and following drainage the sludge was worked into the upper 15 to 23 cm depth of soil. Following this conditioning phase the upper 23 cm depth of soil in the column was removed and mixed with an additional 1.3 cm depth of digested sludge and sufficient rain water to adjust the final moisture content to 5 percent. Additional rain water was added to other samples to give moisture levels of 10, 15, and 20 percent. These samples at the four moisture conditions, along with a sample of sludge not mixed into soil, were then monitored for a period of time to observe the rate of disappearance of the fecal coliforms originating in the sludge. Moisture contents were maintained at the initial levels by sealing the samples in containers with "Saran Wrap" covers and by adjusting the moisture content periodically as needed. Samples from the atmosphere overlying the sludge were collected with a syringe and analyzed with a gas chromatograph to assure that the oxygen content remained near normal.

Discussion of results - Results of the study of survival of fecal coliforms in sludge-soil mixture at 5 percent moisture content are shown in Figure 66. The points shown as squares represent the actual values obtained while the circles represent the 95 percent confidence interval. An initial sharp 100-fold increase in the fecal coliform population will be noted. This might be explained by the fact that metabolic intermediates formed in the anaerobic digestion process become available to the fecal coliforms as a food source under aerobic conditions.

Figure 67 shows the survival of fecal coliforms in sludge conditioned soil at 10 percent moisture. Again an initial period of growth is evident. The fecal coliform density was increased by almost 10 fold. The fecal coliform survival in sludge conditioned soil at 15 percent moisture is shown in Figure 68. At this moisture concentration, a 10 fold increase in fecal coliform density is again realized.

Fecal coliform survival at 20 percent moisture in sludge conditioned soil is shown in Figure 69. No initial growth is exhibited by the data for this curve. This is probably due to the fact that at this high moisture concentration the saturation capacity of the soil was exceeded. Essentially anaerobic conditions were maintained as evidenced by the appearance of black sulfide precipitates typically evident with anaerobic conditions. For this reason the organic intermediates of anaerobic digestion were not readily available to the fecal coliforms as a food source.

Data for fecal coliform survival in sludge are presented in Figure 70. Again, as in sludge conditioned soil at 20 percent moisture, no initial phase of growth was realized. The reason for this was again probably

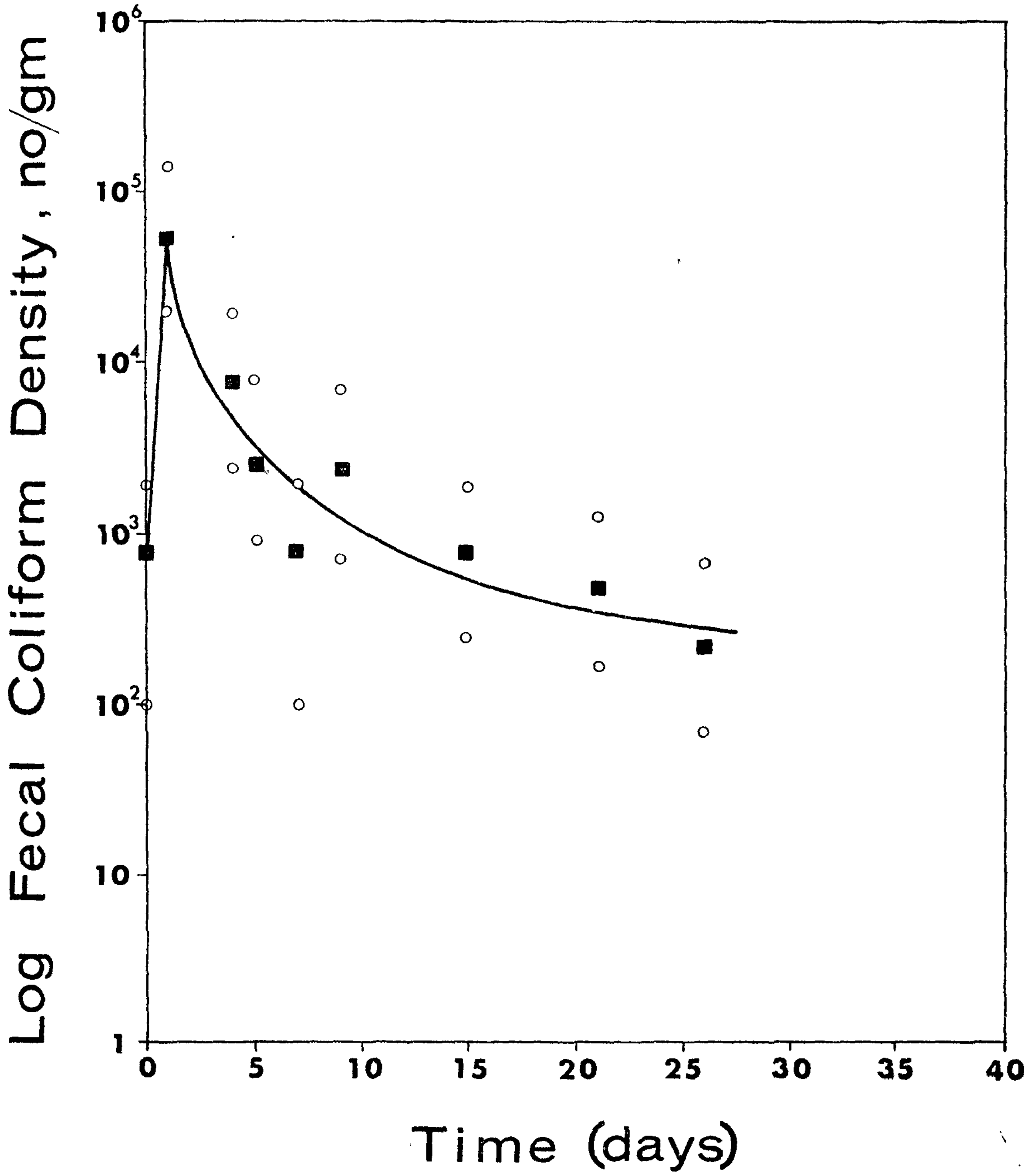


Figure 66. Fecal coliform survival in sludge-conditioned soil adjusted to 5 percent moisture content. Circles represent 95 percent confidence limits.

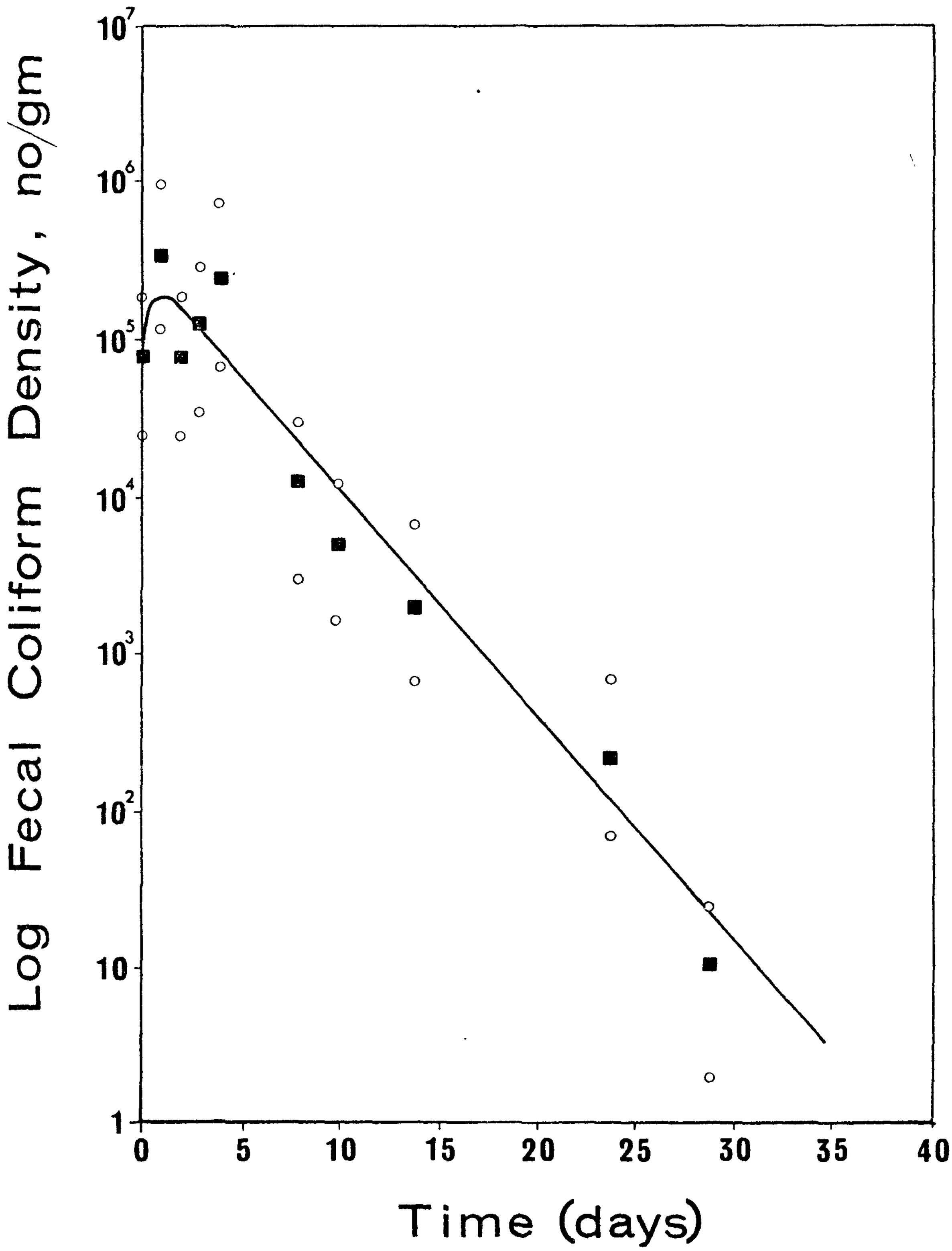


Figure 67. Fecal coliform survival in sludge-conditioned soil adjusted to 10 percent moisture content. Circles represent 95 percent confidence limits.

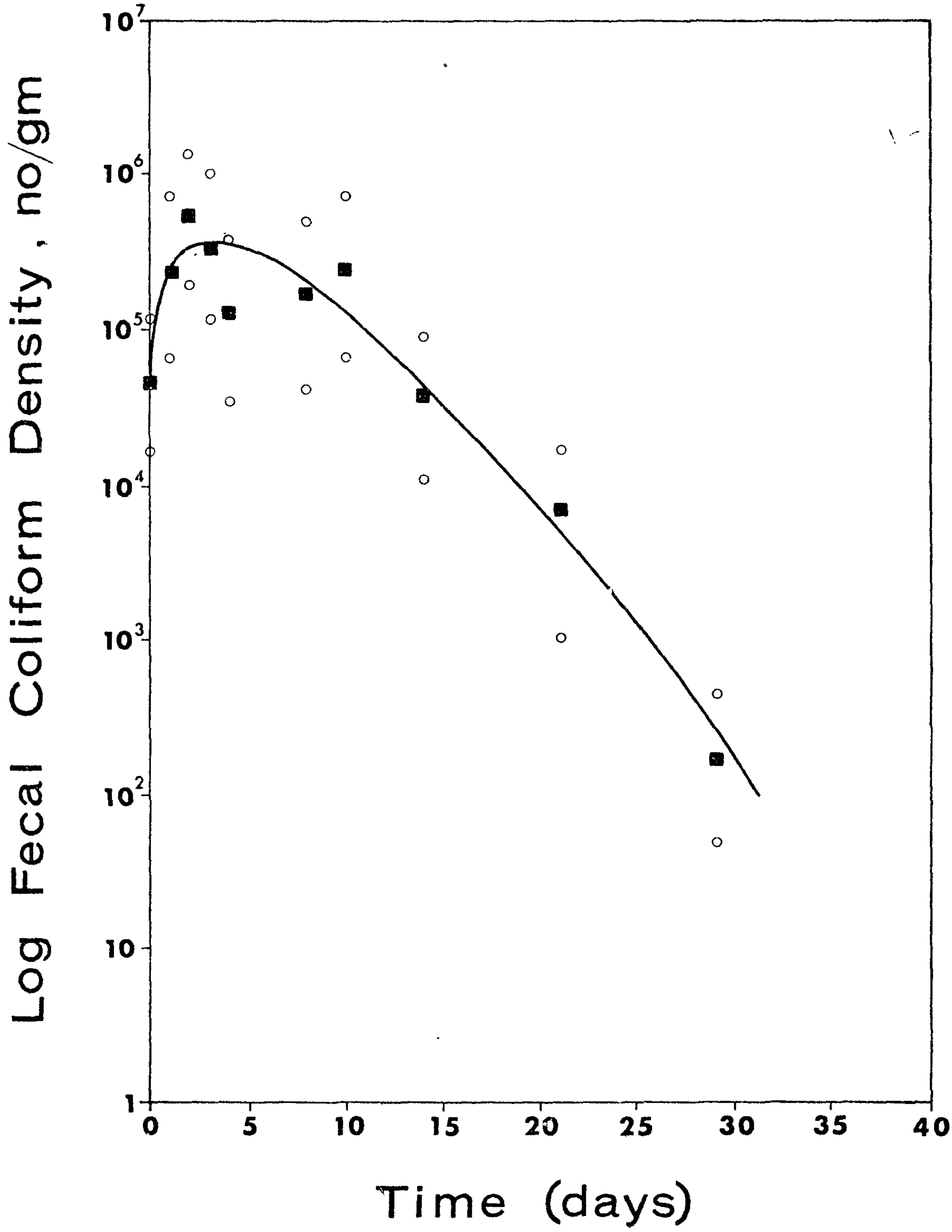


Figure 68. Fecal coliform survival in sludge-conditioned soil adjusted to 15 percent moisture content. Circles represent 95 percent confidence limits.

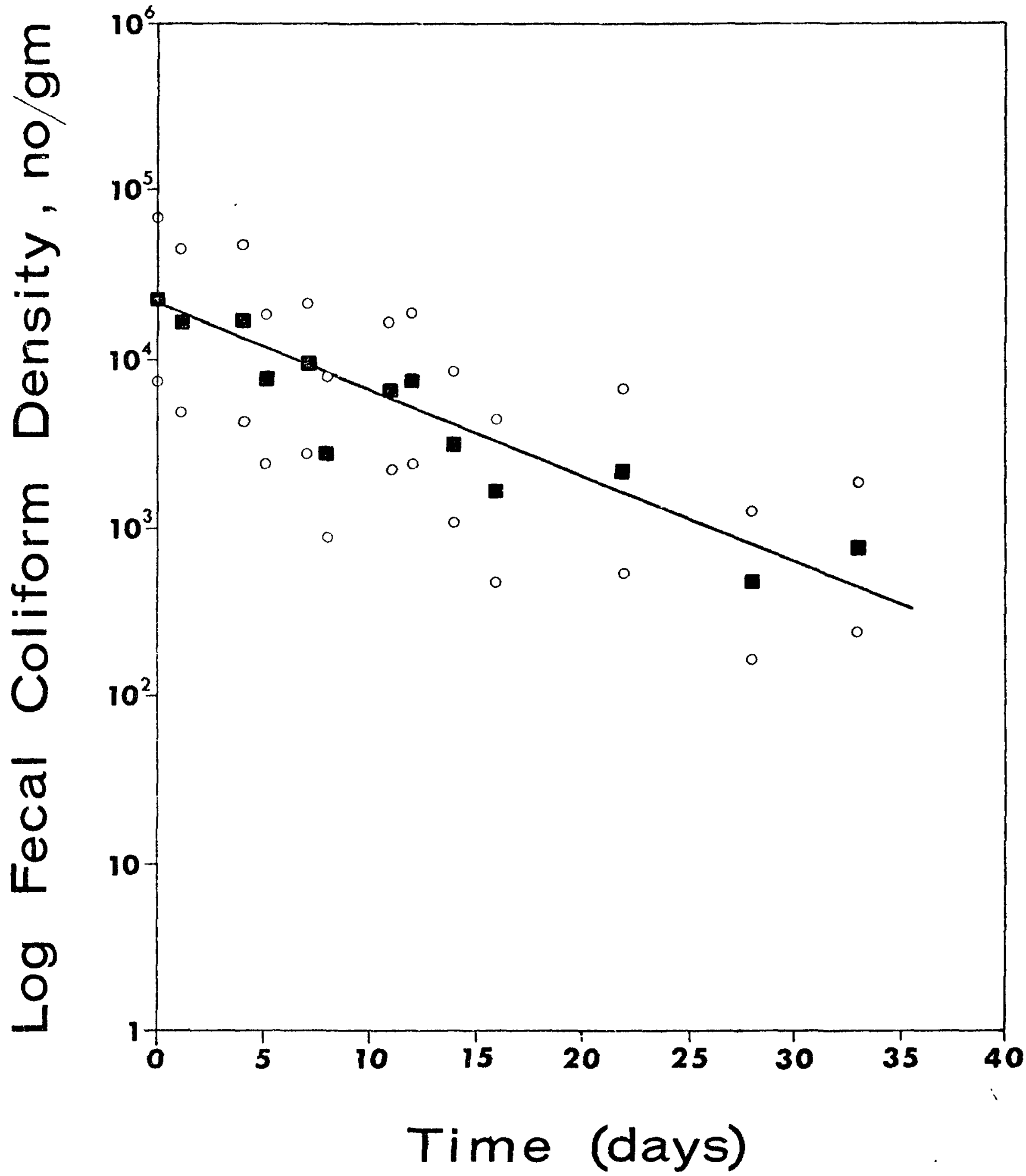


Figure 69. Fecal coliform survival in sludge-conditioned soil adjusted to 20 percent moisture content. Circles represent 95 percent confidence limits.

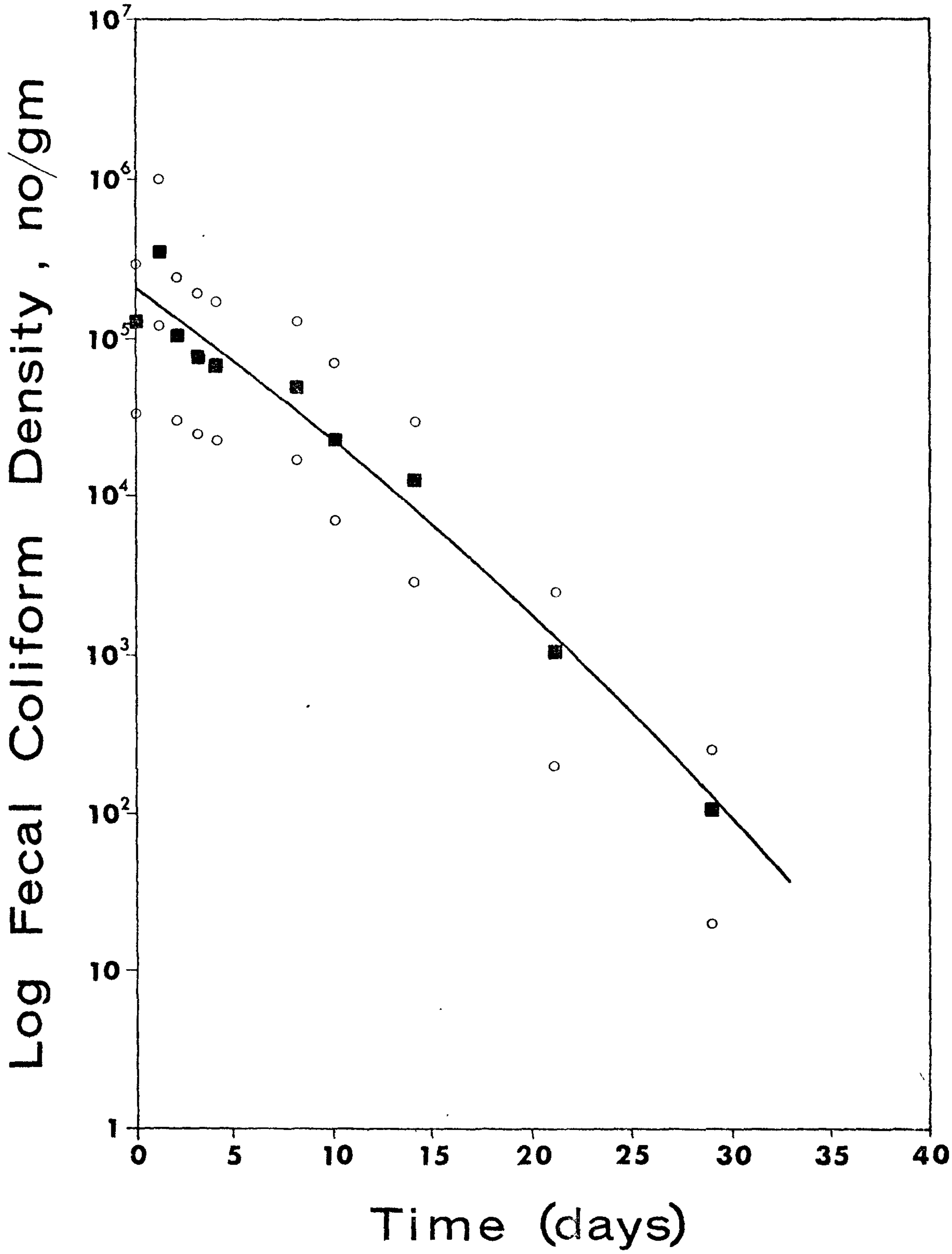


Figure 70. Fecal coliform survival in sludge. Circles represent 95 percent confidence limits.

the maintenance of anaerobic conditions, with the resulting inavailability of the intermediates of anaerobic digestion to the fecal coliforms as a food source.

Table 50 shows the average percentage die off of fecal coliforms after 30 days. The data indicate that at 5 percent moisture condition the fecal coliforms were best able to survive. This is surprising as it might be expected that a higher rate of die off would occur at lower moisture concentrations due to the inavailability of moisture. Perhaps the "aeration porosity limit" as suggested by Bhaumik and Clark (18) for the condition when a favorable balance between moisture concentration and aeration exists is near 5 percent moisture for the conditions of this study.

Table 50. Survival of fecal coliforms in soil and sludge.

Moisture Content (Percent)	Die Off of Average Fecal Coliforms in 30 Days (Percent)
5	72.5
10	99.9
15	99.6
20	96.6
Sludge	99.9

<u>Summary</u> - Following the initial period of growth, die off of fecal coliforms in sludge and in soil containing sludge followed first order kinetics. That is, a constant fraction of remaining organisms died during each time interval. The only exception to this was for the 5 percent moisture condition where the die off rate decreased with time.

Studies of the survival of fecal coliforms in Plainfield sand which had received sludge indicated that the organisms were able to survive for extended periods of time at moisture concentrations from 5 to 20 percent. The rate of die off may be significant from the standpoint of pollution of surface water when precipitation causes runoff soon after the sludge is applied and especially if it causes soil erosion from sludge-treated land. Travel of the fecal coliforms through the soil with leachate was not investigated as a part of this work but, based on reports in the literature, would not be expected to be as severe a problem. Many of the questions regarding the transport of fecal coliform off of the soil surface and through soil will be answered from the results obtained from the NEARC lysimeter studies.

At 5 percent moisture a sizeable increase in fecal coliform numbers occurred after sludge addition and the subsequent rate of die off was slowest. This is of particular interest as 5 percent is probably more representative of the average moisture concentration of the surface layers of the Plainfield sand in the field than the other moisture levels studied. However, in the field rapid changes in soil surface temperatures and exposure to sunlight may exert an over-riding influence on fecal coliform longevity.

Porcine Enterovirus Survival and Anaerobic Sludge Digestion

Introduction - Although, to our knowledge, a disease outbreak has never been known to have resulted from the application of anaerobically digested sludge to land, we really know very little about the virologic aspects of municipal sludges. In light of recent proposals and programs for sludge utilization as a fertilizer on crop lands, additional information is needed on possible virus content and their survival during the anaerobic digestion period.

The following study was undertaken in an attempt to gain some insight into the fate of viruses which cause disease in man when they are subjected to the environmental conditions normally maintained in a heated anaerobic digester. However, to overcome a number of technical problems, an animal enterovirus was used in the study which possesses many characteristics common to human enteroviruses.

Materials and methods - The virus employed in this study was a characterized swine enterovirus designated ECPO-1 (20)(143). The agent was propagated in primary porcine kidney cell cultures prepared in 114 gram prescription bottles employing a medium of 0.5 percent lactalbumin hydrolysate and 10 percent fetal calf serum in Hank's Balanced Salt Solution (HBSS).

When viral induced cellular degeneration was nearly complete and involved approximately 90 percent of the cells, the culture fluid was collected and pooled. A representative sample of the harvest was taken at that time and the titer of the virus determined. The remainder was frozen in 85 ml quantities at -65°C. Just prior to use, sufficient virus stock for the seeding of laboratory scale digesters was thawed and adjusted to yield 100 ml volumes containing 10^6 plaque forming units (PFU) per ml.

The laboratory digesters consisted of six tightly sealed, one liter stainless steel vessels similar in design and operation to those described by Vatthauer *et al*. (159). The initial charge of sludge and inoculum consisted of 900 ml of digester draw-off from the Champaign-Urbana wastewater treatment plant. When in operation each unit was maintained in a 34.5°C water bath and continuously agitated by means of a magnetically driven glass stirring rod. All digesters were monitored daily using gas production and composition, digestion of dry matter, and pH of draw-off. Gas collections were made in metalized plastic bags on each digester and volumes measured by water displacement. Gas composition was determined on an F & M Model 720 gas chromatograph (F & M Scientific Corp., Avondale, Penn.) equipped with a gas sampling valve and thermal conductivity detector. CH_4 and CO_2 were separated using a three-meter long column of 80-100 mesh porapak Q (Waters Associates, Inc., Farmingham, Mass.). Dry matter and organic matter determinations were by standard drying and ashing techniques. The hydrogen ion determinations were made with a Corning model 8 pH meter with a standard combination electrode.

Once in operation, 60 ml of digested sludge were removed from each digester daily through a small stoppered port in the top of each unit and replaced with 60 ml of fresh sewage. On appropriate days the draw-off from each unit was used for viral and other monitoring purposes.

When stabilization of the digesters occurred as determined by gas production, two units were seeded with 100 ml of the virus suspension. Two additional units for comparison remained uninoculated with virus.

All piglets were obtained by hysterectomy and maintained in isolators of stainless steel and flexible plastic design. Equipment, methods of procurement and rearing were as previously described by Meyer et al. (107). All piglets were free of detectable microbes, and at least 10 days old when first exposed to sludge.

Fecal samples were collected from each pig twice a day (AM and PM) on the 3rd and 4th days post-challenge and pooled. Twenty percent fecal suspensions were prepared for inoculation using complete tissue culture medium with antibiotics as diluent and filtered through a microsyringe filter holder containing a millipore HA filter. All cultures once inoculated were examined daily for viral cytopathic effect and held for one week before discarded as negative. In those cases where viruses were recovered from infected piglets, the agents were identified as ECPO-1 by neutralization with specific antiserum.

Three separate trials, approximately six weeks apart were carried out employing piglets of different genetic backgrounds. In the 1st trial, 25 ml samples of sludge were collected from each of the two virus seeded digesters and pooled. Such samples were obtained after 30 minutes, 1 day, 4 days, 7 days, and 12 days. The pooled samples of each time interval were mixed well, antibiotics (penicillin and dihydrostreptomycin) added and 10 ml volumes force fed to two piglets.

In the 2nd trial the general procedures were the same as above except that samples were withdrawn from the digesters after 1 hour, 1 day, 2 days, 3 days, and 4 days; and that the volume fed to the piglets was increased to 20 ml, 10 ml being fed orally and the remaining 10 ml added to a small quantity of sterile milk.

In the 3rd trial the procedure was the same as in the 2nd trial except the samples were collected after 1 hour, 4 days, 5 days, and 6 days.

Results - Generally a period of approximately four days was required for the digesters to stabilize. Data collected during the individual trials indicated comparable and uniform rates for CH_4 production and the other parameters monitored for units with or without virus except that units shortly after the addition of virus showed marked but temporary increases in CO_2 production compared to control digesters.

Virus could not be detected or demonstrated by pig infection after the 4th day of exposure to the environment of the anaerobic digesters. All pigs challenged with virus material exposed in digesters 72 hours or less became infected and 2 to 6 became infected with material withdrawn on the 4th day. As shown in Table 51, none of the piglets were infected with material which had been in the digesters 5 or more days.

Table 51. Survival of swine enterovirus ECPO-1 in anaerobic sludge digesters.

Trial	Time Virus Exposed in Digester								
	1 hr*	1 day	2 days	3 days	4 days	5 days	6 days	7 days	12 days
I	+	+			+			0	0
	+	+			0			0	0
II	+	+	+	+	0				
	+	+	+	+	+				
III	+				0	0	0		
	+				0	0	0		

* Trial I 30 min
\+ Infected piglet
0 Non-infected piglet

While emphasis was on recovery of virus from piglets exposed to sludge, it should be noted that in trial III, six of the eight piglets exposed to sludge at that time contracted a bacterial infection and succumbed to the bacterial pathogen Salmonella typhimurium.

Discussion - Raw sewage contains a wide variety of viruses and a significant number of human origin. Little or no information, however, exists on their isolation from digested sludge and the few attempts recorded in the literature were unsuccessful.

Because viruses may be readily absorbed onto the surface of sludge particles resulting in rather stable virus-sludge complexes, they are difficult to isolate. As a result microbiologists have been reluctant to say that the failure to isolate viruses was due to their inactivation by the digestive process *per se*.

The decision to use germ-free swine as an indicator animal for detecting infectious levels of virus stems from a number of factors. First a characterized swine enterovirus was available which possessed biophysical properties similar to agents commonly encountered in sewage. Second, ECPO-1 had various desirable attributes such as its eas of cultivation, identification and a minimal disease potential for man. Third, early preliminary studies indicated that available sludge was inherently toxic to our cell culture system and that direct isolation by cell culture inoculations would be of limited value.

It should be recognized that in the operation of the digesters a dilution of the virus occurred with the daily draw-off and the addition of 60 ml of fresh sewage. With a turnover rate of approximately 1/15 of the total volume per day, one could anticipate the removal of 1/2 of the original virus inoculum after 7 1/2 days of operation.

To help compensate for the loss of virus, as a normal consequence of the daily feeding of the digesters, we arbitrarily started out with what we thought was a high concentration and one not likely to be encountered in a conventional sludge (10^5 PFU). In addition, we increased the amount of sludge fed to each pig form 10 ml to 20 ml during trials II and III.

Considerably more work will naturally be required before the virologic aspects of digested sludge will be completely known. In the case of the Salmonella typhimurium infection that occurred in trial III, it appears that all facets relative to the presence of bacterial pathogens may also need additional consideration. Even so, it would appear, at least in this limited study and circumstances provided in these experiments, that viruses with characteristics, similar to ECPO-1, whether complexed with sludge or not, would not appear to constitute a serious infectious hazard after five days in digesters of the type employed.

Conclusions - Although additional work on a wide variety of viruses may be required to ascertain the effectiveness of anaerobic digestion as a means to inactivate viruses, preliminary results indicate a reduction and loss of infectivity could be expected upon a suitable five days exposure to the environmental conditions provided by anaerobic digestion of municipal sewage sludge. Considering that sewage solids are held in heated anaerobic digesters for periods of time several times greater than needed to inactivate the swine virus used in this study, it appears that the use of digested sludge as a fertilizer presents very little risk with regard to spreading diseases caused by viruses.

Hygienic Aspects of Liquid Digested Sludge

Fecal coliform die off studies - For routine analyses of water, soil and digested sludge samples, the coliform group of bacteria has been taken as an indicator of the degree of microbial pollution. Since non-fecal coliforms are known to be part of the normal soil flora, only the fecal coliforms have been considered for use in the studies discussed here.

The determination of fecal coliforms is rapidly and easily performed by the membrane filter technique with incubation at 44.5°C in the M-FC medium (54). This method (referred to as MFC) reveals the presence in the liquid digested sludge of a large population of fecal coliforms which gradually decreases upon removal of the sludge from the digester. A reduction from 4×10^4 to 7×10^3 and 2×10^2 fecal coliforms per ml was observed after 19 and 32 days, as can be seen from the data presented in Table 52. A similar die off can be observed for the fecal coliform organisms contained in sludge supernatant. In contrast to the gradual decrease or die off of fecal coliform organisms originally present in digested sludge, when laboratory grown populations of *Escherichia coli* (neotype, ATCC 11775) are added to non-treated or autoclaved digested sludge they die off very rapidly, as evident by the data presented in Table 53. In view of this difference of behavior, the question is raised as to whether the organisms found in the digested sludge by the MFC technique are truly of the fecal coliform group. Short of serological tagging, the IMViC test and the elevated temperature MPN-EC (most probable number - EC medium) method, as a confirmatory test from positive presumptive tubes, are the only other two ways to identify fecal coliforms. Both techniques have indicated the presence of fecal coliforms in the liquid digested sludge. From the data presented in Table 54 it can be seen that agreement in fecal coliform counts was obtained with the MFC and MPN-EC methods. Furthermore, on the basis of the IMViC test, Fuller and Litsky (49) have shown that digested sludge harbors a population of fecal coliforms, which is on the order of 10^5 cells per milliliter.

Table 52. Number of fecal coliforms per ml of impounded liquid digested sludge.

Sludge Sample	Days 0	19	32
Total sludge	4×10^4	7×10^3	2×10^2
Sludge supernatant	3×10^3	2×10^1	0

Table 53. Bactericidal properties of digested sludge toward laboratory-grown *Escherichia coli* as determined by the membrane filter-high temperature method.

	Fecal coliform, cells/ml		
Incubation, hr	Digested sludge	Digested sludge supplemented with E. coli	Autoclaved digested sludge plus E. coli
0	25×10^2	25×10^6	26×10^6
24	20×10^2	41×10^2	<10

Table 54. Comparison between the membrane filter-high temperature (MFC) and the MPN-EC medium techniques for counting fecal coliforms.

	Fecal coliform, cells/ml	
Source of digested sludge	MPN-EC medium 95% confidence limit	MFC
Chicago, Calumet Plant	$4.9 \times 10^3 - 4.2 \times 10^2$	1.6×10^3
Urbana-Champaign	$1.5 \times 10^5 - 1.2 \times 10^4$	1.6×10^5
Urbana-Champaign	$3.4 \times 10^5 - 3.7 \times 10^2$	8.0×10^3

The bacteriolytic effect on the *E. coli* of stock culture may have been a function of the bacterial strain. However, the phenomenon assumed a higher significance when it was discovered that digested sludge-adapted fecal coliforms could be transformed into susceptible strains. Specifically, eight distinctive fecal coliform strains, isolated from digested sludge, were maintained in lactose broth. Following one transfer in the broth, six strains survived reintroduction into autoclaved digested sludge, but two were killed. After several transfers covering a two-month period, all isolates no longer adapted to the digested sludge environment and thus behaved like *E. coli* ATCC 11775. The above results prompted further study of the bactericidal properties.

Various treatments were performed on the digested sludge to determine the nature of its apparent toxicity toward *E. coli*, as determined by the MFC technique. Results of these studies are presented in Table 55. Although the digested sludges were always collected at the same wastewater treatment plant (Calumet, Chicago) a few batches turned out to be devoid of toxicity. This fact rules out many factors which otherwise would have been considered as possible causative agents for the toxicity: the low redox potential, the lack of oxygen, the saturation of the sludge liquid phase with carbon dioxide, methane, and possibly the presence of sulfides. Since toxicity was not eliminated by heat sterilization, the bacteriolytic effect could not be attributed to a protein, parasitic relationship, or nutritional competition with other organisms. The bactericidal properties were localized in the liquid phase of digested sludge (Table 55) thus eliminating several of their possible sources. Most components of digested sludge occur in the solid phase. The liquid phase, for example, contains less than 0.05 ppm sulfide, less than 10 ppm organic carbon exclusive of methane and carbon dioxide, and only traces of heavy metals. Because methane and CO_2 saturation of the liquid phase, presence of bicarbonate and ammonium ions (up to 500 ppm-N) at pH values 7.0 to 8.6, low redox potential, and lack of oxygen are properties common to both bacteriotoxic and non-bacteriotoxic sludge samples, they could not be considered as the principle causative factor.

Heat-sterilized sludge and its liquid phase were assayed for antibiotics by the diffusion technique in nutrient agar (Difco). Incubation was carried out aerobically at 27°C. Under these conditions, no antibiotic activity was detectable.

Volatile fatty acids have been held responsible for the exclusion of *E. coli* and salmonellas in the rumen of bovines by Hollowell and Wolin (72). However, their range of bacteriostatic and bacteriolytic action is limited to pH values below 7.0 and to concentrations above 60 μmoles/ml; conditions which are not prevalent in digested sludge. Moreover, Brounlie and Grau (24) presented evidence that the elimination of salmonellas and *E. coli* from bovine rumen cannot be accounted for by volatile fatty acids alone.

Table 55. Localization of bactericidal properties in digested sludge liquid phase*

Medium	Escherichia coli after 24 hr incubation, cells/ml
Saline solution, 1.0%	3×10^7
Digested sludge, autoclaved	10^5
Liquid phase, autoclaved	10^5
Liquid phase, nonautoclaved	10^5
Precipitate, resuspended with liquid phase, autoclaved	10^5
Precipitate, resuspended with distilled water, autoclaved	11×10^7

* Liquid phase obtained by two successive centrifugations; the first one at 5000 g for 20 min; the second one done at 30,000 g for 60 min on the supernatant liquor from the first centrifugation. Both precipitates were resuspended to original volume with either distilled water of supernatant liquor.

Langley et al. (86) upheld the view that elimination of S. typhosa is not caused by toxic compounds, but rather results from a nutritional deficiency which can be satisfied by additions of tryptophane. We found that with E. coli, reversal of the toxicity could be achieved by addition of 5 g/l tryptone (Difco) to the digested sludge. This amount was in excess of the usual nutritional needs. At 2.5 g/l, tryptone did not prevent die off of E. coli. The energy and growth factors brought with tryptone could not be replaced by lactose and/or yeast extract (Difco). Deficiencies in oxidizable organic material and an essential nutrient do not create a toxic medium. For example, the same fecal coliform population which rapidly died off in digested sludge liquor remained stationary in physiological saline solution.

By way of summarizing the findings up to this point it may be said that fecal coliforms aerobically grown in a lactose broth and transferred to the sludge, die off more rapidly if obtained from stock cultures than if freshly isolated from the sludge. This resistance of sludge isolates disappears after several transfers in a lactose broth. The bactericidal properties are localized in the liquid phase of the sludge, but no antibiotics are detected by the diffusion agar method. The boiling of the sludge destroys the bactericidal properties, autoclaving does not. The lack of oxygen and the low oxido-reduction potential of the sludge are not solely responsible for the killing. Reversal of the bactericidal action is obtained by the addition to the sludge of high concentrations (5 gm/liter) of tryptone (Difco). At the same concentrations, tryptophane and tyrosine reduce the death rate slightly. Lactose or yeast extract, or both, have no effect. Digested sludge made from tryptone as sole source of carbon is toxic; however, sludge made from butyrate is not.

Effect of heavy metals on fecal coliform organisms - Differences in die off rates of *E. coli* in digested sludge as measured by the pour plate and MFC procedures coupled with the characteristics of the toxicity previously discussed suggested that heavy metals should be investigated as a source of bacteriolytic behavior. The pour plate technique gave higher fecal coliform counts and a lower rate of die off than the MFC method. Shipe and Fields (143) had similar results with *E. coli* cell which had been suspended in zinc or copper sulfate solutions. They assumed that either toxic metals were concentrated on the membrane surface or that some cells weakened by the metals could no longer form colonies on the membrane. Nearly all of the heavy metals present in digested sludge occur in the solid phase. Analyses of centrifuged sludge liquor showed 0.057 to 0.10 ppm Cu, nondetectable to 0.10 ppm Ni, and 0.075 to 0.15 ppm Zn. These concentrations were lower than those which were shown to cause a reduction of 5 percent or less in digester efficiency (Public Health Service, 1965). Cadmium and Cr were not detected in the liquid phase although they were present in the solids. Even at low concentrations, metals can be toxic to *E. coli*. For example, Malaney, *et al*. (99) reported that as little as 0.3 ppm Cu or 0.5 ppm Cd affects the metabolism of *E. coli*. Moreover, sublethal concentrations of several metals can accumulate to toxic levels.

An objection to assigning the observed toxicity to heavy metals could be raised because of precipitation of metals by carbonate, hydroxide, and phosphate anions in the liquid phase of sludge. To check the effect, 2×10^4 cells/ml of *E. coli* ATCC 11775, as determined by the pour plate technique with nutrient agar, were suspended in buffered and unbuffered Cu solution. The buffered solution was composed of 10^{-2}M $CuSO_4 \cdot 5H_2O$ and 0.1 M phosphate at pH 7.0. Based on the solubility product of Copper phosphate ($10^{-36.7}$), the theoretical solubility of Cu in the buffered solution was $10^{-11.6}$ M. The concentration of Cu in the unbuffered solution was 10^{-5} M (0.83 ppm). As expected, within 12 hours the *E. coli* cells were killed in unbuffered solution, but survived in buffered solution. One mechanism for metals to enter solution in digested sludge is through chelation with naturally occurring organic compounds. In this form they might retain toxic properties. To test this possibility, a model system was developed with Cu and protocatechuic acid as a complexing agent. It can be seen from the results presented in Table 56 that when the Cu salt and protocatechuic acid were present in equimoler quantities, the die off rate was approximately proportional to the Cu concentration. Neither the buffer-Cu nor the buffer-chelate solution was toxic. Complexation of Cu and protocatechuic acid apparently took place increasing the Cu toxicity. Chelation of metals is possible in digested sludge. The digestion process depends on a large population of viable microorganisms. If heavy metals in solution caused toxicity as was observed with *E. coli*, digestion itself would probably have been severely inhibited unless the microbial population in the digester had acquired a high level of tolerance toward chelated metals. An observation of such a tolerance was reported by Malaney *et al.* (99). Within six days or less after the addition of as much as 50 ppm Zn, 20 ppm Cu, or 16 ppm Ni, a microbial population from sewage recovered 35 to 50% of its original activity. They described the toxicity as inhibitory, not lethal.

Table 56. Toxicity of chelated copper toward *Escherichia coli*

	Escherichia coli, cells/ml[a]			
Incubation, hr	$CuSO_4 \cdot 5H_2O$, protocatechuic acid molar ratio[b]			
	$10^{-3c}/10^{-3}$	$10^{-4c}/10^{-4}$	$10^{-3}/0$	$0/10^{-3}$
0	53×10^2	54×10^2	77×10^2	62×10^2
4	32×10^2	42×10^2	119×10^2	56×10^2
11	0	35×10^2	79×10^2	55×10^2
48	0	20	137×10^2	31×10^2

a Counts made by the pour plate technique with nutrient agar.

b Solutions made in 0.1 M phosphate buffer pH 7.0, heat-sterilized.

c Protocatechuic acid added only after copper solution in phosphate buffer had stood at room temperature for 12 hr.

Fecal coliform survival on soils and in water - The behavior of the sludge fecal coliforms as determined by the MFC technique has been examined under various environmental conditions. A gradual decrease of the fecal coliform population was observed in the sludge cake which develops on a soil surface amended with digested sludge, can be seen from the results presented earlier in Table 5. These results are in agreement with those already obtained from various studies done on the behavior of fecal coliforms and *E. coli* in digested sludge, water and soil samples (40)(86)(156).

Routine analyses for fecal coliform densities have been performed on drainage and runoff water samples originating from the Northeast Agronomy Research Center lysimeters. From an analysis of the data collected the sandy soils (Plainfield) have performed as expected, i.e. no fecal coliforms were detected in tile drainage waters. However, drainage water samples from many Blount and Elliott plots were sometimes higher in fecal coliform counts than expected.

Samples collected in the spring were generally low in fecal coliforms. That situation was probably a reflection of the fact that only four sludge applications were made during the spring sampling period. As expected, surface runoff samples were generally contaminated with fecal coliform throughout most of the sampling period. The relatively high contamination in tile drainage water can only be explained by contamination through cracks or animal holes in the soil. It was also noted that as the soil temperature decreased, fecal coliform contamination increased. Increased longevity of fecal coliforms in cool soil and accumulating sludge residues probably accounted for the increase.

Runoff water from the check plots was almost as consistently contaminated as water from the plots with maximum treatment. No specific explanation for this pehnomenon can be given. Warm-blooded animals all excrete fecal organisms. Gophers, ground squirrels, deer, and birds frequent the plot area and may be responsible for the phenomenon.

The reader who would like to find general considerations on the hygienic aspects of sludge disposal on land is referred to other publications such as those by Gordon (56) and Hanks (62).

Microbiological purification of polluted waters by percolation through artificial filters or soils is known to be an effective method of water treatment. Insofar as inferences can be made from traditions and experiences, one may expect the percolated waters from a biofilter four to five feet thick to be free of pathogens. In the present case, the challenge is at the soil-atmosphere interface, where digested sludge will cover acres and be accessible to runoff waters, insects, birds, and animals. The danger of infection from these fields will, to a great extent, be controlled by the persistence of pathogens at the soil surface.

SECTION VII

GREENHOUSE STUDIES

The Effect of Heavy Metals on Nutrient Uptake and Growth of Corn

Introduction - This study on the effect of Pb, Cu, Cr, Zn, and Ni on nutrient uptake and growth of corn was prompted by their presence in relatively large concentrations in digested sewage sludge.

Rohde (130) found toxic levels of Zn and Cu on soils treated with sewage for many years near Paris, France. Lunt (96) found Zn and Cu toxicity symptoms in a few vegetable crops following additions of acidic sludges. Build-up seems likely in the upper soil horizons of most of the heavy metals added by sludge, since studies on heavy metal retention in soils have shown that they tend to accumulate in upper horizons rather than leaching (85). Sludge has also been known to acidify soils. This further enhances the possibility of available toxic amounts of heavy metals in soils, since the heavy metals in question become more soluble as pH decreases (51).

Although Pb, Cr and Ni have not been studied in connection with sludge fertilization of soils, other studies have indicated a number of potential problems due to excess levels in soil. Lead has not been found to be toxic, at least, to deep-rooted crops (79)(80). It, along with the other four heavy metals investigated here, does reduce Fe uptake by plants, however. Chromium has been found to be toxic in some serpentine soils, sand cultures, and nutrient solutions (144). Chromium and Cu also interfere with P uptake. Nickel can be even more toxic than either Cu or Zn at similar concentrations (38)(41)(60)(66)(123)(127).

Experimental procedure - Lead, Zn, Cu, Cr, and Ni were applied to Plainfield fine loamy sand as chemical salts at rates corresponding to quantities added from 15 cm per year sludge applications for 5, 10, 15, and 20 years. That is, rates of the added metals were equivalent to the quantities that would be applied in 75, 150, 225, and 300 cm of sludge.

Concentration levels of heavy metals in sludge assumed here are given in Table 57. It is realized that this technique over-emphasized the actual situation that would occur under field conditions. However, the intent was to simulate the highest possible concentrations of metals that might occur from the respective sludge rates. Thus, metal additions were made on the assumptions that all metals in the sludge would be released from organic form, and that the metal salts would be retained in the upper soil horizon. The authors are aware that under actual conditions the organic material would only partially decompose and that salts would tend to leach from the upper horizon.

Table 57. Heavy metal concentrations in sludge from the Calumet sewage plant.

Element	kg/ha-cm
Copper	2.42
Zinc	9.14
Lead	9.14
Chromium	5.32
Nickel	0.18

Each heavy metal was tested for its individual effect on corn growth. No interactions between the heavy metals were studied. Each treatment was replicated three times in a randomized complete block design.

Some treatments were tested on an Elliott silt loam soil type for Pb, Zn, Cu, and Cr. These treatments were replicated in the same manner as above.

The heavy metals were applied as the following chemical salts: lead acetate, zinc sulfate, cupric sulfate, chromic acetate, and nickel sulfate.

Each greenhouse pot contained 3000 grams of soil to which 200 mg of nitrogen was added as ammonium sulfate, 200 mg of phosphorus as monocalcium phosphate, and 200 mg of potassium as potassium chloride. The "pots" were number 10 cans which were lined with polyethylene bags to prevent rust from contaminating the soil. Ten kernels of corn were planted in each pot, and the stand thinned to eight plants after germination and emergence. The corn plants were allowed to grow for four weeks before harvesting. They were cut off just above the soil. Oven-dry weight was used to determine stover yield.

Zinc, Fe, Mn, P, and Cu were determined in the plant tissue by emission spectrograph. Lead, Cr and Ni were determined in the plant tissue by atomic absorption.

Regression analysis was used to determine significant effects on corn growth and its chemical content. When a decrease or increase in yield or chemical content is declared significant, it will be so at the 95 percent confidence level or higher. All data given in tables and figures are mean values.

Yield of elements was calculated by multiplying concentration levels in corn stover by oven-dry yield of corn stover.

Results and discussion - The various Pb treatments affected corn growth in an erratic manner on the Plainfield sand (Figure 71). Regression analysis of the yield data failed to show any significant effect on corn yield by adding Pb at rates up to 1224 ppm (twenty years accumulation) to either the sand or the silt loam. However, the trend with the Elliott silt loam soil appears to be decreasing yield with increasing Pb rate. Keaton's (80) data on barley growth showed no general trend as rates of Pb were increased. His highest treatment was 2785 ppm of Pb as lead carbonate. At most rates, yields of barley tops were slightly higher than the control. Scharrer and Schropp (134) found very little effect on plant growth when Pb was added to soils as lead acetate. They also reported some growth stimulation from Pb had no toxic effect on deep-rooted crops. Jones and Hatch (79) reported that Pb had no toxic effect on deep-rooted crops. Yet, they found some damage when some shallow-rooted vegetable crops were planted in Pb-treated soils.

Lead was not present in the aerial portions of the corn except in trace amounts (Table 58). This is in fair agreement with Keaton's (80) work, although he was able to show detectable concentrations of about 2-3 ppm in barley tops. His concentrations may be nothing more than an artifact due to incomplete washing, since the concentrations were so small and constant with all rates of lead. He found very high concentrations in root samples though. Jones and Hatch (79) rarely found concentrations of lead over 10 ppm in the vegetable and legume crops they analyzed. They did not find, however, concentrations in roots of their plants as great as Keaton had reported. Lead concentrations in root samples were not determined in our experiment.

A significant linear decrease in Fe content in corn stover occurred as Pb rates increased on the Plainfield fine loamy sand (Table 58). There was no effect on iron content in the stover grown on the Elliott silt loam treated with Pb. A highly significant linear decrease in P content of the corn tissue occurred for both soils as lead rates increased. None of the other elements reported in Table 58 were significantly affected by Pb.

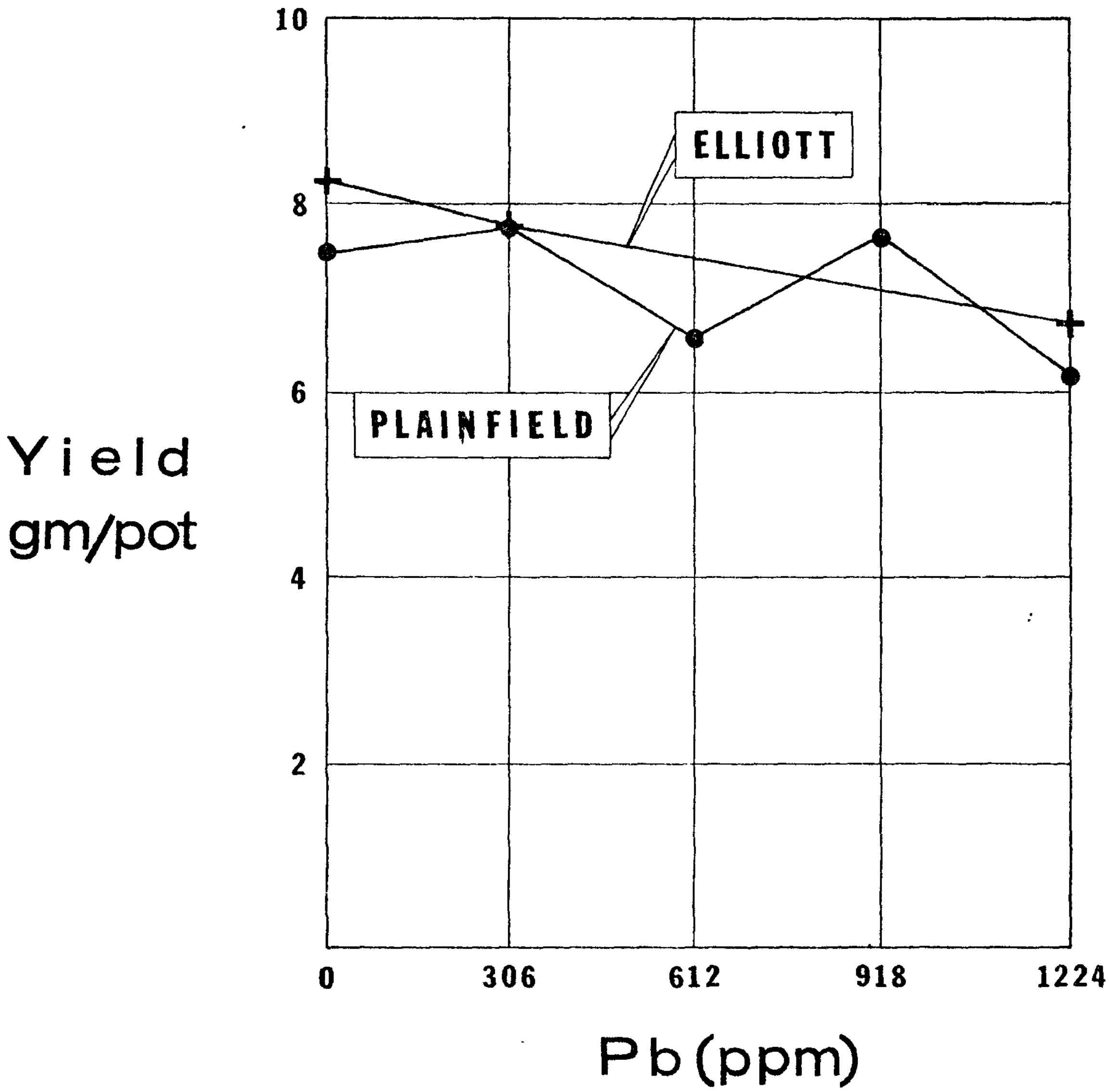


Figure 71. Yield (oven-dry weight) of corn at four weeks in the presence of Pb admixed with Elliott silt loam and Plainfield sand.

Table 58. Heavy metal and P content of corn stover as influenced by Pb, Cu, Cr, Zn, and Ni additions to soils.

Treatments		Concentrations in corn stover					
Element	ppm	Zn ppm	Fe ppm	Mn ppm	P %	Cu ppm	Element varied ppm
				Plainfield			Pb
Pb	0	110	129	239	.62	13	trace
Pb	306	141	149	205	.48	15	trace
Pb	612	139	123	186	.39	14	trace
Pb	918	91	83	144	.26	11	trace
Pb	1224	104	93	206	.26	19	trace
Cu	0	110	129	239	.62	13	
Cu	81	70	58	274	.28	28	
Cu	162	87	60	389	.48	76	
Cu	243	108	88	520	.72	122	
Cu	324	132	81	600	.69	260	
							Cr
Cr	0	110	129	239	.62	13	4
Cr	178	93	110	436	.35	12	26
Cr	356	77	87	377	.27	13	24
Cr	534	66	73	394	.28	8	40
Cr	712	80	94	538	.31	11	32
Zn	0	110	129	239	.62	13	
Zn	306	4969	94	317	.50	12	
Zn	612	7535	95	208	.76	13	
Zn	918	9152	140	254	1.23	12	
Zn	1224	11776	96	302	1.25	12	
							Ni
Ni	0	110	129	239	.62	13	trace
Ni	6	147	142	269	.77	16	9
Ni	12	174	189	267	1.02	19	23
Ni	18	219	148	279	1.02	21	48
Ni	24	199	153	293	1.02	20	64
				Elliott silt loam			
							Pb
Pb	0	59	86	79	.30	6	trace
Pb	306	59	83	74	.27	7	trace
Pb	1224	57	84	59	.18	9	trace

Table 58 (cont). Heavy metal and P content of corn stover as influenced by Pb, Cu, Cr, Zn, and Ni additions to soils.

Treatments		Concentrations in corn stover					
Element	ppm	Zn ppm	Fe ppm	Mn ppm	P %	Cu ppm	Element varied ppm
		Elliott silt loam (cont)					
Cu	0	59	86	79	.30	6	
Cu	81	66	77	66	.21	10	
Cu	324	84	67	77	.16	18	
							Cr
Cr	0	59	86	79	.30	6	2
Cr	534	46	72	39	.17	8	14
Cr	712	59	62	50	.38	9	14
Zn	0	59	86	79	.30	6	
Zn	306	1000	76	64	.23	5	
Zn	1224	5170	81	72	.20	8	

Germination was reduced by high Pb addition to the Plainfield but not the Elliott soil (Table 59).

Yield of elements was generally reduced as rate of Pb increased (Table 60). Some of these reductions were usually due to both reduced yield and concentration levels in the corn. Yield of Cu was affected relatively less than the other elements. Yield of P was reduced relatively more than the other elements.

Rates of Cu corresponding to 15- and 20-year additions of sludge (243 and 324 ppm Cu) were extremely detrimental to germination of corn in the sandy soil (Table 59). Germination was not affected by Cu added to the silt loam soil.

Copper was very detrimental to growth of corn at the rates used in this experiment. On the Plainfield sand, both linear and quadratic regression terms for growth were highly significant (Figure 72). Much of the growth depression occurred with Cu rates no higher than 162 ppm. On the Elliott silt loam, there was a slight increase in corn yield at 81 ppm Cu. This was not a significant increase, however. The overall effect of Cu was to significantly decrease growth of corn on the silt loam.

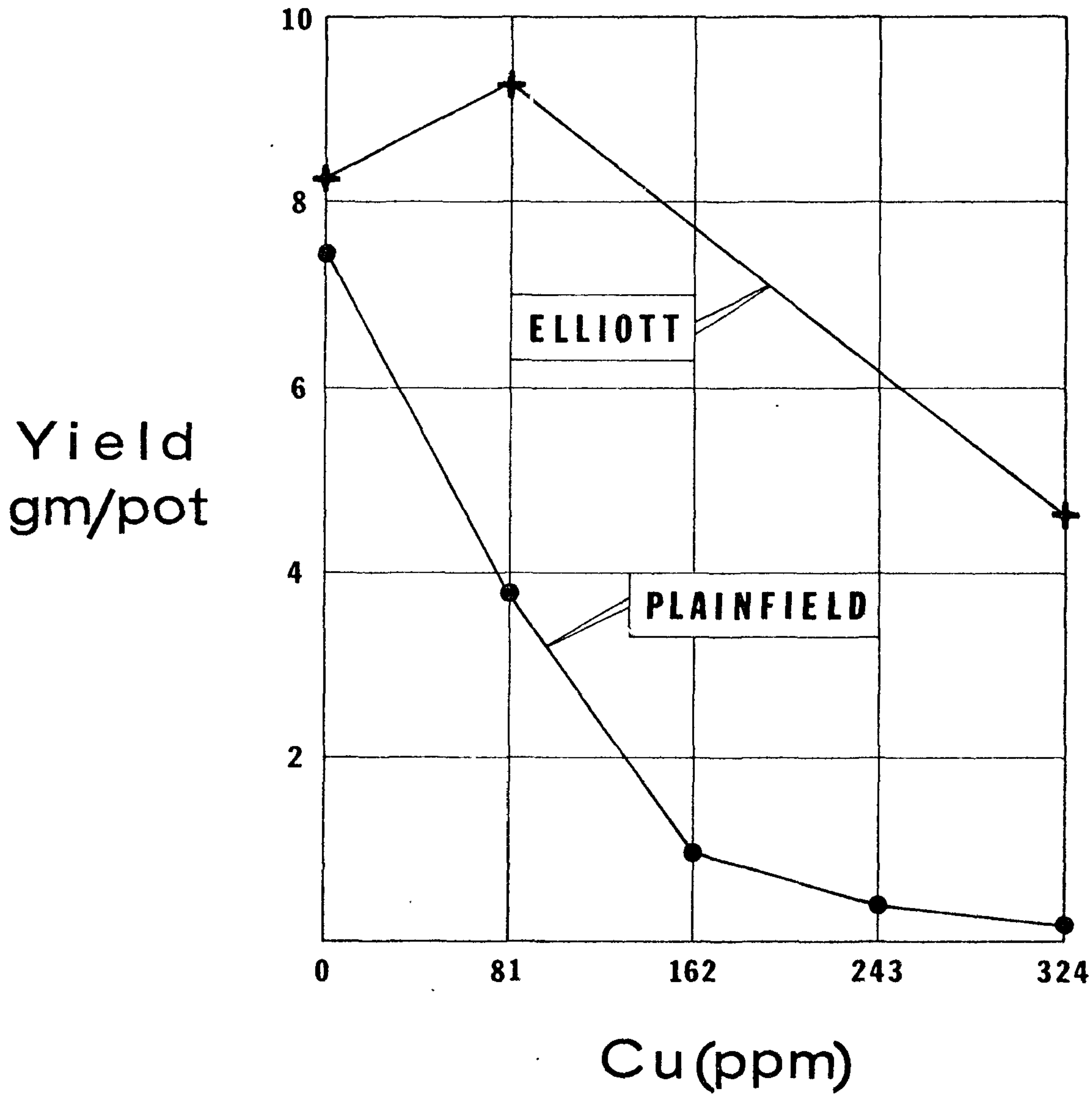


Figure 72. Yield (oven-dry weight) of corn at four weeks in the presence of Cu admixed with Elliott silt loam and Plainfield sand.

Table 59. Germination of corn 10 days after planting as influenced by Pb, Cu, Cr, Zn, and Ni additions to soils.

Treatment		Germination, %	
Element	ppm	Plainfield sand	Elliott silt loam
Pb	0	93	93
Pb	306	93	97
Pb	612	83	--
Pb	918	90	--
Pb	1224	73	97
Cu	0	93	93
Cu	81	100	93
Cu	162	77	--
Cu	243	47	--
Cu	324	30	100
Cr	0	93	93
Cr	178	80	--
Cr	356	60	--
Cr	534	97	100
Cr	712	53	93
Zn	0	93	93
Zn	306	100	90
Zn	612	93	--
Zn	918	90	--
Zn	1224	87	90
Ni	0	93	--
Ni	6	87	--
Ni	12	93	--
Ni	18	93	--
Ni	24	97	--

Table 60. Yield of Zn, Fe, Mn, P, and Cu as influenced by Pb, Cu, Cr, Zn, and Ni.

Treatment		Yield, mg				
Element	ppm	Zn	Fe	Mn	P	Cu
			Plainfield sand			
Pb	0	.822	.964	1.78	46.3	.0972
Pb	306	1.090	1.150	1.59	37.2	.1160
Pb	612	.916	.811	1.23	25.7	.0923
Pb	918	.703	.641	1.11	20.1	.0850
Pb	1224	.646	.598	1.28	16.1	.1180
Cu	0	.822	.964	1.78	46.3	.0972
Cu	81	.262	.217	1.03	10.5	.1050
Cu	162	.083	.057	0.37	4.58	.0726
Cu	243	.040	.033	0.19	2.68	.0454
Cu	324	.024	.015	0.11	1.27	.0480
Cr	0	.822	.964	1.78	46.3	.0972
Cr	178	.652	.771	3.06	24.5	.0841
Cr	356	.418	.473	2.05	14.7	.0706
Cr	534	.437	.483	2.61	18.5	.0529
Cr	712	.371	.435	2.49	14.4	.0510
Zn	0	.822	.964	1.78	46.3	.0972
Zn	306	17.000	.322	1.09	17.1	.0411
Zn	612	3.630	.046	0.10	3.67	.0063
Zn	918	2.150	.033	0.06	2.89	.0028
Zn	1224	2.660	.027	0.07	2.82	.0027
Ni	0	.822	.964	1.78	46.3	.0972
Ni	6	.970	.937	1.77	50.8	.1050
Ni	12	.896	.973	1.38	52.5	.0979
Ni	18	.884	.597	1.13	41.1	.0847
Ni	24	.837	.644	1.23	42.9	.0842
			Elliott silt loam			
Pb	0	.488	.711	.653	24.8	.0496
Pb	306	.459	.645	.575	21.0	.0544
Pb	1224	.385	.567	.398	12.2	.0608

Table 60 (cont). Yield of Zn, Fe, Mn, P, and Cu as influenced by Pb, Cu, Cr, Zn, and Ni.

Treatment		Yield, mg				
Element	ppm	Zn	Fe	Mn	P	Cu
		Elliott silt loam (cont)				
Cu	0	.488	.711	.653	24.8	.0496
Cu	81	.612	.715	.612	19.5	.0928
Cu	324	.389	.310	.356	7.41	.0833
Cr	0	.488	.711	.653	24.8	.0496
Cr	534	.111	.173	.094	4.08	.0192
Cr	712	.058	.060	.049	3.70	.0088
Zn	0	.488	.711	.653	24.8	.0496
Zn	306	6.590	.501	.422	15.2	.3330
Zn	1224	22.200	.348	.309	8.6	.0344

Concentration levels of Cu in the aerial portions of the corn were not very high considering the concentrations added to the Elliott silt loam (Table 58). This is in agreement with results reported by Reuther (127). Copper concentration levels in corn grown on the Plainfield sand increased significantly as the rate of added Cu was increased.

At the 5-year rate of Cu (81 ppm), corn grown on the sand had a severe interveinal chlorosis symptomatic of an iron deficiency. Iron content of plants was significantly reduced (Table 58). On the silt loam soil this chlorosis occurred at the 20-year rate of Cu although it took longer to develop and was not as severe as the chlorosis which developed at the 81-ppm treatment on the sandy soil. Iron content, however, was not significantly affected by Cu treatment at the 95 percent confidence level. Manganese content of corn on the sandy soil was increased as Cu additions were increased. This may have been due to pH changes in the soils since the soils treated with Cu sulfate were more acid (0.5 - 0.7 pH units less) than the control. No similar increase in Mn content occurred for corn grown on the silt loam. Zinc content of corn grown on the sandy soil was depressed to 70 ppm at the 81-ppm Cu treatment and then increased steadily thereafter as plants were more adversely affected by a Cu toxicity.

Copper additions reduced corn growth so much that yield of all elements was greatly reduced as Cu rates were increased in the sandy soil (Table 60). Yield of elements was reduced on the silt loam soil, except for Cr, as Cu was added. The reduction in yield of P was much greater than the reduction in stover yield on the silt loam soil.

Chromium was more toxic to corn grown on the silt loam soil than when grown on the sandy soil (Figure 73). This was unexpected since the silt loam soil is more highly buffered than the sandy soil. Growth reduction, although less than on the silt loam soil occurred on the sandy soil. The linear regression term was significant for the sand even though the effect due to treatment was somewhat erratic. On the silt loam soil a highly significant decrease in corn growth occurred as Cr additions were increased. Severe stunting and purpling of the leaves occurred at the highest rate of Cr.

Germination of corn in the sandy soil was affected by Cr, but this, too, did not seem to follow rates of application very closely (Table 59).

Chromium significantly affected Fe content. On both soils, Cr significantly decreased the Fe content of corn (Table 58). At 714 ppm of Cr, P concentration of corn on the sandy soil was reduced to half that of the control, and this reduction was highly significant. Total P uptake by the corn was significantly reduced on both soils (Table 60). Yield of Mn on sandy soil was the only element whose yield was not reduced by increasing Cr application rates.

Precipitation of phosphates by Cr in the soil is not likely since Cr phosphate salts are soluble in dilute acid solutions and water. Therefore, it would seem most likely that this reduction in P content in the plant due to high Cr additions to the soil is a physiological phenomenon. Zinc content was also significantly reduced in corn grown on the sandy soil (Table 58). This kind of reduction did not occur on the silt loam soil.

Manganese content of corn and yield of Mn was significantly increased as amounts of Cr applied to the sandy soil were increased (Tables 58 and 60). This would seem to be directly related to Cr additions to the soil since pH of the soil did not change with treatment as it did with the Cu treatments. On the silt loam soil, however, the reverse occurred. Manganese content in the corn stover decreased significantly as the rate of Cr applied to the silt loam increased.

Concentrations of Cr in the corn grown on the two soils leads to another paradox. Content of the above-ground portion of corn was less on the silt loam soil where more damage to growth occurred, than on the sandy soil (Table 58).

It would seem that perhaps a change in the valence of chromium occurred in the silt loam soil which did not occur in the sandy soil. Perhaps this change in valence could have occurred through a biological oxidation of the chromic ion to the chromate or dichromate ion. Microbial activity has been known to oxidize the manganous ion, so the possibility of chromic ion oxidation may be quite good. A purely chemical oxidation in the soil is remote since the chromic ion is only oxidized by

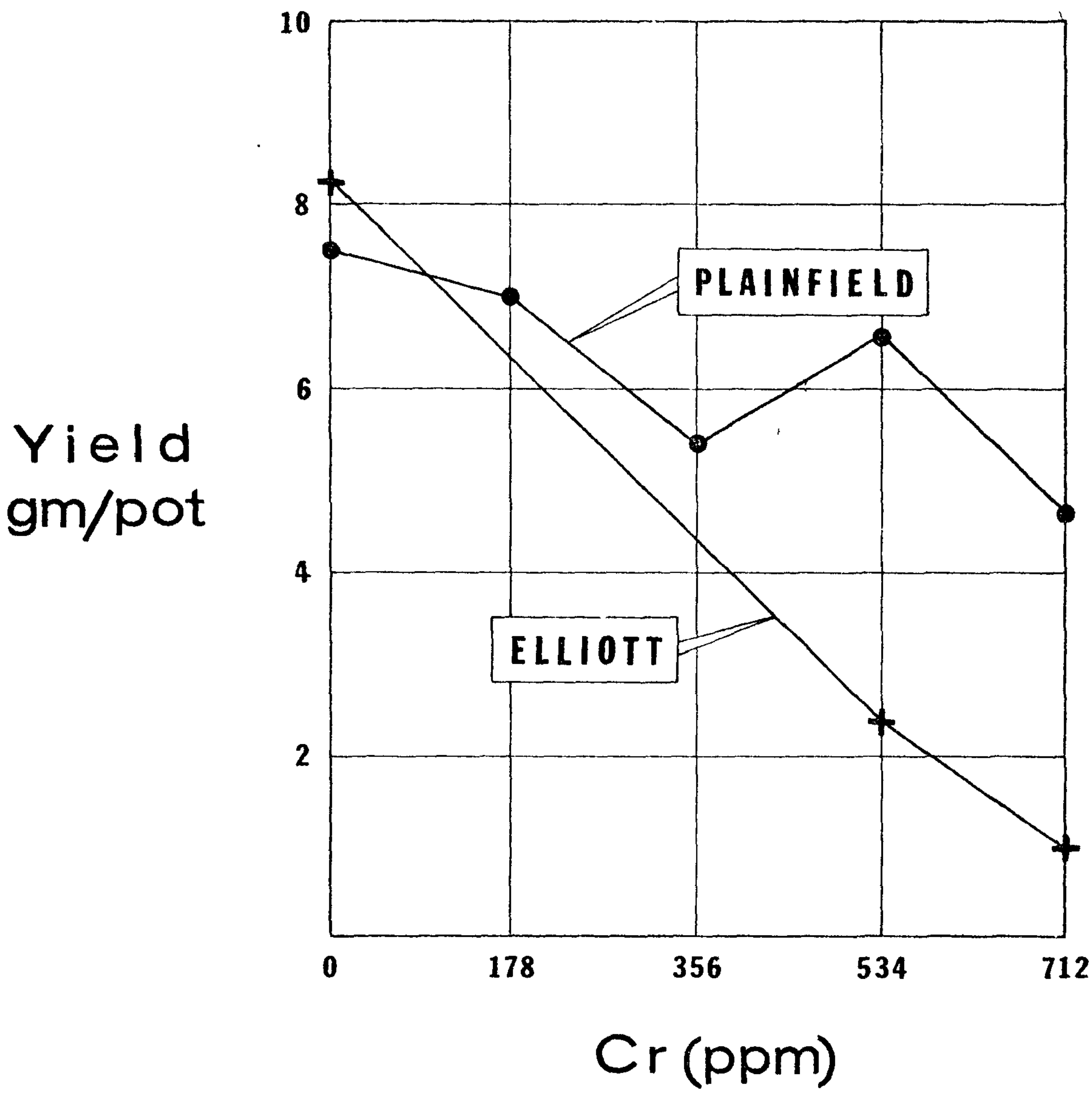


Figure 73. Yield (oven-dry weight) of corn at four weeks in the presence of Cr admixed with Elliott silt loam and Plainfield sand.

rather strong oxidizers. Since the silt loam had a good supply of organic matter and the sandy soil hardly any, this would mean the microbe population should be at a minimum in the sandy soil but high in the silt loam soil. Microbes which could possibly oxidize the chromic ion are facultative autotrophs. Thus, the silt loam soil being more abundant in organic matter content than the sandy soil can harbor a larger population of these microbes.

Soane and Saunder (144) suggested that the degree of Cr toxicity could be influenced by Ca and P levels in soil. Since our two soils differed considerably in their Ca and P content, this, too, may explain the differences in Cr toxicity between the two soils.

Oxidation state of Cr is very important because its toxicity is greatly influenced by the valence. Hewitt (66) reported that chromate was more toxic than chromic, although it is doubtful that in slightly acid soils chromate will exist for long periods of time before it is converted to dichromate. This conversion, however, does not lead to a further change in valence. Soane and Saunder (144) used dichromate in their sand-culture study. They found 50 ppm of Cr as dichromate caused the same severe symptoms that the highest rate of Cr caused in this study on the silt loam soil. Comparing the effect of chromic ion concentrations on growth of corn on the sandy soil in this experiment with the effect of dichromate ion concentrations on growth of corn on the sand-culture experiment of Soane and Sanuders would seem to substantiate Hewitt's finding.

Zinc was very toxic on the sandy soil even at the 5-year rate (306 ppm) (Figure 74). The plants were stunted and the lower leaves were bright red. The two highest rates of Zn caused growth to terminate shortly after germination. All the plants turned to a brilliant red. On the silt loam soil where only the 5-year and 20-year rates of Zn were applied, there was a slight growth depression at the first Zn rate and a yield of only one-half that of the control at the highest rate.

Zinc increased to very high concentration levels in the corn for both soils treated with Zn (Table 58). Where 306 ppm of Zn were added, concentration levels in corn grown on the sandy and silt loam soils were 4969 and 1000 ppm of Zn, respectively. Phosphorus deficiency does not seem to be responsible for the red color which developed in the corn on the Zn-treated sandy soil. Phosphorus content of the plants increased as the rates of Zn increased on the sandy soil (Table 58). The high concentration level of P was undoubtedly due to the lack of plant growth that prevented a dilution effect. Phosphorus concentration levels in the corn stover were significantly reduced as Zn application rates were increased on the silt loam soil.

Germination was not appreciably affected by the Zn treatments (Table 59).

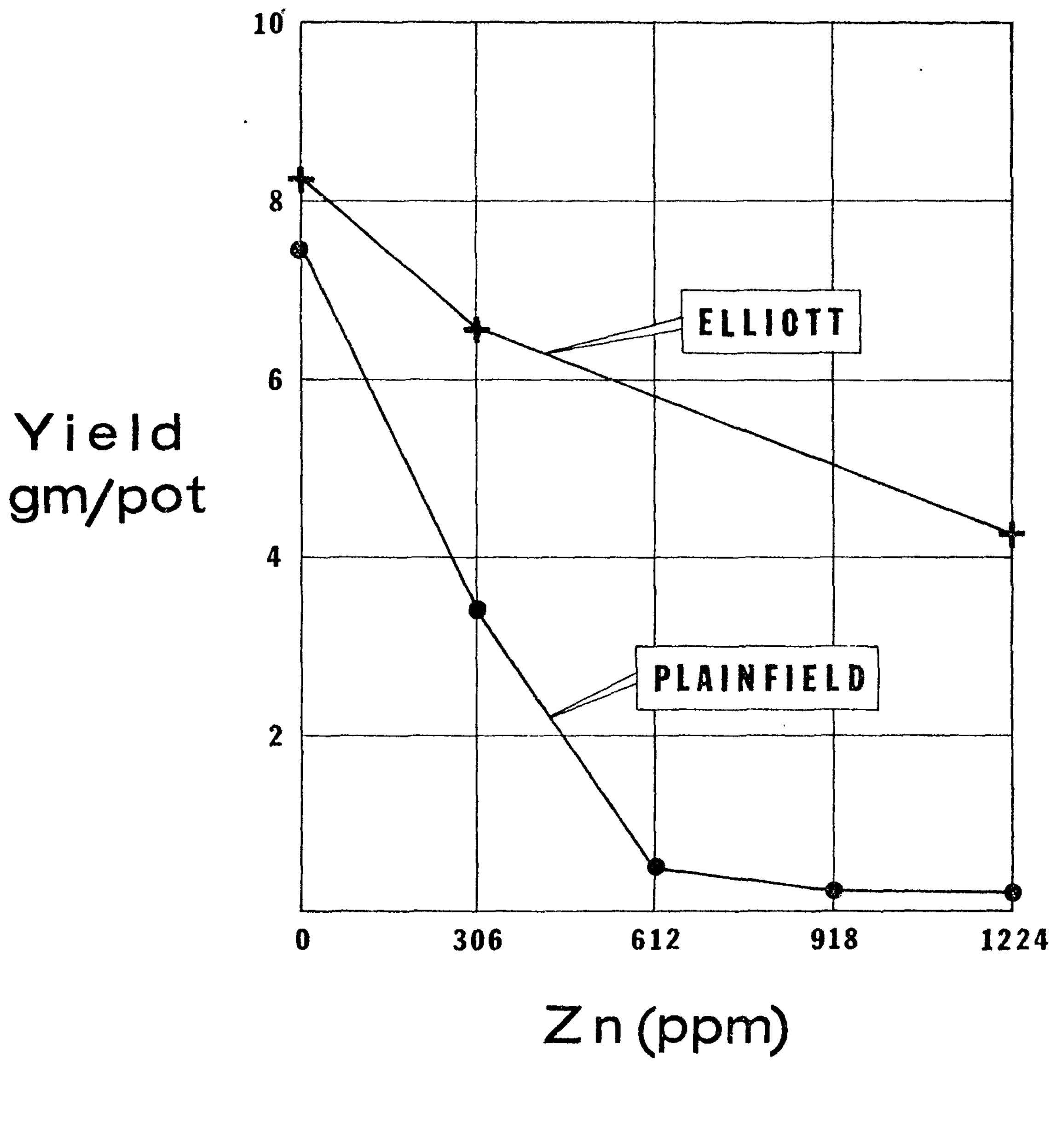


Figure 74. Yield (oven-dry weight) fo corn at four weeks in the presence of Zn admixed with Elliott silt loam and Plainfield sand.

Nickel was applied only to the Plainfield sand. Germination was not decreased by the Ni treatments (Table 59). Corn stover yield was significantly decreased as the application rate of Ni was increased (Figure 75). Added Ni caused a highly significant increase in Ni concentration levels in plants (Table 58). All chemical elements considered tended to increase in plant tissue as Ni application rates were increased. Yield of all elements, except Zn, tended to decrease as application rates of Ni were increased (Table 60).

Nickel induced symptoms were very similar to those symptoms associated with a Ca deficiency. The corn growing on the Ni-treated soils started to have very gummy, whip-like terminal leaves after two and one-half weeks of growth. These leaves failed to part completely; the tips were glued together. Leaf tips which appeared to be glued together died after three or four days. Tips of older separated leaves were necrotic and eventually died. Hewitt (66) reported similar symptoms in potatoes grown in sand culture studies with various concentration levels of added Ni. The older leaves of potatoes withered and the growing tips died in the latter stages of toxicity, but this was preceded by a chlorosis which Hewitt considered similar to Mn deficiency symptoms. Soane and Saunder (144) also noted that toxic levels of Ni caused an interveinal chlorosis. Chlorosis occurred only three days after emergence when corn seedlings growing in a sand culture where 30 ppm Ni were added. Yet the Ca content of the corn increased as the application rate of Ni was increased. Crooke (38) observed an increase in Ca content when 2.5 ppm Ni was present in a sand culture. Perhaps Ni can create a greater need for Ca in the plant. This would explain why the apparent Ca deficiency symptoms became worse despite increased Ca content in the corn stover as application rates of Ni were increased.

Effects of Digested Sewage Sludge Added to Soil on Growth and Composition of Soybean: Part I

Introduction -- The experiment described here was carried out to determine the effects of heated anaerobically digested sludge mixed in large quantities with soil which was subsequently planted to soybeans. In addition, an attempt was made to determine how different levels of elements occurring in sludge, added as salts to simulate a readily available form of the element, might interact with freshly-applied sludge to affect plant growth. The experiment, carried out under greenhouse conditions, was planned in a factorial manner with three levels of salt-simulated sludge and of digested sludge. The increments between treatments increased by a factor of two, giving the highest combined treatment a weight equivalent of 144 t/ha of sludge solids. The design allosw comparison of the availability of the elements in sludge to be compared with those added in salt form.

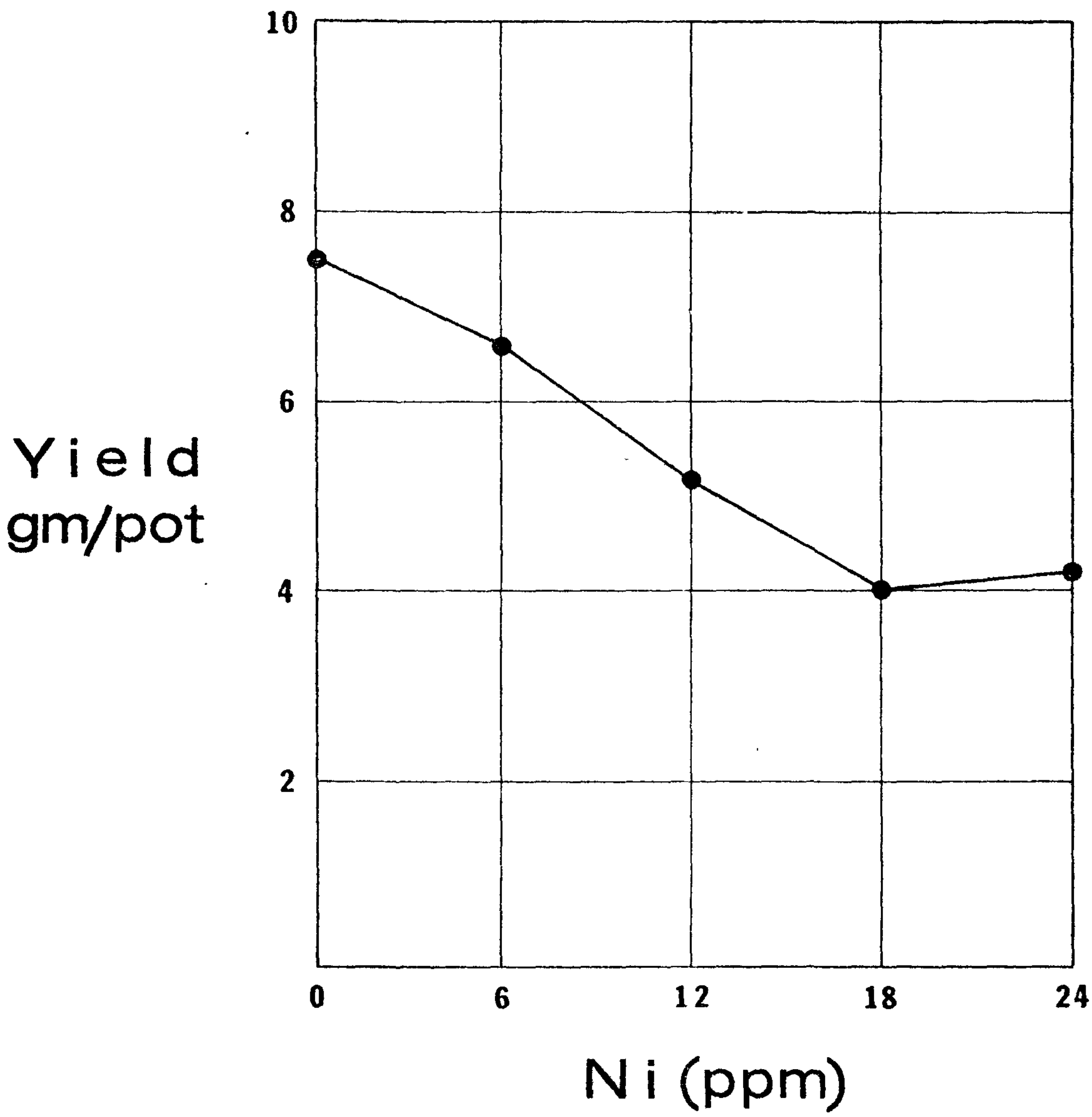


Figure 75. Yield (oven-dry weight) of corn at four weeks in the presence of Ni admixed with Plainfield sand.

Methods - Anaerobically digested sewage sludge was obtained from the Calumet Waste Treatment Plant of the Metropolitan Sanitary District of Greater Chicago. The sludge represents sewerage from both industrial and domestic sources. This sludge was analyzed (Table 61) for the elements subsequently used to prepare the salt-simulated sludge. The salts were chosen to combine as many of the elements involved in the simulated sludge and still maintain a highly soluble form. For several of the metals it was necessary to add acetate salts. It was felt that the acetate would be rapidly utilized by the soil bacteria and not affect plants subsequently grown in the soil. Sludge and simulated sludge were added incrementally to four kilograms of Elliott silt loam soils until the total amount for each treatment had been added. The treatments amounted to 36, 72 and 144 t/ha of solids or equivalent in the case of simulated sludge. The dry soil was crushed and four kilograms placed in a 20-cm diameter by 20-cm high plastic pot and distilled water was added from above and below to bring moisture content to 20 percent by weight. Soybean seeds [Glycine max (L.) Merr., var Corsoy] were germinated in sand and when 2-to 3-cm high were transplanted to the pots.

Moisture content of the soil was maintained by daily watering, taking care to moisten the soil throughout its depth, and the amount of water added was recorded. After 27 days, moisture content of soil was increased to 37.5 percent. Surface area of the fourth leaf from the top on all plants was determined on the 35th day and leaf surface area of all leaves on the largest plant in each pot was determined on the 45th day. Height of each plant was measured on day 12 and each week thereafter. Six plants from each pot were harvested to 2.5 cm above the soil surface after 22 days and during the rapid growth phase and three plants were harvested after 37 days, the date of initiation of blooming. The plants were washed with distilled water, dried at 60°C and ground in a Wiley Mill to pass 40 mesh.

Tissue was analyzed for Ca, Mg, Fe, Mn, Zn, Cu, Cd, and Ni by atomic absorption analysis after ashing at 500°C with care being taken to raise the temperature gradually and dissolving the ash in hydrochloric acid. Phosphorus was determined by the vanadomolybdate yellow method and N by the Kjeldahl method. Sodium and K were analyzed by flame emission spectroscopy. Also, the above elements, with the exception of N, were determined in a 0.1 N HCl extract of an alloquot of the soil taken before planting. A ratio of 0.5 gm soil to 10 ml of acid was used.

Incidence of weeds was determined by counting the number of plants emerging in the pots after 13 days. Germination of soybeans in the soil of each treatment was assessed by placing 100 seeds in Petri dishes containing the particular treatment and moistening the soil. The number of seeds germinating was counted from day 2 through 13. This germination experiment was not replicated.

Table 61. Elemental composition of salt-simulated sludge and salts used in its formulation and composition of sludge from Calumet Sewage Treatment Works. The Calumet sludge contained 3.13 percent solids of which 43.7 percent were volatilized by heating to 500°C.

Simulated Sludge				Calumet Sludge
Element	Salt Used	g salt/l	mg element/l	mg/l
N	$(NH_4)_2SO_4$	.309	800	900
	NH_4Cl	.634		
	$(NH_4)H_2PO_4$	3.0006		
	$NH_4Mo_7O_{24}$	.0006		
	NH_4OH	.244		
P	KH_2PO_4	.574	850	626
	$NH_4H_2PO_4$	3.066		
K	KH_2PO_4	.574	165	205
Ca	$Ca(C_2H_3O_2)_2$	5.275	1200	1243
Mg	$Mg(C_2H_3O_2)_2$	1.610	275	366
Na				126
Fe	$FeC_6H_5O_7 \cdot 3H_2O$	5.354	1000	1230
Mn	$Mn(C_2H_3O_2)_2$	.045	10	16
Zn	$Zn(C_2H_3O_2)_2$	.822	170	148
Cu	$Cu(C_2H_3O_2)_2$	.132	42	33
S	$(NH_4)_2SO_4$	.309	75	n.d.
B	H_3BO_3	.009	1.5	n.d.
Mo	$NH_4Mo_7O_{24}$	.0006	.05	n.d.
Cr	$Cr(C_2H_3O_2)_3$	.171	36	23
Pb	$Pb(C_2H_3O_2)_2$	.070	38	56
Cd	$Cd(C_2H_3O_2)_2$	.033	14	4
Ni	$Ni(C_2H_3O_2)_2$	.017	4	3

Results and discussion, general statement - All except the highest combined treatment - equal to 288 t of solids/ha - supported plant growth to the first harvest at 22 days. However, plants died in a number of treatments during the period between this harvest and the second harvest at 37 days. Plants affected were in the highest treatment of salt-simulated sludge and combinations of the second and highest levels of simulated sludge and sludge. Reference to the section of Table 65 dealing with the second cutting indicates those treatments which would not support growth. Death of plants was preceded by a series of early symptoms progressing from interveinal chlorosis, drying and rolling of the primary leaves, through interveinal chlorosis and wrinkling of the second through fourth trifoliate leaves. Higher leaves appeared normal, especially in the lower treatments.

The experiment was abandoned after 58 days because of the loss of treatments and poor vigor in some of the surviving lower treatments. Although toxicity due to the high levels of metals was an obvious and appealing answer to the cause of loss of plants, we suspected salt effects. Saturation extracts of the soils were prepared and conductivities were determined. The conductivities (Table 62) indicate that salt accumulation would be a considerable problem for growth of soybeans in such a sludge-amended medium where percolating water would not remove salts. The salt-simulated sludge also created intolerable conditions for soybean growth. Sodium content of the paste extract (Table 63) increases through each kind of treatment probably because the Na ion is easily displaced from exchange positions on soil colloids by the more abundant and strongly adsorbed two and higher valent ions in the sludges. The nature of soybean growth after the soil in each treatment had been leached to a conductivity of less than 1.0 mmho/cm or slightly more than the 0.47 mmho/cm of the control soil is the subject of part two of this report. Data for pH of the soils are gathered into Table 64. Addition of salts to the soil depressed pH by about one unit and analysis of the data indicates main effects for both sludge and salt-simulated sludge are present. The range in pH involved in these treatments should not markedly influence nutrient uptake or be physiologically detrimental, in fact, the absorbtion of metals should be reduced substantially by the slightly alkaline conditions.

Means of elemental contents of tissue for the first and second harvests are given in Tables 65 through 76 for N, P, K, Ca, Mg, Fe, Mn, Zn, Cu, Na, Ni, and Cd, respectively. The data were treated by analysis of variance to determine the presence of treatment effects. To determine mutual relationships among concentrations of elements, the data were analyzed by correlation analysis.

Table 62. Effect of digested sludge and salt-simulated sludge on conductivity of saturation paste extract of soil. Data in mmhos/cm.

DIGESTED SLUDGE	SALT-SIMULATED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	0.47	6.70	13.30	14.37	8.71
36	1.58	11.47	13.03	13.83	9.98
72	8.67	12.63	11.47	11.13	10.98
144	11.57	11.20	11.87	25.10	14.93
Average Effect	5.57	10.50	12.42	16.11	

Table 63. Effect of digested sludge and salt-simulated sludge on Na content of saturation paste extract of soil. Data in parts per million of extract.

DIGESTED SLUDGE	SALT-SIMULATED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	4	34	69	128	59
36	9	47	67	118	60
72	21	51	77	135	71
144	45	75	89	167	94
Average Effect	20	52	75	137	

Table 64. Effect of digested sludge and salt-simulated sludge on pH of soil in which soybeans were grown.

DIGESTED SLUDGE	SALT-SIMULATED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	6.6	6.6	6.4	5.7	6.3
36	7.0	7.0	6.8	5.8	6.7
72	7.8	7.5	7.1	6.7	7.3
144	7.9	7.5	7.2	6.4	7.2
Average Effect	7.3	7.2	6.9	6.2	

Least significant differences

Interaction 0.6 (19:1)
Average effect
Salt-simulated sludge 0.4 (99:1)
Digested sludge 0.3 (99:1)

Table 65. Effect of digested sludge and salt-simulated sludge on N content in soybean. Data are in percent of oven-dry (60°C) tissue. Analysis of variance performed on 4 by 3 treatments for 22-day cutting and 4 by 2 treatments for 37-day cutting. Average effects for these treatments are in parentheses.

1st Cutting, 22 days

DIGESTED SLUDGE	SALT-SIMULATED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	5.74	6.67	7.19	7.59	5.76(6.48)
36	4.64	7.75	8.26	8.34	7.25(6.88)
72	8.27	8.16	8.51	8.54	8.37(8.31)
144	8.94	8.00	8.49	--	(8.47)
Average Effect	(6.86)	(7.64)	(8.11)	8.16	

Least significant difference (P)

Interaction 0.74 (99:1)
Average effect
Salt-simulated sludge 0.37 (99:1)
Digested sludge 0.28 (99:1)

2nd Cutting, 37 days

DIGESTED SLUDGE	SALT-SIMULATED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	3.68	3.76	4.22	--	3.89(3.72)
36	4.39	5.37	6.09	--	5.28(4.88)
72	5.51	5.34	--	--	(5.42)
144	6.40	7.08	--	--	(6.74)
Average Effect	(5.00)	(5.39)	5.16	--	

Least significant difference (P)

Interaction 0.37 (99:1)
Average effect
Salt-simulated sludge 0.25(99:1)
Digested sludge 0.25 (99:1)

Table 66. Effect of digested sludge and salt-simulated sludge on P content in soybean. Data are in percent of oven-dry (60°C) tissue. Analysis of variance performed on 4 by 3 treatments for 22-day cutting and 4 by 2 treatments for 37-day cutting. Average effects for these treatments are in parentheses.

1st Cutting, 22 days

DIGESTED SLUDGE	SALT-SIMULATED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	.42	.62	.70	.50	.56(.58)
36	.50	.52	.48	.44	.48(.50)
72	.48	.48	.37	.59	.48(.44)
144	.44	.48	.44	--	(.45)
Average Effect	(.46)	(.52)	(.50)	.51	

Least significant difference (P)

Interaction .13 (19:1)
Average effect
Salt-simulated sludge none
Digested sludge .05 (19:1)

2nd Cutting, 37 days

DIGESTED SLUDGE	SALT-SIMULATED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	.43	.62	.56	--	.54(.52)
36	.51	.49	.47	--	.49(.50)
72	.58	.35	--	--	(.46)
144	.52	.34	--	--	(.43)
Average Effect	(.51)	(.45)	.52	--	

Least significant difference (P)

Interaction .08 (99:1)
Average effect
Salt-simulated sludge .03 (19:1)
Digested sludge .04 (99:1)

Table 67. Effect of digested sludge and salt-simulated sludge on K content in soybean. Data are in percent of oven-dry (60°C) tissue. Analysis of variance performed on 4 by 3 treatments for 22-day cutting and 4 by 2 treatments for 37-day cutting. Average effects for these treatments are in parentheses.

1st Cutting, 22 days

DIGESTED SLUDGE	SALT-SIMULATED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	1.90	2.45	2.50	2.42	2.32(2.28)
36	2.26	2.11	2.08	2.34	2.20(2.15)
72	1.90	1.92	1.95	2.51	2.07(1.92)
144	1.85	2.07	2.26	--	(2.06)
Average Effect	(1.98)	(2.14)	(2.20)	2.39	

Least significant difference

Interaction
Average effect
Salt-simulated sludge
Digested sludge

2nd Cutting, 37 days

DIGESTED SLUDGE	SALT-SIMULATED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	.98	.89	.80	--	.89(.93)
36	.84	.72	--	--	(.78)
72	.71	.74	--	--	(.72)
144	.80	.96	--	--	(.88)
Average Effect	(.83)	(.83)	.80		

Least significant difference

Interaction
Average effect
Salt-simulated sludge
Digested sludge

Table 68. Effect of digested sludge and salt-simulated sludge on Ca content in soybean. Data are in percent of oven-dry (60°C) tissue. Analysis of variance performed on 4 by 3 treatments for 22-day cutting and 4 by 2 treatments for 37-day cutting. Average effects for these treatments are in parentheses.

1st Cutting, 22 days

DIGESTED SLUDGE	SALT-SIMULATED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	1.85	2.36	1.91	1.61	1.93(2.04)
36	2.37	2.02	1.24	1.26	1.73(1.88)
72	1.96	1.38	1.03	.59	1.24(1.45)
144	1.26	1.00	1.00	--	(1.09)
Average Effect	(1.86)	(1.69)	(1.30)	1.15	

Least significant difference (P)

Interaction 0.28 (99:1)
Average effect
Salt-simulated sludge 0.14(99:1)
Digested sludge 0.11 (99:1)

2nd Cutting, 37 days

DIGESTED SLUDGE	SALT-SIMULATED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	1.51	2.49	4.08	--	2.69(2.00)
36	3.52	4.63	--	--	(4.08)
72	3.63	3.51	--	--	(3.57)
144	3.24	3.07	--	--	(3.16)
Average Effect	(2.98)	(3.42)	4.08	--	

Least significant difference (P)

Interaction 0.46 (99:1)
Average effect
Salt-simulated sludge 0.23 (99:1)
Digested sludge 0.23 (99:1)

Table 69. Effect of digested sludge and salt-simulated sludge on Mg content in soybean. Data are in percent oven-dry (60°C) tissue. Analyses of variance performed on 4 by 3 treatments for 22-day cutting and 4 by 2 treatments for 37-day cutting. Average effects for these treatments are in parentheses.

1st Cutting, 22 days

DIGESTED SLUDGE	SALT-SIMULATED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	.89	.73	.64	.60	.71(.75)
36	.78	.71	.52	.50	.63(.67)
72	.76	.63	.48	.36	.56(.62)
144	.69	.60	.50	--	(.60)
Average Effect	(.78)	(.67)	(.53)	.49	

Least significant difference (P)

Interaction 0.05 (19:1)
Average effect
Salt-simulated sludge 0.03 (99:1)
Digested sludge 0.02 (99:1)

2nd Cutting, 37 days

DIGESTED SLUDGE	SALT-SIMULATED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	.62	.90	.98	--	.83(.76)
36	1.01	1.17	--	--	(1.09)
72	1.04	.99	--	--	(1.02)
144	1.00	.95	--	--	(.98)
Average Effect	(.92)	(1.00)	.98		

Least significant difference (P)

Interaction 0.15 (99:1)
Average effect
Salt-simulated sludge 0.06 (19:1)
Digested sludge 0.08 (99:1)

Table 70. Effect of digested sludge and salt-simulated sludge on Fe content in soybean. Data are in parts per million of oven-dry (60°C) tissue. Analysis of variance performed on 4 by 3 treatments for 22-day cutting and 4 by 2 for treatments for 37-day cutting. Average effects for these treatments are in parentheses.

1st Cutting, 22 day

DIGESTED SLUDGE	SALT-SIMULATED SLUDGE				
t/ha	0	36	72	144	Average Effects
0	75	72	77	64	72(74)
36	85	86	75	61	77(82)
72	90	68	51	59	67(70)
144	79	85	58	--	(74)
Average Effects	(82)	(78)	(65)	61	

Least significant difference (P)

Interaction 16 (99:1)
Average effect
Salt-simulated sludge 11 (99:1)
Digested sludge none

2nd Cutting, 37 day

DIGESTED SLUDGE	SALT-SIMULATED SLUDGE				
t/ha	0	36	72	144	Average Effects
0	51	63	57	--	57(57)
36	59	29	--	--	(44)
72	40	32	--	--	(36)
144	49	54	--	--	(52)
Average Effect	(50)	(44)	57	--	

Least significant difference
None

Table 71. Effect of digested sludge and salt-simulated sludge on Mn content in soybean. Data are in parts per million of oven-dry (60°C) tissue. Analysis of variance performed on 4 by 3 treatments for 22-day cutting and 4 by 2 treatments for 37-day cutting. Average effects for these treatments are in parentheses.

1st Cutting, 22 days

DIGESTED SLUDGE	SALT-SIMULATED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	40	64	63	81	62(55)
36	44	74	59	47	56(59)
72	80	78	52	27	59(70)
144	100	82	51	---	(78)
Average Effect	(66)	(74)	(56)	52	

Least significant difference (P)

Interaction 18 (99:1)
Average effect
Salt-simulated sludge 9 (99:1)
Digested sludge 7 (99:1)

2nd Cutting, 37 days

DIGESTED SLUDGE	SALT-SIMULATED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	28	33	163	---	75(31)
36	89	241	---	---	(165)
72	212	158	---	---	(186)
144	364	208	---	---	(286)
Average Effect	(173)	(160)	163		

Least significant difference (P)

Interaction 54 (99:1)
Average effect
Salt-simulated sludge none
Digested sludge 27 (99:1)

Table 72. Effect of digested sludge and salt-simulated sludge on Zn content in soybean. Data are in parts per million of oven-dry (60°C) tissue. Analysis of variance performed on 4 by 3 treatments for 22-day cutting and 4 by 2 treatments for 37-day cutting. Average effects for these treatments are in parentheses.

1st Cutting, 22 days

DIGESTED SLUDGE	SALT-SIMULATED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	351	919	1079	832	795(783)
36	966	892	501	465	706(783)
72	771	633	403	353	540(602)
144	761	637	478	--	(625)
Average Effect	(712)	(770)	(615)	550	

Least significant difference (P)

Interaction 185 (99:1)
Average effect
Salt-simulated sludge 93 (99:1)
Digested sludge 70 (99:1)

2nd Cutting, 37 days

DIGESTED SLUDGE	SALT-SIMULATED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	172	228	420	--	273(200)
36	291	352	--	--	(322)
72	367	188	--	--	(278)
144	353	216	--	--	(284)
Average Effect	(296)	(246)	420		

Least significant difference (P)

Interaction 135 (99:1)
Average effect
Salt-simulated sludge
Digested sludge 68 (99:1)

Table 73. Effect of digested sludge and salt-simulated sludge on Cu content in soybean. Data are in parts per million of oven-dry (60°C) tissue. Analysis of variance performed on 4 by 3 treatments for 22-day cutting and 4 by 2 treatments for 37-day cutting. Average effects for these treatments are in parentheses.

1st Cutting, 22 days

DIGESTED SLUDGE	SALT-SIMULATED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	9.3	15	22	21	17(15)
36	14	20	20	18	18(18)
72	17	17	18	24	19(17)
144	22	19	17	--	(19)
Average Effect	(16)	(18)	(19)	21	

Least significant difference (P)

Interaction 6 (99:1)
Average effect
Salt-simulated sludge 2 (19:1)
Digested sludge none

2nd Cutting, 37 days

DIGESTED SLUDGE	SALT-SIMULATED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	76	27	25	--	43(51)
36	73	20	--	--	(46)
72	21	30	--	--	(26)
144	27	46	--	--	(36)
Average Effect	(49)	(31)			

Least significant difference (P)

Interaction 39 (99:1)
Average effect
Salt-simulated sludge none
Digested sludge 14 (19:1)

Table 74. Effect of digested sludge and salt-simulated sludge on Na content in soybean. Data are in parts per million of oven-dry (60°C) tissue. Analysis of variance performed on 4 by 3 treatments for 22-day cutting and 4 by 2 treatments for 37-day cutting. Average effects for these treatments are in parentheses.

1st Cutting, 22 days

DIGESTED SLUDGE	SALT-SIMULATED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	364	372	452	433	405(396)
36	850	519	577	848	698(649)
72	995	680	802	870	837(826)
144	1192	1001	1209	--	(1134)
Average Effect	(850)	(643)	(760)	717	

Least significant difference (P)

Interaction none
Average effect
Salt-simulated sludge 139 (99:1)
Digested sludge 105 (99:1)

2nd Cutting, 37 days

DIGESTED SLUDGE	SALT-SIMULATED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	134	146	198	--	159(140)
36	208	414	--	--	(311)
72	763	1554	--	--	(1158)
144	1215	2574	--	--	(1894)
Average Effect	(580)	(1172)	198	--	(1894)

Least significant difference (P)

Interaction 529 (99:1)
Average effect
Salt-simulated sludge 264 (99:1)
Digested sludge 264 (99:1)

Table 75. Effect of digested sludge and salt-simulated sludge on Ni content in soybean. Data are in parts per million of oven-dry (60°C) tissue. Analysis of variance performed on 4 by 2 treatments. Average effects for these treatments are in parentheses.

2nd Cutting, 37 days

DIGESTED SLUDGE	SALT-SIMULATED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	5.3	8.0	4.0	--	5.8(6.7)
36	5.3	10.7	--	--	(8.0)
72	4.0	4.0	--	--	(4.0)
144	4.0	4.0	--	--	(4.0)
Average Effect	(4.6)	(6.7)	4.0	--	

Least significant difference

None

Table 76. Effect of digested sludge and salt-simulated sludge on Cd content in soybean. Data are in parts per million of oven-dry (60°C) tissue. Analysis of variance performed on 4 by 3 treatments for 22-day cutting and 4 by 2 treatments for 37-day cutting. Average effects for these treatments are in parentheses.

1st Cutting, 22 days

DIGESTED SLUDGE	SALT-SIMULATED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	1.0	5.3	6.7	8.6	5.4(4.3)
36	3.0	5.1	3.2	3.2	3.6(3.8)
72	3.4	2.8	2.5	1.6	2.6(2.9)
144	2.7	2.9	2.3	--	(2.6)
Average Effect	(2.5)	(4.0)	(3.7)	4.5	

Least significant difference (P)

Interaction 1.1 (99:1)
Average effect
Salt-Simulated sludge 0.6 (99:1)
Digested sludge 0.4 (99:1)

2nd Cutting, 37 days

DIGESTED SLUDGE	SALT-SIMULATED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	1.0	1.9	8.1	--	3.7(2.8)
36	1.1	4.4	--	--	(2.8)
72	2.0	3.2	--	--	(2.6)
144	2.2	4.0	--	--	(3.1)
Average Effect	(1.6)	(3.4)	8.1		

Least significant difference (P)

Interaction 1.1 (99:1)
Average effect
Salt-simulated sludge 0.6 (99:1)
Digested sludge 0.6 (99:1)

Treatment effects for elemental content of the first cutting (22 days) and miscellaneous data - The results of analysis of variance for main effects and interaction for treatments are gathered into Table 84. Inspection of this table indicates that only the main effect for sludge on Fe content and the interaction of the treatments for Na are not significant among the eleven elements analyzed. Reference to Tables 68, 69, 72, and 76 indicates contents of Ca, Mg, Zn, and Cd, especially with digested sludge treatment, decrease in content with increasing application of sludge or simulated sludge. Ash content (Table 77), reflecting these decreases, also undergoes a marked decline. Na increases with digested sludge treatment. Production of dry matter or yield (Table 78) and leaf surface area of all leaves at 35 days and of the fourth top leaf at 45 days (Tables 79 and 80) are significantly related to digested sludge treatment wherein increasing amounts of sludge applied depresses yield.

Growth rate (Table 81) from the 12th through 21st days shows significant negative treatment effects for both sludge and simulated-sludge treatments. Interaction effects also occur. Increasing treatments of each kind depresses the incidence of weeds (Table 82) which were primarily grasses occurring in the soil. Similarly, the test of germination of soybean seeds (Table 83) shows significant negative effects of digested sludge and interaction effects. This interaction suggests that inhibition is more closely associated with some toxicity than with strictly osmotic effects.

Compared with values for soybean tissue reported by Walker (164) for soybeans sampled across Illinois, N, Zn and perhaps K and Na contents are very high. Phosphorus contents are within the upper regions of ranges reported. Iron contents are smaller than the lower limit of the range reported by Walker and Mn values fall within the lower region. Magnesium, P, K, and Ca, in the lower treatments, are in the range of compositions cited by Ohlrogge (115). Contents of both N and P are almost two times the values given by Hanway and Weber (63) for field grown plants in Iowa. Potassium is in the range given by Hanway and Weber. Compared with the data reported by Harper (64), K, Cu and, particularly, Fe are lower at this growth stage and Zn is appreciately higher.

Correlations among elements and ash in the first cutting are plotted in Figure 76. A considerable number of significant correlations exist among the elements. The large number of negative correlations associated with Ca - note particularly the correlation coefficient of -.71 associated with N and Ca - and Mg (for N-Mg the r value is -.65) are noteworthy. The positive correlations among Zn, Cd, Fe, and Mn and the other elements are also of interest, especially the very strong interaction (r = .70) between Zn and Cd.

Table 77. Effect of digested sludge and salt-simulated sludge on ash content in soybean. Data are in percent of oven-dry (60°C) tissue. Analysis of variance performed on 4 by 3 treatments for 22-day cutting and 4 by 2 treatments for 37-day cutting. Average effects for these treatments are in parentheses.

1st Cutting, 22 days

DIGESTED SLUDGE	SALT-SIMULATED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	9.61	11.62	9.73	7.99	9.74(10.32)
36	11.16	9.05	6.33	6.38	8.23(8.85)
72	9.37	7.66	5.39	6.07	7.12(7.47)
144	8.16	7.50	6.36	--	(7.34)
Average Effect	(9.58)	(8.96)	(6.95)	6.81	

Least significant difference (P)

Interaction 1.21 (99:1)
Average effect
Salt-simulated sludge 0.61 (99:1)
Digested sludge 1.21 (99:1)

2nd Cutting, 37 days

DIGESTED SLUDGE	SALT-SIMULATED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	7.00	10.93	14.66	--	10.86(8.96)
36	12.64	16.62	12.54	--	13.93(14.63)
72	14.00	13.38	--	--	(13.69)
144	13.09	12.67	--	--	(12.88)
Average Effect	(11.69)	(13.40)	13.60		

Least significant difference

Interaction 1.16 (99:1)
Average effect
Salt-simulated sludge 0.58 (99:1)
Digested sludge 0.58 (99:1)

Table 78. Effect of digested sludge and salt-simulated sludge on dry matter production in soybean. Data are in grams per plant reported on oven-dry (60°C) basis. Analysis of variance performed on 4 by 3 treatments for 22-day cutting and 4 by 2 treatments for 37-day cutting. Average effects for these treatments are in parentheses.

1st Cutting, 22 days

DIGESTED SLUDGE	SALT-SIMULATED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	1.26	1.09	.97	1.08	1.10(1.04)
36	.95	.96	.88	.73	.88(0.93)
72	.97	.87	.99	.55	.84(0.94)
144	.93	.89	.83	--	(.88)
Average Effect	(1.03)	(.95)	(.92)	.79	

Least significant difference (P)

Interaction None
Average effect
Salt-simulated sludge None
Digested sludge .09 (19:1)

2nd Cutting, 37 days

DIGESTED SLUDGE	SALT-SIMULATED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	1.37	3.14	1.66	--	2.06(2.25)
36	1.37	.76	.34	--	.82(1.06)
72	1.06	.59	--	--	(.82)
144	.94	.67	--	--	(.80)
Average Effect	(1.18)	(1.29)	1.00		

Least significant difference (P)

Interaction 0.82 (99:1)
Average effect
Salt-simulated sludge None
Digested sludge 0.41 (99:1)

Table 79. Effect of digested sludge and salt-simulated sludge on leaf surface area of all leaves of the largest plant in each pot. Data are in square centimeters. Analysis of variance performed on 4 by 2 treatments. Average effects for these treatments are in parentheses.

DIGESTED SLUDGE	SALT-SIMULATED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	127	376	174	56	183(251)
36	123	95	--	--	(109)
72	131	56	--	--	(94)
144	91	75	--	--	(83)
Average Effect	(118)	(150)	174	56	

Least significant difference (P)

Interaction 72 (99:1)
Average effect
Salt-simulated sludge none
Digested sludge 36 (99:1)

Table 80. Effect of digested sludge and salt-simulated sludge on leaf surface of fourth top leaf of the tallest plant in each pot. Data are in square centimeters. Analysis of variance performed on 4 by 2 treatments. Average effects for these treatments are in parentheses.

DIGESTED SLUDGE	SALT-SIMULATED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	18	25	21	9	18(22)
36	17	12	4	--	11(14)
72	14	8	--	--	(11)
144	10	7	--	--	(8)
Average Effect	(15)	(13)	12	9	

Least significant difference (P)

Interaction 7 (99:1)
Average effect
Salt-simulated sludge none
Digested sludge 4 (99:1)

Table 81. Effects of digested sludge and salt-simulated sludge on growth rate of soybean over two periods. Data are in centimeters per day. Analysis of variance performed on 4 by 3 treatments for 12-to 21-day period and on 4 by 2 treatments for 21-to 41-day period. Average effects for these treatments are in parentheses.

12 to 21 Days

DIGESTED SLUDGE	SALT-SIMULATED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	.371	.403	.365	.184	.331(.380)
36	.319	.262	.155	--	.245(.246)
72	.275	.219	.081	--	(.192)
144	.265	.281	.109	--	(.218)
Average Effect	(.308)	(.291)	(.178)	.184	

Least significant difference (P)

Interaction None
Average effect
Salt-simulated sludge .060 (99:1)
Digested sludge .046 (99:1)

21 to 41 Days

DIGESTED SLUDGE	SALT-SIMULATED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	.258	.432	.212	--	.301(.345)
36	.196	.100	--	--	(.148)
72	.125	.133	--	--	(.129)
144	.095	.070	--	--	(.082)
Average Effect	(.168)	(.184)	.212		

Least significant difference (P)

Interaction .100 (99:1)
Average effect
Salt-simulated sludge None
Digested sludge .050 (99:1)

Table 82. Effect of digested sludge and salt-simulated sludge on weed germination. Data in number of plants germinating.

DIGESTED SLUDGE	SALT-SIMULATED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	142	70	18	5	58
36	40	43	18	2	26
72	45	42	22	2	28
144	33	15	8	1	14
Average Effect	65	42	16	2	

Table 83. Germination of soybean seed in soil used for unleached experiment. Data are in percent of seed planted. Only one trial per treatment was performed.

DIGESTED SLUDGE	SALT-SIMULATED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	94	86	94	91	91
36	100	92	98	55	86
72	94	91	88	41	78
144	56	87	86	8	59
AVERAGE Effect	86	86	92	49	

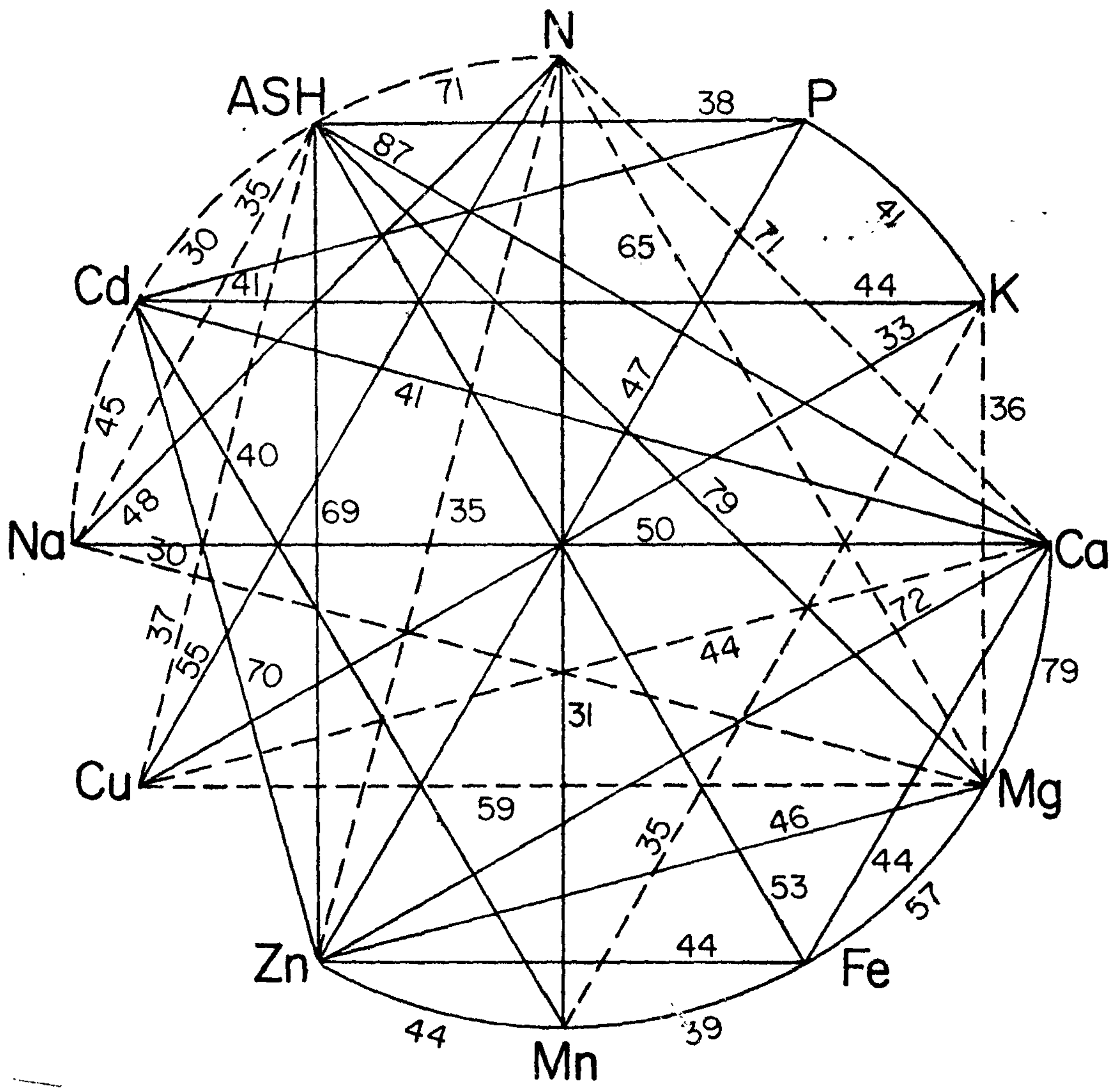


Figure 76. Graphical representation of simple linear correlations among macroelements and microelements in soybean aerial plant parts harvested at 22 days, the beginning of the rapid growth phase. Coefficients greater than 28 are significant at .05 level and coefficients greater than 37 are significant at the .01 level. Dashed lines are negative correlations.

Treatment effects for elemental contents of the second cutting (37 days), or the initiation of blooming - Except for the intermediate level of salt-simulated sludge at the zero level of digested sludge, all treatment blocks at the intermediate and highest treatment levels of salt-simulated sludge were lost because of death during the time interval between the first and second cuttings (compare, e.g., cuttings in Table 78 above). The data for 0 and 36 t/ha of simulated sludge across the four treatment levels of digested sludge were treated by analysis of variance. Zinc, Mg, Ca, P, Na, N, Cd, and ash show interaction and main effects for both sludge and salt-simulated treatments. Manganese and K show highly significant sludge treatment and interaction effects. Copper levels are affected by salt-simulated treatments and interaction effects are present. Iron is conspicuous for absence of treatments and interaction effects of either kind. Nitrogen and Mn, particularly with digested sludge, and Ca increase in content with treatment, whereas P declines. Compared with the first cutting, Cu undergoes a two-fold increase in content. Yield of the second cutting shows a highly significant main effect with sludge treatment and there is a highly significant interaction effect. Growth rate measured from the 21st through 41st day (Table 81), leaf surface area of all leaves on the 35th day and of the fourth leaf on the 45th day (Table 79), and evaporation on the day of harvest (data not presented here) all show significant main effect for digested sludge treatment and interaction with the salt-simulated sludge. Evaporation also is influenced by simulated sludge (Table 84).

Compared with values for soybean tissue reported by Walker (164) for soybeans in Illinois, Ca, Mn, Zn, Cu, and Na contents are very high. Potassium contents fall within the lower range as reported by Walker. Phosphorus, K, Ca, and Mg are in the range of contents summarized by Ohlrogge (115) whereas N, as in the first cutting, remains high - almost two times the upper range found by Ohlrogge. Nitrogen is also above the highest contents found by Hanway and Weber (63), and P is two times that in field-produced plants at a similar growth stage. With regard to N contents, it should be noted that only roots of plants in the control and lowest simulated sludge treatment were nodulated. Potassium is at the lowest level observed by Hanway and Weber for nodulating plants without K fertility. Potassium and Fe are about four times lower and Cu two times lower than Harper (64) found at this growth stage, whereas, Ca, Mg and Mn are about two times higher.

Correlations among elements and ash in the second cutting are plotted in Figure 77. As in the first cutting, a considerable number of significant correlations exist among the elements. The fewer negative correlations associated with Ca and Mg are noteworthy with the high positive correlations between Ca and Mg (r = .92) and Na and N (r = .90) especially strong. Copper and Cd are negatively correlated in contrast to the positive correlation of Cd and Zn. Modest but noteworthy correlations of the first cutting not existing in the second cutting are N and Zn, P and K, Fe and Mg, Fe and Mn, and Cd and Na.

Table 84. Table of F values for salt-simulated sludge and liquid digested sludge effects and their interactions on contents of various elements in the aerial portion of soybean and on dry matter production (yield), growth rate, leaf surface area, evaporation, number of weeds, and soil pH. F ratios for significance at the five and one percent levels are given, in allother cases the asterisks have their usual meaning. This table summarizes the effects described in the least significant difference captions in previous tables.

EFFECT	N	P	K	Ca	Mg	Fe	Mn	Zn	Cu	Na	Ni	Cd	Ash	F/.05	F/.01
							SOIL EXTRACT								
Salt		189.7	210.80	127.2	7.96	332.8	29.16	579.3	230.5	8.03	143.9	1106.5	40.83	2.92	4.51
Sludge		96.50	106.95	127.8	19.70	37.19	9.54	280.4	204.2	237.7	13.19	74.9	51.45	2.92	4.51
Interaction		20.31	24.25	1.46	1.01	3.43	3.04	5.10	1.87	1.89	2.22	2.81	0.76	2.21	3.07
							FIRST CUTTING, 22 DAYS								
Salt	81.29	2.22	9.89	65.37	237.20	9.72	16.54	11.45	6.85	8.89	n.d.[1]	27.86	81.20	3.44	5.72
Sludge	152.52	6.06	12.82	108.01	53.68	2.59	15.24	13.61	3.39	59.51		20.70	63.08	3.05	4.82
Interaction	53.14	3.88	6.83	20.49	3.09	3.31	15.48	37.30	6.84	1.99		32.27	15.94	2.55	3.76
							SECOND CUTTING, 37 DAYS								
Salt	45.78	24.73	0.01	34.15	10.54	0.54	2.12	4.84	8.21	44.39	n.d.	97.70	77.77	4.60	8.86
Sludge	225.98	9.95	9.44	131.65	30.17	1.64	135.20	4.99	3.08	83.49		15.89	162.76	4.60	8.86
Interaction	4.77	53.45	5.01	20.17	10.09	1.65	50.78	7.67	8.51	11.75		9.69	43.89	3.11	5.56

EFFECT	YIELD		GROWTH RATE		LEAF SURFACE		EVAPORATION	pH
	22 Day	37 Day	1[2]	2[3]	35 Day	45 Day		
Salt	0.88	0.58	22.77**	0.80	2.25	7.34	17.67**	20.95**
Sludge	3.32*	24.94**	22.76**	47.38**	22.73**	42.25**	74.27**	16.98**
Interaction	.78	16.46**	2.27	11.60**	5.79**	36.45**	3.23**	0.54

[1] not determined

[2] 12 through 21 days

[3] 21 through 41 days

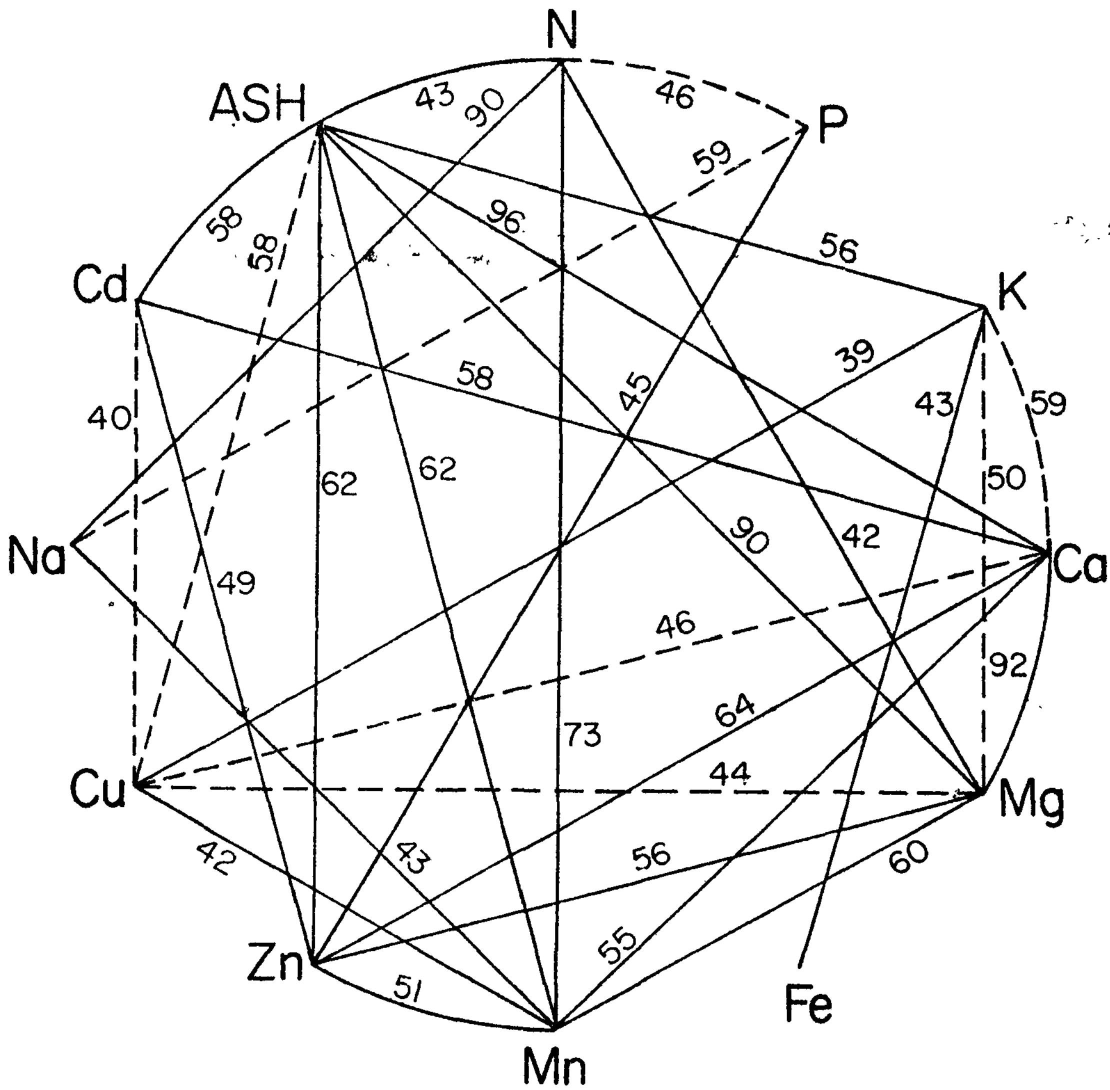


Figure 77. Graphical representation of simple linear correlations among macroelements in soybean aerial plant parts harvested at 37 days, appearance of first blooms. Numbers are correlation coefficients multiplied by 100. Coefficients greater than 33 are significant at the .05 level and coefficients greater than 40 are significant at the .01 level. Dashed lines are negative correlations.

Effect of sodium on soybean - To assess the effect on plant growth of the level of Na occurring in the digested sludge utilized in the experiment, two treatments of salt-simulated sludge were prepared using Elliott silt loam. One treatment was prepared at the level of 18 and one at 36 t of solids/ha. The reader will recall that Na was not added to the salt combination (Table 61). After the plants had grown 30 days, Na, as NaCl, equivalent to a sludge application of 144 t of solids/ha was added to the lower treatment and Na, as CH_3COONa, equivalent to the same sludge application was added to the higher salt-simulated treatment. After seven days the plants in both treatments developed chlorosis, browning and drying symptoms indistinguishable from those observed in the factorial experiment.

Relationships among cuttings and soil extracts - Data for contents of P, K, Ca, Mg, Fe, Mn, Zn, Cu, Na, Ni, Cd, and Pb in the acid extracts of the soils are given in Tables 85 through 96, respectively. These data are set down so that the reader can observe the similar amounts of the elements extractable with dilute acid from either salt-simulated or digested sludge. Inspection of the average effects of each treatment indicates that only Fe, Cd and Ni differ markedly, there being a substantial interaction effect of unknown source for Fe. Of course, Na was not added in the salt mixture. These data and similar results laid out in part two of this report can be taken as empirical substantiation of the choice of salts and method of soil preparation.

Values for correlation coefficients of the elements as determined in the two cuttings and in the acid extracts of the soils of the respective treatments are arranged in Table 97. For the elements analyzed, only Mn and P in the first cutting are not significantly correlated with the acid-extract contents of these elements. Among the elements that are significantly correlated, Zn, Fe, Mn, Ca, Mg, and P are negatively correlated. These would usually be considered to correlate positively with the extract values. Correlation of elements in the second cutting with the same extract values results in notably fewer significant relationships, probably in response to the considerable physiological stress experienced during the period between cuttings. Sodium, P and Cd are the only elements that can be correlated with the soil extract, and among these P is negative. Between cuttings, K, Mg, Fe, and Cu are negatively correlated and Mn, Na and Cd are positively correlated. Extractable Na is especially highly correlated with plant levels, a fact that lends support to the contention that salinity has affected plant development. The increase in Cd is important because of concern for this potentially toxic element to animals.

Summary and conclusions - Under the conditions of moisture content maintained, that is, at or slightly above field capacity, and the levels of sludge applied in this experiment, considerably negative effects on plant growth are to be expected. The effects of suppression of growth and toxicity are expressed primarily through osmotic influences on soil

Table 85. Effect of digested sludge and salt-simulated sludge on P in 0.1 N HCl extracts of soils in which soybeans were grown. Data in parts per million oven-dry (110°C) soil.

DIGESTED SLUDGE	SALT-SIMULATED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	60	280	550	1077	494
36	210	413	727	1137	622
72	377	623	927	1220	787
144	617	863	1113	3013	1402
AVERAGE EFFECT	316	545	829	1612	

Least significant difference

Interaction 319 (99:1)
Average effect
Digested sludge 101 (99:1)
Salt-Simulated sludge 159 (99:1)

Table 86. Effect of digested sludge and salt-simulated sludge on K in 0.1 N HCl extracts of soils in which soybeans were grown. Data in parts per million oven-dry (110°C) soil.

DIGESTED SLUDGE	SALT-SIMULATED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	156	195	254	377	246
36	172	225	287	413	274
72	208	261	330	418	304
144	265	331	399	885	470
AVERAGE EFFECT	200	253	318	523	

Least significant difference

Interaction 76 (99:1)
Average effect
Salt-simulated sludge 38 (99:1)
Digested sludge 24 (99:1)

Table 87. Effect of digested sludge and salt-simulated sludge on Ca in 0.1 N HCl extracts of soils in which soybeans were grown. Data in percent of oven-dry (110°C) soil.

DIGESTED SLUDGE	SALT-SIMULATED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	.37	.46	.48	.64	.49
36	.44	.52	.59	.70	.56
72	.49	.59	.68	.72	.62
144	.62	.69	.80	.85	.74
Average Effect	.48	.56	.64	.73	

Least significant difference

Interaction .05 (19:1)
Average effect
Salt-simulated sludge .04 (99:1)
Digested sludge .02 (99:1)

Table 88. Effect of digested sludge and salt-simulated sludge on Mg in 0.1 N HCl extracts of soils in which soybeans were grown. Data are in parts per million of oven-dry (110°C) soil.

DIGESTED SLUDGE	SALT-SIMULATED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	780	796	744	902	806
36	848	753	826	1006	858
72	918	852	920	937	907
144	978	996	1060	1148	1046
Average Effect	881	849	888	998	

Least significant difference

Interaction 134 (19:1)

Average effect

Digested sludge 57 (99:1)

Salt-simulated sludge 90 (99:1)

Table 89. Effect of digested sludge and salt-simulated sludge on Fe in 0.1 N HCl extracts of soils in which soybeans were grown. Data are in parts per million of oven-dry (110°C) soil.

DIGESTED SLUDGE	SALT-SIMULATED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	233	340	519	1097	547
36	425	399	612	1596	758
72	557	457	633	1441	772
144	652	579	791	1751	943
Average Effect	467	444	639	1471	

Least significant difference

Interaction 207 (99:1)

Average effect

Salt-simulated sludge 103 (99:1)

Digested sludge 65 (99:1)

Table 90. Effect of digested sludge and salt-simulated sludge on Mn in 0.1 N HCl extracts of soils in which soybeans were grown. Data are in parts per million of oven-dry (110°C) soil.

DIGESTED SLUDGE	SALT-SIMULATED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	145	222	231	276	218
36	194	199	228	284	226
72	265	233	220	315	258
144	200	255	288	330	268
Average Effect	201	227	242	301	

Least significant difference

Interaction 61 (99:1)

Average effect

Salt-simulated sludge 30 (99:1)

Digested sludge 19 (99:1)

Table 91. Effect of digested sludge and salt-simulated sludge on Zn in 0.1 N HCl extracts of soils in which soybeans were grown. Data are in parts per million of oven-dry (110°C) soil.

DIGESTED SLUDGE	SALT-SIMULATED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	12	115	217	418	190
36	84	165	303	483	259
72	158	264	382	510	328
144	293	392	453	587	431
Average Effect	137	234	339	500	

Least significant difference

Interaction 47 (99:1)

Average effect

Salt-simulated sludge 24 (99:1)

Digested sludge 15 (99:1)

Table 92. Effect of digested sludge and salt-simulated sludge on Cu in 0.1 N HCl extracts of soils in which soybeans were grown. Data in parts per million of oven-dry (110°C) soil.

DIGESTED SLUDGE	SALT-SIMULATED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	12	25	51	86	44
36	27	45	70	100	60
72	52	68	88	116	81
144	80	96	112	131	105
Average Effect	43	58	80	108	

Least significant difference

Interaction 11 (19:1)
Average effect
Digested sludge 4 (99:1)
Salt-simulated sludge 7 (99:1)

Table 93. Effect of digested sludge and salt-simulated sludge on Na in 0.1 N HCl extracts of soils in which soybeans were grown. Data are in parts per million of oven-dry (110°C) soil.

DIGESTED SLUDGE	SALT-SIMULATED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	79	52	64	86	70
36	110	96	96	142	111
72	160	130	143	132	141
144	234	215	241	252	236
Average Effect	146	123	136	153	

Least significant difference

Interaction 26 (19:1)
Average effect
Digested sludge 11 (99:1)
Salt-simulated sludge 18 (99:1)

Table 94. Effect of digested sludge and salt-simulated sludge on Ni in 0.1 N HCl extracts of soils in which soybeans were grown. Data are in parts per million of oven-dry (110°C) soil.

DIGESTED SLUDGE	SALT-SIMULATED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	3.0	5.2	7.7	11	6.7
36	4.3	5.7	7.8	10	7.0
72	5.0	6.5	8.3	10	7.4
144	6.2	7.8	9.0	11	8.5
Average Effect	4.6	6.3	8.2	10	

Least significant difference

Interaction 1.2 (19:1)

Average effect

Salt-simulated sludge 0.8 (99:1)

Digested sludge 0.5 (99:1)

Table 95. Effect of digested sludge and salt-simulated sludge on Cd in 0.1 N HCl extracts of soils in which soybeans were grown. Data are in parts per million of oven-dry (110°C) soil.

DIGESTED SLUDGE	SALT-SIMULATED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	1.0	6.5	14	16	11
36	2.8	8.2	16	26	13
72	4.3	11	18	27	15
144	7.9	14	19	27	17
Average Effect	4.0	9.9	17	26	

Least significant difference

Interaction 2.2 (99:1)

Average effect

Salt-simulated sludge 1.1 (99:1)

Digested sludge 0.7 (99:1)

Table 96. Effect of digested sludge and salt-simulated sludge on Pb in 0.1 N HCl extracts of soil in which soybeans were grown. Data are in parts per million of oven-dry (110°C) soil.

DIGESTED SLUDGE	SALT-SIMULATED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	15	19	34	61	32
36	26	31	50	71	44
72	47	50	64	95	64
144	64	73	96	99	83
Average Effect	38	43	61	82	

Least significant difference

Interaction 18 (19:1)

Average effect

Salt-simulated sludge 12 (99:1)

Digested sludge 7 (99:1)

Table 97. Simple linear correlation matrices for several elements among analyses of tissues from two successive cuttings of soybean and 0.1 N HCl extract of soil in which plants were grown. First cutting correlations are based on 45 observations, whereas second cutting correlations are based on 27 observations.

	CUTTING			CUTTING	
	1st	2nd		1st	2nd
PHOSPHORUS			MANGANESE		
2nd cutting	.281		2nd cutting	.488**	
Soil	-.048	-.417*	Soil	-.061	.320
POTASSIUM			ZINC		
2nd cutting	-.621**		2nd cutting	.194	
Soil	.330**	.092	Soil	-.440**	.114
CALCIUM			COPPER		
2nd cutting	-.189		2nd cutting	-.455*	
Soil	-.809	.306	Soil	.551**	-.330
MAGNESIUM			SODIUM		
2nd cutting	-.603**		2nd cutting	.535**	
Soil	-.424**	.078	Soil	.836**	.762**
IRON			CADMIUM		
2nd cutting	-.158		2nd cutting	.723**	
Soil	-.470**	.035	Soil	.162	.790**

water and subsequently on elemental absorption by the plant. The effect of Na appears strongest among the elements analyzed, although the effects of other elements under the conditions of salt-induced physiological stress are not evaluated. Nitrogen, P and Zn appear to be easily available from sludge sources to soybeans and occurred at levels in aerial plant parts up to the time of first bloom exceeding those found in field-cultivated plants. Germination is inhibited by sludge but this effect is somewhat counteracted by simulated sludge.

Effects of Digested Sewage Sludge Added to Soil on Growth and Composition of Soybean: Part II

Introduction - In part one of the experiment described here application of relatively high amounts of liquid anaerobically digested sludge and salt-simulated sludge to soil created salt conditions unfavorable for growth of soybeans. Results are presented here for the growth of soybeans in the same treatments after the treated soils had been leached to a conductivity nearly equivalent to that of the untreated or control soil. Field conditions such as those represented in this experiment would be expected in a humid climate where soluble salts are removed by percolating ground water.

Methods - Soils from each of the treatments used in the earlier experiment were leached to a conductivity in the leachate of less than 1.0 mmhO/cm, slightly higher than the conductivity level of the leachate from untreated Elliott silt loam used in the trial. After leaching, the soils were crushed, mixed and returned to their respective plastic pots and 12 soybean [*Glycine max* (L.) Merr. var. Corsoy] seedlings 2-3 cm in height were planted in each pot. Moisture content of the soil was maintained, by weighing, at 37.5 percent. The plants were grown under greenhouse conditions during late July, August, September, and October. Daylength after August 10 was controlled at 14 hours. The mean high and low temperatures for the period of the experiment were 86.3 and 63.8°C, respectively. After 19 days, six plants were harvested, washed with distilled water, dried and ground in a Wiley Mill to less than 40 mesh for analysis. Similarly, three plants were harvested after 30 days. The remaining three plants were left to mature - 93 days - and seed as well as the aerial plant parts were harvested for analysis. Germination in the soil of each treatment was assessed by placing 100 seeds in Petri dishes containing the particular treatment and moistening the soil. The number of seeds germinating was counted from days 3 through 5.

Tissue was analyzed for Ca, Mg, Fe, Mn, Zn, Cu, Cd, Cr, and Ni by atomic absorption analysis after ashing at 500°C and dissolving the ash in hydrochloric acid. Phosphorus was determined by the vanadomolybdate yellow method and N by the Kjeldahl method. Sodium and K were analyzed by flame emission spectroscopy. Also, the above elements, with the exception of N, were determined in a 0.1 *N* HCl extract of the soil using a ratio of 0.5 gm soil to 10 ml acid and two-hour equilibration time. A 1 to 1 soil-water paste was prepared for pH determination.

Results and discussion, general statement - All treatments supported plants to maturity, although one replicate in the treatment combining maximum rates of salt-simulated sludge and digested sludge at the maximum rate of 144 t of solids/ha of each and one replicate in the treatment combining salt-simulated sludge at the rate of 72 t of solids/ha with the maximum rate of digested sludge, did not produce pods. In subsequent analysis and correlation of the third cutting and seed data, values for these missing data were calculated as the average of the remaining two replicates within the treatment. Germination of soybean seeds in the soil after leaching experienced none of the adverse treatment effects occurring before leaching, with from 96 to 100 percent of the seeds germinating in all treatment rates. Soil pH (Table 98) is closely related to sludge treatment, with a negative simulated-sludge interaction. For all, except perhaps the lowest salt treatments, pH is satisfactory for soybean nutrition.

Treatment effects of sludge and simulated sludge on soil levels of macro- and microelements - On extraction of elements from soil, the levels of all elements measured in 0.1 N HCl extracts (Tables 99 through 110 for P, K, Ca, Mg, Fe, Mn, Zn, Cu, Na, Ni, Cd, and Pb, respectively) are very significantly ($P < .01$ in all cases, treatment effects collected in Table 138) affected by the treatments. Also, comparison of the mean effects for each sludge type for the elements considered suggests that the original levels added, the process of leaching and the acid extraction employed combine to yield remarkably similar concentrations for each incremental increase of sludge type added. These similarities are empirical evidence that the attempt to simulate the levels and availability of the elements in sludge was successful. Of special interest is the fact that interaction effects between the treatments occur for all elements except Zn, Ni and Cu. Correlation of the leached values with unleached data indicate that levels of Zn, Mg and Cd extracted after the leaching procedure are nearly equivalent to those extracted before leaching (for concentrations prior to leaching compare Tables 85 through 96 in part one of this section). Copper, Fe, Mn, Ca, P, K, and Ni are from one-half to three-quarters of the unleached levels. In contrast, for simulated treatments, Na levels decline to about one-third of those in the unleached series and Pb is about one-third the unleached values in both sludge and simulated series. Coefficients of determination for Zn, Ca, P, K, Cu, and Cd in the above comparisons were above 0.80. The lowest coefficients were Na at 0.29, a reflection of the rather complete removal of readily accessible sources of this ion by the leaching procedure, and Mn at 0.32, perhaps a reflection of transformation of this oxidation responsive ion into forms not readily available to hydrochloric acid. It should be noted that Na was the only element among the analytes not added in the simulated sludge. It occurs at concentrations of about 125 ppm in the sludge used in the experiment.

Treatment effects, first cutting or beginning of rapid growth phase - Data for yield of the first cutting are presented in Table 111. Data for tissue analyses of the elements determined in the plants at 19 days are presented in Tables 112 through 136. The tables are arranged in the order N, P, K, Ca, Mg, Fe, Mn, Zn, Cu, Na, Ni, Cd, and ash weight.

Table 98. Effects of digested sludge and salt-simulated sludge on pH of soil in which soybeans were grown. Determinations made on a 1:1, by weight, soil-water paste.

SIMULATED SLUDGE	DIGESTED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	6.3	6.0	6.1	6.5	6.2
36	6.4	6.4	6.4	6.5	6.4
72	6.5	6.6	6.8	6.9	6.7
144	7.3	7.3	7.4	6.6	7.1
AVERAGE EFFECT	6.6	6.6	6.7	6.6	

Least significant difference (P)
Interaction = 0.2 (99:1)
Average effect
Salt-simulated sludge = 0.1 (99:1)

Table 99. Effects of digested sludge and salt-simulated sludge on P contents in 0.1 N HCl extracts of soils in which soybeans were grown. Data are reported in parts per million of oven-dry (110°C) soil.

SIMULATED SLUDGE	DIGESTED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	103	200	427	747	369
36	157	387	607	563	428
72	390	647	857	1177	768
144	837	947	1157	1973	1228
AVERAGE EFFECT	372	545	762	1115	

Least significant difference (P)

Interaction = 168 (99:1)
Average effect
Digested sludge = 53 (99:1)
Salt-simulated sludge = 84 (99:1)

Table 100. Effects of digested sludge and salt-simulated sludge on K contents in 0.1 N HCl extracts of soils in which soybeans were grown. Data are reported in parts per million of oven-dry (110°C) soil.

SIMULATED SLUDGE	DIGESTED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	110	117	150	197	143
36	110	173	197	230	178
72	160	213	240	267	220
144	287	333	357	480	364
AVERAGE EFFECT	167	209	236	293	

Least significant difference (P)
Interaction = 23 (99:1)
Average effect
Digested sludge = 7 (99:1)
Salt-simulated sludge = 11 (99:1)

Table 101. Effects of digested sludge and salt-simulated sludge on Ca contents in 0.1 N HCl extracts of soils in which soybeans were grown. Data are reported in percent of oven-dry (110°C) soils.

SIMULATED SLUDGE	DIGESTED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	.30	.29	.30	.38	.32
36	.30	.30	.35	.43	.34
72	.33	.36	.42	.49	.40
144	.44	.50	.52	.59	.51
AVERAGE EFFECT	.34	.36	.40	.47	

Least significant difference
Interaction = .03 (99:1)
Average effect
Digested sludge = .01 (99:1)
Salt-simulated sludge = .01 (99:1)

Table 102. Effects of digested sludge and salt-simulated sludge on Mg contents in 0.1 N HCl extracts of soils in which soybeans were grown. Data are reported in parts per million of oven-dry (110°C) soil.

SIMULATED SLUDGE	DIGESTED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	747	660	667	783	714
36	670	643	733	880	732
72	627	693	853	990	791
144	837	930	1013	1230	1002
AVERAGE EFFECT	720	732	817	971	

Least significant difference (P)
Interaction = 76 (99:1)
Average effect
Digested sludge = 24 (99:1)
Salt-simulated sludge = 38 (99:1)

Table 103. Effects of digested sludge and salt-simulated sludge on Fe contents in 0.1 N HCl extracts of soils in which soybeans were grown. Data are reported in parts per million of oven-dry (110°C) soil.

SIMULATED SLUDGE	DIGESTED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	188	307	348	702	386
36	240	299	480	774	448
72	333	397	594	973	574
144	543	793	989	1363	922
AVERAGE EFFECT	326	449	603	953	

Least significant difference (P)
Interaction = 124 (19:1)
Average effect
Digested sludge = 53 (99:1)
Salt-simulated sludge = 84 (99:1)

Table 104. Effects of digested sludge and salt-simulated sludge on Mn contents in 0.1 N HCl extracts of soils in which soybeans were grown. Data are reported in parts per million of oven-dry (110°C) soil.

SIMULATED SLUDGE	DIGESTED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	127	169	214	271	195
36	129	224	255	256	216
72	168	216	250	256	222
144	226	226	251	348	262
AVERAGE EFFECT	162	209	242	283	

Least significant difference (P)
Interaction = 50 (99:1)
Average effect
Digested sludge = 16 (99:1)
Salt-simulated sludge = 25 (99:1)

Table 105. Effects of digested sludge and salt-simulated sludge on Zn contents in 0.1 N HCl extracts of soils in which soybeans were grown. Data are reported in parts per million of oven-dry (110°C) soil.

SIMULATED SLUDGE	DIGESTED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	12	77	141	268	124
36	102	175	246	392	229
72	201	281	357	476	329
144	403	470	532	681	522
AVERAGE EFFECTS	180	251	319	454	

Least significant difference (P)
Interaction = 25 (19:1)
Average effect
Digested sludge = 11 (99:1)
Salt-simulated sludge = 17 (99:1)

Table 106. Effects of digested sludge and salt-simulated sludge on Cu contents in 0.1 N HCl extracts of soils in which soybeans were grown. Data are reported in parts per million of oven-dry (110°C) soil.

SIMULATED SLUDGE	DIGESTED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	3.2	13	30	56	26
36	15	27	44	65	38
72	31	42	61	81	53
144	61	70	85	110	81
AVERAGE EFFECT	27	38	55	78	

Least significant difference (P)
Interaction = 9 (19:1)
Average effect
Digested sludge = 4 (99:1)
Salt-simulated sludge = 6 (99:1)

Table 107. Effects of digested sludge and salt-simulated sludge on Na contents in 0.1 N HCl extracts of soils in which soybeans were grown. Data are reported in parts per million of oven-dry (110°C) soil.

SIMULATED SLUDGE	DIGESTED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	37	40	50	62	47
36	42	45	50	54	48
72	22	33	42	51	37
144	32	39	73	50	48
AVERAGE EFFECT	33	39	54	54	

Least significant difference (p)
Interaction = 14 (99:1)
Average effect
Digested sludge = 4 (99:1)
Salt-simulated sludge = 7 (99:1)

Table 108. Effects of digested sludge and salt-simulated sludge on Ni contents in 0.1 N HCl extracts of soils in which soybeans were grown. Data are reported in parts permillion of oven-dry (110°C) soil.

SIMULATED SLUDGE	DIGESTED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	2.9	4.1	5.3	5.9	4.6
36	4.5	5.3	6.7	7.2	5.9
72	6.3	5.2	6.5	6.7	6.2
144	8.1	8.0	8.1	8.6	8.2
AVERAGE EFFECT	5.5	5.7	6.7	7.1	

Least significant difference (P)
Interaction = 1.8 (19:1)
Average effect
Digested sludge = 0.8 (99:1)
Salt-simulated sludge = 1.2 (99:1)

Table 109. Effects of digested sludge and salt-simulated sludge on Cd contents in 0.1 N HCl extracts of soils in which soybeans were grown. Data are reported in parts per million of oven-dry (110°C) soil.

SIMULATED SLUDGE	DIGESTED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	.1	1.8	3.7	7.4	3.2
36	5.9	7.6	9.3	12	8.8
72	11	13	15	18	14
144	23	23	23	34	26
AVERAGE EFFECT	10	11	13	18	

Least significant difference (P)
Interaction = 3.3 (99:1)
Average effect
Digested sludge = 1.0 (99:1)
Salt-simulated sludge = 1.7 (99:1)

Table 110. Effects of digested sludge and salt-simulated sludge on Pb contents in 0.1 N HCl extracts of soils in which soybeans were grown. Data are reported in parts per million of oven-dry (110°C) soil.

SIMULATED SLUDGE		DIGESTED SLUDGE			
t/ha	0	36	72	144	Average Effect
0	1.3	9.2	16	23	12
36	8.7	15	21	23	17
72	19	24	29	31	26
144	24	27	26	11	22
AVERAGE EFFECT	13	19	23	22	

Least significant difference (P)
Interaction = 3.7 (99:1)
Average effect
Digested sludge = 1.2 (99:1)
Salt-simulated sludge = 1.8 (99:1)

Table 111. Effects of digested sludge and salt-simulated sludge on dry matter production (aerial plant parts). Data are reported in grams of oven-dry (60°C) tissue for 3 plants in cutting 2 and 3 and 6 plants in cutting 1.

	FIRST CUTTING - 21 DAYS					SECOND CUTTING - 32 DAYS					THIRD CUTTING - 95 DAYS				
SIMULATED SLUDGE	DIGESTED SLUDGE				AVERAGE EFFECT	DIGESTED SLUDGE				AVERAGE EFFECT	DIGESTED SLUDGE				AVERAGE EFFECT
t/ha	0	36	72	144		0	36	72	144		0	36	72	144	
0	.91	.93	1.14	.86	.96	3.24	2.27	1.86	2.28	2.41	2.04	4.02	5.73	8.02	4.95
36	1.03	1.07	1.18	1.22	1.12	2.70	2.93	1.62	2.86	2.53	4.02	4.20	8.00	7.40	5.91
72	1.03	1.29	.88	1.09	1.07	1.79	2.28	2.37	2.06	2.12	4.32	7.34	7.44	7.98	6.77
144	1.19	.99	1.04	1.12	1.08	3.57	2.71	1.27	2.13	2.42	7.12	6.11	7.02	5.48	6.43
AVERAGE EFFECT	1.04	1.07	1.06	1.07		2.82	2.55	1.78	2.33		4.37	5.42	7.05	7.22	

First cutting: Least significant difference (P)
Interaction = .33 (99:1)

Second cutting: Least significant difference (P)
Interaction = 1.02 (99:1)
Average effect
Digested sludge = .43 (99:1)

Third cutting: Least significant difference (P)
Interaction = 4.14 (99:1)
Average effect
Digested sludge = 1.31(99:1)
Salt-simulated sludge = 2.07 (99:1)

Table 112. Effects of digested sludge and salt-simulated sludge on N contents in soybean tissues at three stages of growth. Data are reported in percent of oven-dry (60°C) tissue.

FIRST CUTTING - 21 DAYS

SIMULATED SLUDGE	DIGESTED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	3.60	3.91	4.59	5.32	4.38
36	3.84	4.37	5.60	5.63	4.86
72	3.66	5.10	5.94	6.18	5.22
144	4.92	5.37	5.70	6.41	5.60
AVERAGE EFFECT	4.03	4.69	5.46	5.88	

Least significant difference (P) -
Interaction = .63 (19:1)
Average effect
Digested sludge = .27 (99:1)
Salt-simulated sludge = .42 (99:1)

SECOND CUTTING - 32 DAYS

SIMULATED SLUDGE	DIGESTED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	4.57	3.96	5.64	4.79	4.74
36	4.18	4.64	3.63	4.62	4.27
72	3.64	3.51	4.55	3.33	3.76
144	4.97	4.43	3.52	5.52	4.61
AVERAGE EFFECT	4.34	4.14	4.34	4.56	

Least significant difference (P)
Interaction = .86 (99:1)
Average effect
Salt-simulated sludge = .43 (99:1)

THIRD CUTTING - 95 DAYS

SIMULATED SLUDGE	DIGESTED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	1.42	1.32	1.39	2.00	1.54
36	1.35	1.46	1.39	1.92	1.53
72	1.53	1.68	1.66	2.65	1.88
144	1.06	1.47	3.07	4.36	2.49
AVERAGE EFFECT	1.34	1.48	1.88	2.73	

Least significant difference (P)
Interaction = 1.49 (19:1)
Average effect
Digested sludge = .47 (99:1)
Salt-simulated sludge = .75 (99:1)

Table 113. Effects of digested sludge and salt-simulated sludge on N content in soybean seed. Data is percent of oven-dry (60oC) tissue.

SIMULATED SLUDGE	DIGESTED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	6.71	6.24	6.19	6.44	6.40
36	6.53	5.74	5.38	5.87	5.88
72	5.39	6.49	5.89	6.18	5.99
144	5.18	5.92	6.51	7.03	6.16
AVERAGE EFFECT	5.95	6.10	5.99	6.38	

Least significant difference (P)
Interaction = .96 (99:1)
Average effect
Salt-simulated sludge = .36 (19:1)

Table 114. Effects of digested sludge and salt-simulated sludge on P contents in soybean tissues at three growth stages. Data are reported in percent of oven-dry (60°C) tissue.

FIRST CUTTING - 21 DAYS						SECOND CUTTING - 32 DAYS						THIRD CUTTING - 95 DAYS					
SIMULATED SLUDGE	DIGESTED SLUDGE					SIMULATED SLUDGE	DIGESTED SLUDGE					SIMULATED SLUDGE	DIGESTED SLUDGE				
t/ha	0	36	72	144	Average Effect	t/ha	0	36	72	144	Average Effect	t/ha	0	36	72	144	Average Effect
0	.28	.57	.74	.74	.58	0	.97	.73	.62	.46	.70	0	.29	.80	1.44	1.56	1.02
36	.57	.92	.93	.84	.82	36	.64	.58	.55	.78	.64	36	.66	1.29	1.68	2.11	1.43
72	1.00	1.00	.86	.71	.89	72	.54	.76	.66	1.06	.76	72	1.41	2.09	1.78	.96	1.56
144	.77	.77	.69	.84	.77	144	.73	.71	.18	.53	.54	144	1.40	.82	.67	.56	.86
AVERAGE EFFECT	.65	.82	.80	.78		AVERAGE EFFECT	.72	.70	.50	.71		AVERAGE EFFECT	.94	1.25	1.39	1.30	

Least significant difference (P)
Interaction = 0.12 (99:1)
Average effect
Simulated sludge = 0.06 (99:1)
Digested sludge = 0.04 (99:1)

Least significant difference (P)
Interaction = 0.16 (99:1)
Average effect
Simulated sludge = 0.08 (99:1)
Digested sludge = 0.05 (99:1)

Least significant difference (P)
Interaction = 0.50 (99:1)
Average effect
Simulated sludge = 0.25 (99:1)
Digested sludge = 0.16 (99:1)

Table 115. Effects of digested sludge and salt-simulated sludge on P content in soybean seed. Data are reported in percent of oven-dry (60°C) tissue.

SIMULATED SLUDGE	DIGESTED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	.52	.72	.79	.73	.69
36	.66	.80	.82	.78	.76
72	.80	.84	.78	.71	.78
144	.80	.75	.65	.70	.72
AVERAGE EFFECT	.70	.78	.76	.73	

Least significant difference (P)
Interaction = .07 (99:1)
Average effect
Salt-simulated sludge = .03 (99:1)
Digested sludge = .07 (99:1)

Table 116. Effects of digested sludge and salt-simulated sludge on K contents in soybean tissues at three stages. Data are reported in percent of oven-dry (60oC) tissue.

FIRST CUTTING - 21 DAYS

SIMULATED SLUDGE t/ha	DIGESTED SLUDGE 0	36	72	144	Average Effect
0	1.87	2.09	2.32	2.38	2.17
36	1.93	2.89	2.83	2.76	2.60
72	2.69	2.78	2.57	2.36	2.60
144	3.15	2.96	2.73	2.94	2.94
AVERAGE EFFECT	2.41	2.68	2.61	2.61	

Least significant difference (P)
Interaction = .44 (99:1)
Average effect
Digested sludge = .14 (19:1)
Salt-simulated sludge = .22 (99:1)

SECOND CUTTING - 32 DAYS

SIMULATED SLUDGE t/ha	DIGESTED SLUDGE 0	36	72	144	Average Effect
0	2.15	2.15	1.97	2.05	2.08
36	2.44	2.44	1.89	2.16	2.23
72	2.20	2.33	1.99	2.63	2.29
144	2.12	1.73	1.61	1.71	1.79
AVERAGE EFFECT	2.22	2.16	1.86	1.71	

Least significant difference (P)
Interaction
Average effect
Digested sludge = .16 (99:1)
Salt-simulated sludge = .25 (99:1)

THIRD CUTTING - 95 DAYS

SIMULATED SLUDGE t/ha	DIGESTED SLUDGE 0	36	72	144	Average Effect
0	.54	.23	.27	.75	.45
36	.18	.49	.44	.82	.48
72	.45	.90	.52	.64	.63
144	.49	.64	.76	.64	.64
AVERAGE EFFECT	.42	.57	.50	.71	

Least significant difference (P)
Interaction = .36 (99:1)
Average effect
Digested sludge = .11 (99:1)
Salt-simulated sludge = .18 (99:1)

Table 117. Effects of digested sludge and salt-simulated sludge on K content in soybean seed. Data are reported in percent of oven-dry (60°C) tissue.

SIMULATED SLUDGE	DIGESTED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	1.74	1.77	1.83	1.77	1.78
36	1.72	1.89	2.03	1.99	1.91
72	2.00	2.06	1.84	1.72	1.90
144	1.91	1.88	1.76	1.70	1.81
AVERAGE EFFECT	1.84	1.90	1.87	1.80	

Least significant difference (P)
Interaction = .15 (99:1)
Average effect
Digested sludge = .05 (99:1)
Salt-simulated sludge = .08 (99:1)

Table 118. Effects of digested sludge and salt-simulated sludge on Ca contents in soybean tissues at three growth stages. Data are reported in percent of oven-dry (60°C) tissue.

FIRST CUTTING - 21 DAYS						SECOND CUTTING - 32 DAYS						THIRD CUTTING - 95 DAYS					
SIMULATED SLUDGE	DIGESTED SLUDGE					SIMULATED SLUDGE	DIGESTED SLUDGE					SIMULATED SLUDGE	DIGESTED SLUDGE				
t/ha	0	36	72	144	Average Effect	t/ha	0	36	72	144	Average Effect	t/ha	0	36	72	144	Average Effect
0	1.37	1.45	1.39	1.25	1.36	0	1.59	1.49	.96	1.09	1.28	0	2.57	3.61	3.52	3.43	3.28
36	1.48	1.35	1.33	1.50	1.42	36	1.31	1.31	1.48	1.72	1.46	36	3.47	3.33	2.99	3.84	3.41
72	1.65	1.43	1.20	1.24	1.38	72	1.67	1.41	1.44	1.43	1.49	72	3.17	2.92	3.73	3.51	3.33
144	1.12	1.17	.99	.88	1.04	144	1.46	1.73	1.93	1.52	1.66	144	3.32	3.08	3.00	2.74	3.04
AVERAGE EFFECT	1.40	1.35	1.23	1.22		AVERAGE EFFECT	1.50	1.48	1.45	1.44		AVERAGE EFFECT	3.13	3.24	3.31	3.38	

First cutting:
Least significant difference (P)
Interaction = .17 (99:1)
Average effect
Salt-simulated sludge = .08 (99:1)
Digested sludge = .05 (99:1)

Second cutting:
Least significant difference (P)
Interaction = .42 (99:1)
Average effect
Salt-simulated sludge = .21 (99:1)

Third cutting:
Least significant difference
None

Table 119. Effects of digested sludge and salt-simulated sludge on Ca content in soybean seed. Data are reported in percent of oven-dry (60°C) tissue.

SIMULATED SLUDGE	DIGESTED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	.31	.29	.25	.26	.28
36	.31	.24	.23	.24	.26
72	.30	.25	.25	.24	.26
144	.27	.24	.27	.24	.26
AVERAGE EFFECT	.30	.26	.25	.25	

Least significant difference (P)
Interaction
Average effect
Digested sludge = .02 (99:1)

Table 120. Effects of digested sludge and salt-simulated sludge on Mg contents in soybean tissue at three growth stages. Data are reported in percent of oven-dry (60°C) tissue.

FIRST CUTTING - 21 DAYS

SIMULATED SLUDGE	DIGESTED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	.72	.62	.53	.51	.59
36	.79	.51	.51	.67	.62
72	.55	.51	.57	.57	.55
144	.53	.57	.55	.54	.55
AVERAGE EFFECT	.65	.55	.54	.57	

Least significant difference (P)
Interaction = .15 (19:1)
Average effect
Digested sludge = .05 (19:1)

SECOND CUTTING - 32 DAYS

SIMULATED SLUDGE	DIGESTED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	.66	.57	.55	.63	.60
36	.65	.60	.66	.66	.65
72	.69	.51	.52	.56	.57
144	.73	.66	.67	.66	.68
AVERAGE EFFECT	.68	.58	.60	.63	

Least significant difference (P)
Interaction
Average effect
Digested sludge = .04 (99:1)
Salt-simulated sludge = .06 (99:1)

THIRD CUTTING - 95 DAYS

SIMULATED SLUDGE	DIGESTED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	.74	1.07	.92	.91	.91
36	1.08	.80	.86	1.02	.94
72	.74	.82	1.09	1.15	.95
144	.84	.95	.97	1.03	.95
AVERAGE EFFECT	.85	.91	.96	1.02	

Least significant difference (P)
Interaction = .25 (99:1)
Average effect
Digested sludge = .08 (99:1)

Table 121. Effects of digested sludge and salt-simulated sludge on Mg content in soybean seed. Data are reported in percent of oven-dry (60°C) tissue.

SIMULATED SLUDGE	DIGESTED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	.28	.30	.27	.29	.28
36	.29	.28	.28	.29	.29
72	.28	.28	.31	.31	.29
144	.30	.30	.31	.32	.31
AVERAGE EFFECT	.29	.29	.29	.30	

Least significant difference
Interaction = .02 (19:1)
Average effect
Salt-simulated sludge = .01 (99:1)

Table 122. Effects of digested sludge and salt-simulated sludge on Fe contents in soybean tissues at three growth stages. Data are reported in parts per million of oven-dry (60°C) tissue.

FIRST CUTTING - 21 DAYS						SECOND CUTTING - 32 DAYS						THIRD CUTTING - 95 DAYS					
SIMULATED SLUDGE	DIGESTED SLUDGE					SIMULATED SLUDGE	DIGESTED SLUDGE					SIMULATED SLUDGE	DIGESTED SLUDGE				
t/ha	0	36	72	144	Average Effect	t/ha	0	36	72	144	Average Effect	t/ha	0	36	72	144	Average Effect
0	111	103	107	122	111	0	83	110	80	80	88	0	205	125	167	151	162
36	98	102	115	106	105	36	68	69	69	89	74	36	163	183	124	166	159
72	112	87	84	82	91	72	100	98	89	76	91	72	140	110	129	124	126
144	80	87	81	72	80	144	88	82	76	74	80	144	101	103	86	88	95
AVERAGE EFFECT	100	95	97	96		AVERAGE EFFECT	84	90	79	80		AVERAGE EFFECT	152	130	126	132	

First cutting:
Least significant difference (P)
Interaction
Average effect
Salt-simulated sludge = 16 (19:1)

Second cutting:
Least significant difference (P)
Interaction = 26 (99:1)
Average effect
Salt-simulated sludge = 13 (99:1)

Third cutting:
Least significant difference (P)
Interaction = 46 (19:1)
Average effect
Salt-simulated sludge = 31 (99:1)

Table 123. Effects of digested sludge and salt-simulated sludge on Fe content in soybean seed. Data are reported in parts per million of oven-dry (60°C) tissue.

SIMULATED SLUDGE	DIGESTED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	54	57	51	49	53
36	48	51	52	47	50
72	45	53	55	59	53
144	56	63	56	46	55
AVERAGE EFFECT	50	56	54	50	

Least significant difference (P)
Interaction = 7 (99:1)
Average effect
Digested sludge = 2 (99:1)
Salt-simulated sludge = 3 (99:1)

Table 124. Effects of digested sludge and salt-simulated sludge on Mn contents in soybean tissues at three stages of growth. Data are reported in parts per million of oven-dry (60°C) tissue.

FIRST CUTTING - 21 DAYS

SIMULATED SLUDGE t/ha	DIGESTED SLUDGE 0	36	72	144	Average Effect
0	95	429	486	354	341
36	61	393	456	358	317
72	118	348	304	398	292
144	187	253	288	178	226
AVERAGE EFFECT	115	356	384	322	

Least significant difference (P)
Interaction = 135 (99:1)
Average effect
Digested sludge = 43 (99:1)
Salt-simulated sludge = 68 (99:1)

SECOND CUTTING - 32 DAYS

SIMULATED SLUDGE t/ha	DIGESTED SLUDGE 0	36	72	144	Average Effect
0	438	486	238	298	365
36	255	337	73	467	283
72	442	458	397	79	344
144	487	363	96	390	334
AVERAGE EFFECT	406	411	201	308	

Least significant difference (P)
Interaction = 121 (99:1)
Average effect
Digested sludge = 38 (99:1)
Salt-simulated sludge = 61 (99:1)

THIRD CUTTING - 95 DAYS

SIMULATED SLUDGE t/ha	DIGESTED SLUDGE 0	36	72	144	Average Effect
0	158	573	1037	873	660
36	107	876	989	1001	743
72	157	717	806	793	618
144	570	488	416	441	479
AVERAGE EFFECT	248	663	812	777	

Least significant difference (P)
Interaction = 380 (99:1)
Average effect
Digested sludge = 120 (99:1)
Salt-simulated sludge = 190 (99:1)

Table 125. Effects of digested sludge and salt-simulated sludge on Mn content in soybean seed. Data are reported in parts per million of oven-dry (60°C) tissue.

SIMULATED SLUDGE	DIGESTED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	41	63	93	83	70
36	28	94	103	78	76
72	29	84	71	65	62
144	64	66	53	67	63
AVERAGE EFFECT	41	77	80	73	

Least significant difference (P)
Interaction = 24 (99:1)
Average effect
Digested sludge = 8 (99:1)
Salt-simulated sludge = 12 (99:1)

Table 126. Effects of digested sludge and salt-simulated sludge on Zn contents in soybean tissues at three growth stages. Data are reported in parts per million of oven-dry (60°C) tissue.

FIRST CUTTING - 21 DAYS						SECOND CUTTING - 32 DAYS						THIRD CUTTING - 95 DAYS					
SIMULATED SLUDGE	DIGESTED SLUDGE					SIMULATED SLUDGE	DIGESTED SLUDGE					SIMULATED SLUDGE	DIGESTED SLUDGE				
t/ha	0	36	72	144	Average Effect	t/ha	0	36	72	144	Average Effect	t/ha	0	36	72	144	Average Effect
0	60	131	163	188	135	0	185	148	159	184	170	0	82	278	314	353	257
36	107	159	183	232	170	36	154	184	119	194	163	36	196	306	327	429	314
72	172	194	208	210	196	72	146	160	201	163	168	72	266	277	310	326	295
144	155	201	213	175	186	144	212	197	59	187	164	144	254	211	315	249	257
AVERAGE EFFECT	124	171	192	201		AVERAGE EFFECT	174	172	134	182		AVERAGE EFFECT	200	268	316	339	

First cutting:
Least significant difference (P)
Interaction = 50 (99:1)
Average effect
Digested sludge = 16 (99:1)
Salt-simulated sludge = 25 (99:1)

Second cutting:
Least significant difference (P)
Interaction = 39 (99:1)
Average effect
Digested sludge = 12 (99:1)

Third cutting:
Least significant difference (P)
Interaction = 115 (99:1)
Average effect
Digested sludge = 36 (99:1)
Salt-simulated sludge = 43 (19:1)

Table 127. Effects of digested sludge and salt-simulated sludge on Zn content in soybean seed. Data are reported in parts per million of oven-dry (60°C) tissue.

SIMULATED SLUDGE	DIGESTED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	40	67	64	60	58
36	62	62	64	67	64
72	75	67	64	69	69
144	64	74	82	80	75
AVERAGE EFFECT	60	67	68	69	

Least significant difference (P)
Interaction = 5 (99:1)
Average effect
Digested sludge = 2 (99:1)
Salt-simulated sludge = 3 (99:1)

Table 128. **Effects of digested sludge and salt-simulated sludge on Cu contents in soybean tissues at three growth stages.** Data are reported in parts per million of oven-dry (60°C) tissue.

FIRST CUTTING - 21 DAYS

SIMULATED SLUDGE t/ha	DIGESTED SLUDGE 0	36	72	144	Average Effect
0	6	10	11	16	11
36	12	12	12	13	12
72	11	14	15	14	14
144	12	15	14	15	[illegible]
AVERAGE EFFECT	10	13	13	15	

Least significant difference (P)
Interaction = 3 (19:1)
Average effect
Digested sludge = 1 (99:1)
Salt-simulated sludge = 2 (99:1)

SECOND CUTTING - 32 DAYS

SIMULATED SLUDGE t/ha	DIGESTED SLUDGE 0	36	72	144	Average Effect
0	11	10	11	11	11
36	9	13	9	11	11
72	6	10	12	8	9
144	12	10	6	12	10
AVERAGE EFFECT	10	11	9	11	

Least significant difference (P)
Interaction = 4 (99:1)

THIRD CUTTING - 95 DAYS

SIMULATED SLUDGE t/ha	DIGESTED SLUDGE 0	36	72	144	Average Effect
0	10	7	5	6	7
36	7	6	5	5	6
72	5	5	4	7	5
144	4	9	11	9	8
AVERAGE EFFECT	6	7	6	7	

Least significant difference (P)
Interaction = 3 (99:1)
Average effect
Salt-simulated sludge = 1 (99:1)

Table 129. Effects of digested sludge and salt-simulated sludge on Cu content in soybean seed. Data are reported in parts per million of oven-dry (60°C) tissue.

SIMULATED SLUDGE	DIGESTED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	11	10	9	11	10
36	8	7	10	12	9
72	8	11	12	13	11
144	11	13	14	11	12
AVERAGE EFFECT	10	10	11	12	

Least significant difference (P)
- Interaction = 2 (99:1)
- Average effect
  - Digested sludge = 1 (99:1)
  - Salt-simulated sludge = 1 (99:1)

Table 130. Effects of digested sludge and salt-simulated sludge on Na contents in soybean tissues at three growth stages. Data are reported in parts per million of oven-dry (60°C) tissue.

FIRST CUTTING - 21 DAYS						SECOND CUTTING - 32 DAYS						THIRD CUTTING - 95 DAYS					
SIMULATED SLUDGE	DIGESTED SLUDGE					SIMULATED SLUDGE	DIGESTED SLUDGE					SIMULATED SLUDGE	DIGESTED SLUDGE				
t/ha	0	36	72	144	Average Effect	t/ha	0	36	72	144	Average Effect	t/ha	0	36	72	144	Average Effect
0	261	99	95	85	135	0	91	99	114	81	96	0	347	459	517	283	402
36	92	118	131	148	122	36	81	89	76	66	78	36	459	429	353	303	386
72	140	69	141	126	119	72	115	95	103	96	102	72	369	309	328	302	327
144	87	99	94	75	88	144	99	93	100	64	89	144	272	336	289	540	359
AVERAGE EFFECT	145	96	115	108		AVERAGE EFFECT	96	94	98	77		AVERAGE EFFECT	362	383	372	357	

First cutting:
Least significant difference (P)
Interaction = 81 (99:1)
Average effect
Digested sludge = 19 (19:1)
Salt-simulated sludge = 30 (19:1)

Second cutting:
Least significant difference (P)
Interaction
Average effect
Digested sludge = 9 (19:1)
Salt-simulated sludge = 19 (19:1)

Third cutting:
Least significant difference (P)
Interaction = 200 (99:1)

Table 131. Effects of digested sludge and salt-simulated sludge on Na contents in soybean seed. Data are reported in parts per million of oven-dry (60°C) tissue.

SIMULATED SLUDGE	DIGESTED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	37	33	35	32	34
36	33	36	31	39	35
72	36	27	39	32	33
144	28	20	30	32	28
AVERAGE EFFECT	34	29	34	34	

Least significant difference (P)
Interaction
Average effect
Salt-simulated sludge = 6 (99:1)

Table 132. Effects of digested sludge and salt-simulated sludge on Ni contents in soybean tissues at three growth stages. Data are reported in parts per million of oven-dry (60°C) tissue.

FIRST CUTTING - 21 DAYS						SECOND CUTTING - 32 DAYS						THIRD CUTTING - 95 DAYS					
SIMULATED SLUDGE	DIGESTED SLUDGE					SIMULATED SLUDGE	DIGESTED SLUDGE					SIMULATED SLUDGE	DIGESTED SLUDGE				
t/ha	0	36	72	144	Average Effect	t/ha	0	36	72	144	Average Effect	t/ha	0	36	72	144	Average Effect
0	4.2	14.7	8.9	8.6	9.1	0	3.2	3.2	6.0	1.8	3.6	0	2.8	2.5	1.5	1.5	2.1
36	6.5	8.3	22.8	4.5	10.5	36	2.3	6.5	3.3	71.6	20.9	36	1.0	1.7	1.2	1.3	1.3
72	5.9	5.4	6.1	7.8	6.3	72	2.1	4.0	4.6	2.6	3.3	72	1.2	1.0	1.1	1.6	1.2
144	4.5	7.4	5.1	5.4	5.6	144	3.8	2.5	1.4	2.4	2.5	144	3.8	1.1	1.5	2.6	2.2
AVERAGE EFFECT	5.3	8.9	10.7	6.6		AVERAGE EFFECT	2.8	4.0	3.8	19.6		AVERAGE EFFECT	2.2	1.6	1.3	1.8	

Least significant difference (P)
Interaction = 9.6 (99:1)
Average effect
Digested sludge = 2.2 (19:1)
Salt-simulated sludge = 3.6 (19:1)

Least significant difference
NONE

Least significant difference
NONE

Table 133. Effects of digested sludge and salt-simulated sludge on Ni content in soybean seed. Data are reported in parts per million of oven-dry (60°C) tissue.

SIMULATED SLUDGE	DIGESTED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	3.4	5.6	6.2	7.1	5.6
36	3.4	4.6	8.0	7.4	5.8
72	4.2	7.9	5.0	5.0	5.6
144	4.6	4.7	5.1	6.8	5.3
AVERAGE EFFECT	3.9	5.7	6.1	6.6	

Least significant difference (P)
Interaction = 2.5 (99:1)
Average effect
Digested sludge = 0.8 (99:1)

Table 134. Effects of digested sludge and salt-simulated sludge on Cd contents in soybean tissues at three growth stages. Data are reported in parts per million of oven-dry (60°C) tissue.

FIRST CUTTING - 21 DAYS

SIMULATED SLUDGE t/ha	DIGESTED SLUDGE 0	36	72	144	Average Effect
0	1.8	5.5	9.1	5.4	5.5
36	2.1	3.9	4.4	5.4	4.0
72	4.7	5.1	5.6	7.4	5.7
144	6.1	6.9	6.3	5.6	6.2
AVERAGE EFFECT	3.7	5.4	6.4	6.0	

Least significant difference (P)
Interaction = 3.3 (99:1)
Average effect
Digested sludge = 1.0 (99:1)
Salt-simulated sludge = 1.6 (99:1)

SECOND CUTTING - 32 DAYS

SIMULATED SLUDGE t/ha	DIGESTED SLUDGE 0	36	72	144	Average Effect
0	5.2	1.6	6.9	6.4	5.0
36	8.3	5.1	2.8	4.8	5.2
72	0.6	3.1	3.7	6.3	3.4
144	5.4	5.6	0.1	6.6	4.4
AVERAGE EFFECT	4.9	3.8	3.4	6.0	

Least significant difference (P)
Interaction = 2.4 (99:1)
Average effect
Digested sludge = 1.3 (99:1)

THIRD CUTTING - 95 DAYS

SIMULATED SLUDGE t/ha	DIGESTED SLUDGE 0	36	72	144	Average Effect
0	1.8	3.6	5.1	5.8	4.1
36	6.4	8.1	8.5	10.1	8.3
72	15.6	9.8	10.9	9.1	11.4
144	18.5	12.6	9.5	10.2	12.7
AVERAGE EFFECT	10.6	8.5	8.5	8.8	

Least significant difference (P)
Interaction = 5.4 (99:1)
Average effect
Salt-simulated sludge = 2.7 (99:1)

Table 135. Effects of digested sludge and salt-simulated sludge on Cd content in soybean seed. Data are reported in parts per million of oven-dry (60°C) tissue.

SIMULATED SLUDGE	DIGESTED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	0.4	0.6	1.0	1.0	0.7
36	2.2	1.3	1.5	1.4	1.6
72	2.9	2.1	1.9	1.6	2.1
144	3.3	2.3	1.6	2.4	2.4
AVERAGE EFFECT	2.2	1.6	1.5	1.6	

Least significant difference (P)
Interaction = 0.5 (99:1)
Average effect
Digested sludge = 0.2 (99:1)
Salt-simulated sludge = 0.2 (99:1)

Table 136. Effects of digested sludge and salt-simulated sludge on ash contents of soybean tissues at three growth stages. Data are reported in percent of oven-dry (60°C) tissue.

FIRST CUTTING - 21 DAYS

SIMULATED SLUDGE	DIGESTED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	9.37	10.00	10.81	9.91	10.02
36	10.10	11.49	10.95	11.15	10.92
72	11.48	11.27	9.88	9.27	10.48
144	8.88	10.70	9.93	10.19	9.92
AVERAGE EFFECT	9.96	10.86	10.39	10.13	

Least significant difference
NONE

SECOND CUTTING - 32 DAYS

SIMULATED SLUDGE	DIGESTED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	11.25	10.35	8.39	8.71	9.68
36	10.62	10.46	9.98	10.46	10.36
72	10.62	10.16	9.29	11.28	10.34
144	10.29	8.78	7.98	8.39	8.86
AVERAGE EFFECT	10.69	9.91	8.91	9.71	9.71

Least significant difference (P)
Interaction = 1.18 (99:1)
Average effect
Digested sludge = .37 (99:1)
Salt-simulated sludge = .59 (99:1)

THIRD CUTTING - 95 DAYS

SIMULATED SLUDGE	DIGESTED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	12.46	16.17	17.08	16.74	15.61
36	14.74	15.53	15.15	19.57	16.25
72	14.77	15.58	17.90	16.50	16.19
144	15.23	13.66	13.73	13.50	14.03
AVERAGE EFFECT	14.30	15.23	15.96	16.58	

Least significant difference (P)
Interaction = 3.08 (19:1)
Average effect
Digested sludge = .97 (19:1)
Salt- simulated sludge = 1.54 (19:1)

A summary of the analyses of variance for these data is given in Table 138. Interaction effects occur for all data except Fe and ash weight. Only Fe and, surprisingly, ash weight are not associated with main effects of digested sludge; similarly, neither are Mg and ash weight with salt-simulated sludge. Examination of the data suggests that digested sludge added alone or in combination with readily available sources, even after a schedule of leaching, will create a unique pattern of concentration for the elements determined. Except for the important macroelements Ca and Mg and Na, which were so highly concentrated in the plants before leaching the soil, digested sludge tends to increase levels of elements in aerial plant parts. These relations are seen in another way in the discussion of negative interactions of Ca and Mg later.

Compared with results of Walker (164) for analyses of soybean leaves from plants in the early growth period sampled in fields across Illinois, the absolute amounts of K, P, Mn, and Zn are high by up to fourfold (in the case of Mn). Contents of Fe and Na and Cu, to a lesser extent, are low, whereas N, Ca and Mg compare with the values reported by Walker. Nitrogen, P and Mg are high or above the optimum according to the data of Ohlrogge (115). Nitrogen, K, Fe, and Cu are lower than values cited by Harper (64) and Mn four times higher than levels found in sand culture grown outdoors.

Treatment effects, second cutting or initiation of blooming - Data for yield of dry matter are presented in Table 111 and contents of elements and ash in Tables 112 through 136. The main effect for digested sludge is either significant or highly significant for the tissue contents of all elements except N, Ca, Fe, Cu, and Ni (Table 111). Only Zn, Cu, Ni, and Cd do not reflect the effect of salt-simulated sludge treatment. Except for K, Mg, Na, and Ni, interaction effects are mostly highly significant. Yield at the second cutting, like the first cutting, shows a significant interaction effect in addition to a highly significant main effect of digested sludge.

Relative to the first cutting, N, P, K, Mg, and Zn values are similar. Iron content decreases particularly in the low treatments. Compared with results of Walker (164) for analyses of soybean leaves from plants at full height sampled in fields across Illinois, the absolute amounts of P, Mn and Zn are substantially higher. Potassium, Ca and Mg are up to 50 percent higher. Nitrogen compares closely, whereas, Fe is lower by about one-half and Na values are one-fifth to one-fourth those reported by Walker.

Copper and Mg are within the range of contents given by Ohlrogge (115) whereas, P is well above his optimum and N is higher than the range of data he sites for this stage growth. Nitrogen and K contents are within the range of data given by Hanway and Weber (63) for field experi-

Table 137. Effects of digested sludge and salt-simulated sludge on ash content of soybean seed. Data are reported in percent of oven-dry (60°C) tissue.

SIMULATED SLUDGE	DIGESTED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	5.54	6.03	6.20	6.17	5.98
36	5.87	6.55	6.68	6.10	6.30
72	6.35	6.78	6.14	5.80	6.27
144	6.59	6.20	5.72	5.69	6.05
AVERAGE EFFECT	6.09	6.39	6.18	5.94	

Least significant difference (P)
  Interaction = .40 (99:1)
  Average effect
    Digested sludge = .13 (99:1)
    Salt-simulated sludge =.20 (99:1)

Table 138. Table of F values for salt-simulated sludge and digested sludge effects and their interactions on contents of various elements in the aerial portion of soybean and on dry matter production (yield), bean weight, seed germination, and oil content of the seed. Note that values for soil extract in ash column are for lead which was not detected in the plant tissues. Values equaling or exceeding 2.92 and 4.51 are significant at the 5 and 1 percent levels in the case of main effects. For interactions, values equaling 2.21 and 3.07 are significant at the 5 and 1 percent levels, respectively.

EFFECT	N	P	K	Ca	Mg	Fe	Mn	Zn	Cu	Na	Ni	Cd	Ash
							First Cutting - 21 Days						
Sludge	56.54	23.7	4.42	18.5	3.55	0.33	49.15	29.44	9.46	4.00	3.89	7.98	1.91
Salt	22.65	76.7	32.02	66.7	1.87	11.26	8.09	17.35	6.79	3.62	3.51	5.33	2.56
Interaction	2.26	27.2	6.91	7.53	2.51	1.63	5.21	4.03	2.29	5.57	3.66	3.32	1.85
							Second Cutting - 32 Days						
Sludge	2.48	24.30	6.01	0.29	6.61	2.36	40.33	17.75	1.89	4.11	0.97	4.85	23.14
Salt	15.60	19.07	11.61	7.98	8.38	5.27	5.02	0.36	2.16	4.66	1.19	2.40	21.46
Interaction	10.84	23.72	1.86	5.52	1.26	2.72	21.11	17.01	4.30	0.94	1.12	5.63	5.31
							Third Cutting - 95 Days						
Sludge	10.63	9.27	7.37	0.52	5.28	2.08	28.13	17.40	0.32	0.21	1.43	2.05	3.39
Salt	5.54	26.43	4.43	1.25	0.33	15.67	5.11	3.73	12.95	1.63	2.87	29.82	3.77
Interaction	2.66	19.09	4.57	1.97	4.60	1.87	5.70	4.17	9.54	4.03	1.25	4.59	2.40
							Seed						
Sludge	2.47	17.64	5.20	12.68	2.01	10.47	33.83	31.48	7.50	2.20	12.60	25.17	13.10
Salt	3.29	23.50	11.18	2.26	8.72	7.31	4.29	105.00	16.95	4.81	0.47	132.08	9.03
Interaction	5.44	24.13	10.20	1.62	2.44	7.64	7.67	28.35	7.57	1.79	4.42	13.29	14.62
							Soil Extract						
Sludge		218.95	322.63	265.80	139.09	160.00	63.27	723.00	208.11	32.80	6.46	64.39	86.43
Salt		332.11	1083.01	584.80	182.72	124.00	19.17	1511.00	243.60	8.53	23.04	494.63	143.53
Interaction		15.69	16.97	8.16	14.06	2.57	3.99	0.92	0.26	4.57	1.15	3.55	49.06

	Yield: 21 Days	Yield: 32 Days	Yield: 95 Days	Bean Wt.	Germination	% Oil
Salt	0.11	6.34	18.16	3.72	2.47	4.40
Sludge	2.72	0.97	10.21	3.32	14.61	1.66
Interaction	2.75	2.53	6.63	8.67	2.40	4.52

ments, but P is about two times higher than their values. As in the first cutting, K, Fe and Cu are less than values cited by Harper (64) for soybeans grown outdoors in sand culture. Manganese values are three to four times those reported by Harper.

Treatment effects, third cutting or mature plant at harvest - Analyses of the third cutting are for the aerial plant parts without seeds and pods. Leaves were collected as they dropped. Data for elemental concentrations and ash content are presented in Tables 112 through 136. With the exceptions of Ca, Fe, Cu, Na, Ni, and Cd, digested-sludge treatments show main effects for the elements analyzed. The contents of more elements are affected by salt-simulated sludge treatment with only Ca, Mg, Na, and Ni lacking in significant treatment effects. Interaction effects occur for all elements except Ca, Fe and Ni. The production of dry matter at maturity is highly significant for both main effects and interaction, the combination of sludges reducing yield somewhat (Figure 78) at the highest treatment levels.

Nitrogen and Mg are higher than values reported by Ohlrogge (115) as is P, which is in the range that Webb et al. (167) cited for Mg deficient plants. Manganese values, except for the two lower treatments of salt-simulated without digested sludge, are much higher than any data cited by Ohlrogge. Compared with data of Hanway and Weber (63) for whole plant analyses at harvest, N tends to be lower, P three-times higher and K is lower by one-third. Nitrogen is one-half and K is one-quarter the levels cited by Harper (64), whereas, Ca and Mg are three-times higher, P two-times and Mn and Zn four-to ten-times higher. Iron is slightly higher.

Treatment effects, seed - Among the data obtained, those for seed analyses (Tables 113 through 137 and summary in Table 138) are of most immediate interest with regard to application of sludge on cropland because seed is the product utilized in commerce. Therefore, it is particularly noteworthy that more main and interaction effects are significant for seed than in any of the three harvests of plant tissue. Only main effects of Mg and Na for digested and Ni for salt-simulated sludge and Ca and Na for interaction are not significant. As in yield of stems and leaves at maturity, the weight of beans is significant for both main effects and interaction and the interaction is similar to that in the third cutting wherein the combination substantially reduces yield (Table 139 and Figure 79). Oil content (Table 140) is reflected in significant treatment effects for interaction and digested sludge, sludge tending to diminish oil content probably partly because of dilution due to protein increases.

Treatment effects, summary - Application of salt-simulated or digested sludge results in significant differences in content of all elements analyzed for in this experiment. Only Ca, Na and, particularly, Ni content of the aerial plant parts show decreasing tendencies for sig-

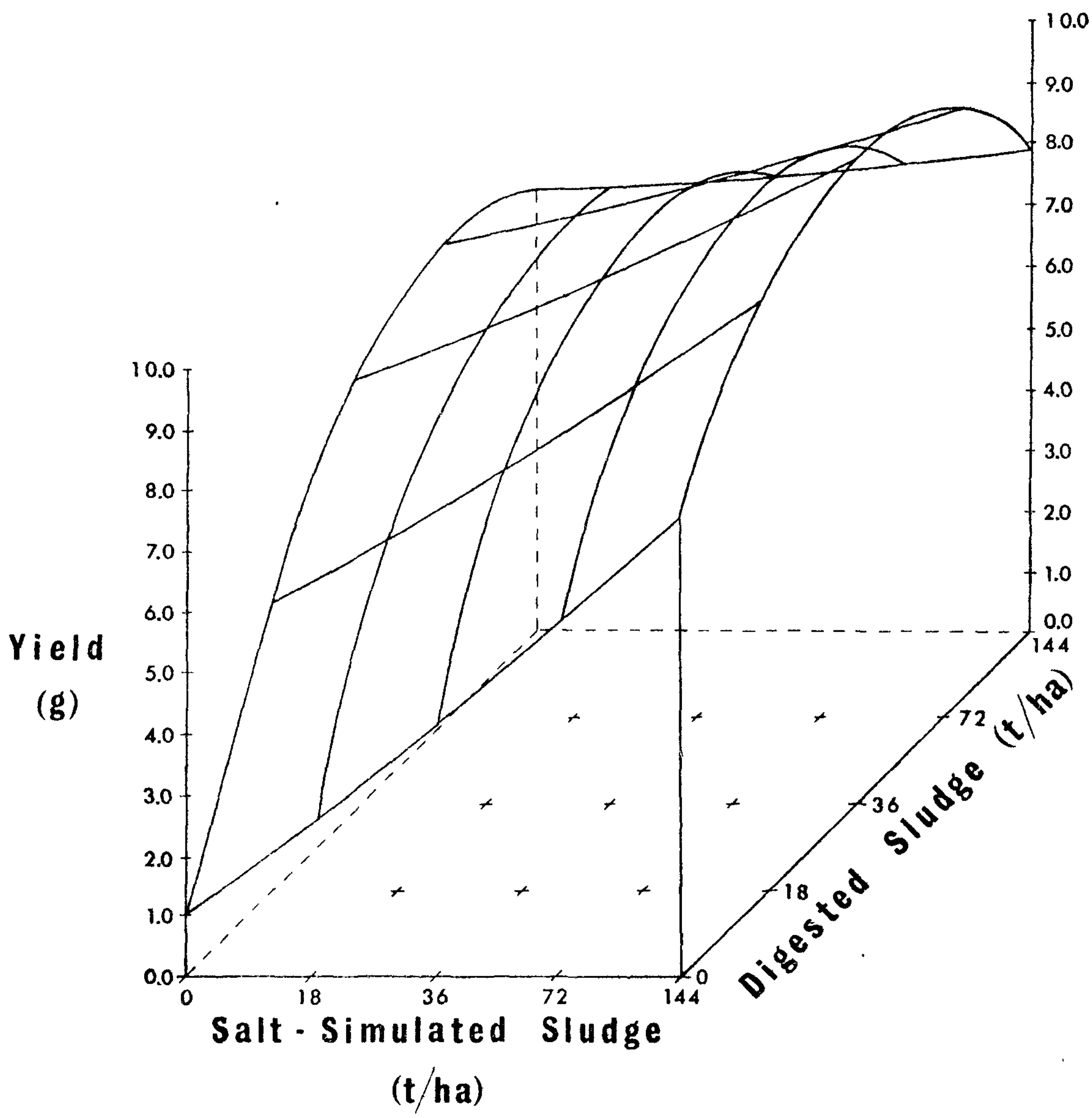


Figure 78. Weight of aerial plant portion of soybean at maturity without seeds and pods. Weights are for one plant. The surface is generated by the equation $Y = 1.03 + 0.75 X, + 2.19 X_2 + .009 X_1^2 - 0.18 X_2^2 - .092 X_1X_2$, where X_1 is simulated salt and X_2 is digested sludge. The standard partial regression coefficients are: X_1, 0.63; X_2, 1.84; X_1^2, .064; X_2^2,-1.29; X_1X_2, -0.45. The multiple correlation coefficient is 0.68.

Table 139. Effects of digested sludge and salt-simulated sludge on seed weight. Data are reported in grams of oven-dry (60°C) seed from 3 plants.

SIMULATED SLUDGE	DIGESTED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	5.41	10.55	12.14	11.63	9.93
36	9.72	10.10	12.57	12.30	11.17
72	10.20	8.95	13.31	14.83	11.82
144	12.78	12.60	8.59	6.56	10.13
AVERAGE EFFECT	9.53	10.55	11.65	11.33	

Least significant difference (P)
Interaction = 5.57 (99:1)
Average effect
Digested sludge = 1.76 (99:1)

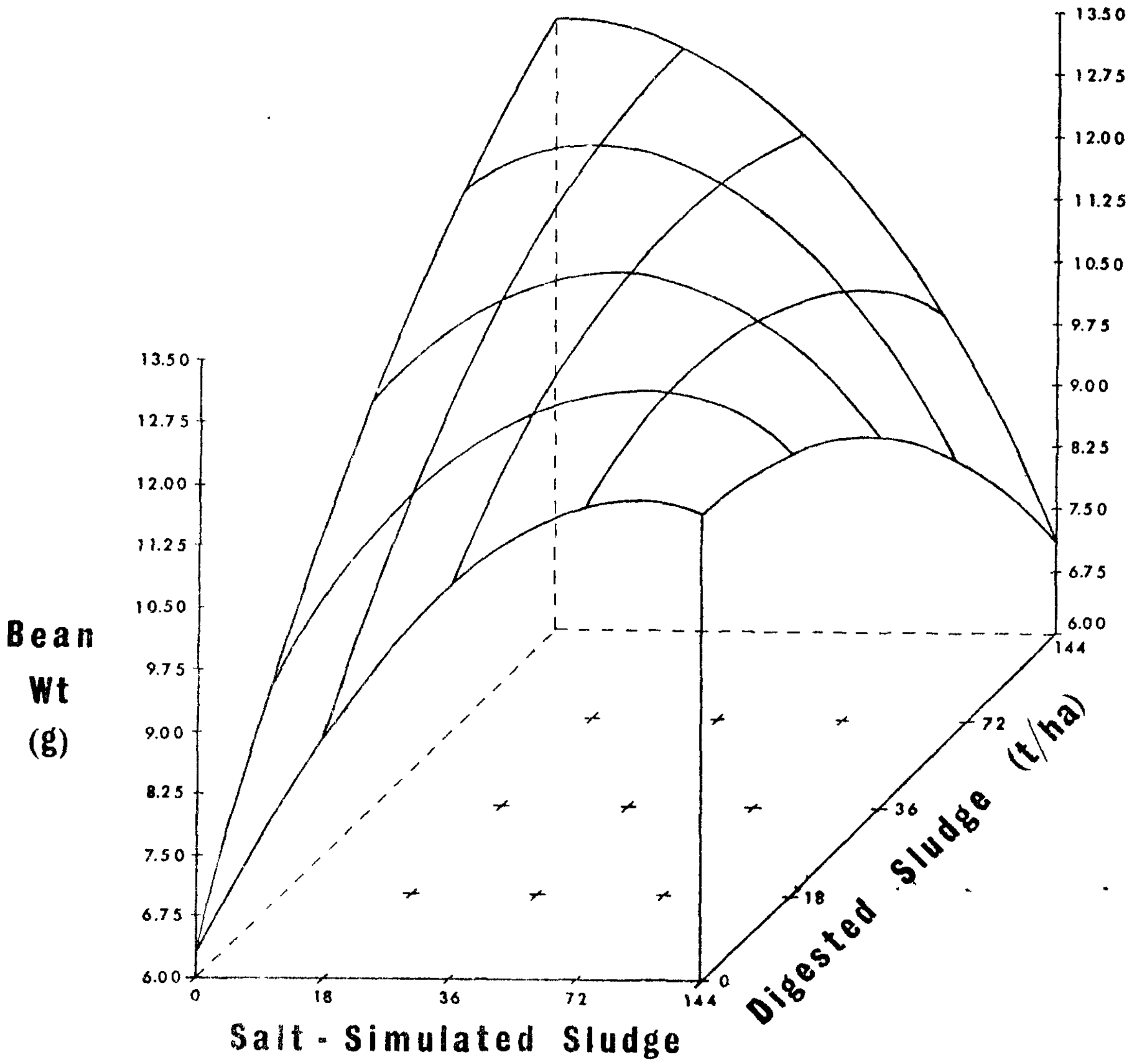


Figure 79. Weight of seeds produced by digested- and simulated-sludge applications to Blount silt loam. Weights are for seeds collected from three plants. The surface is generated by the equation $Y = 6.35 + 1.54\ X_1 + 1.42\ X_2 - 0.11\ X_1^2 - .068\ X_2^2 - 0.18\ X_1X_2$, where X_1 is simulated sludge and X_2 is digested sludge. The standard partial regression coefficients are: X_1, 1.63; X_2, 1.51; X_1^2, -.99; X_2^2, -.63; X_1X_2, -1.11. The multiple correlation coefficient is 0.70.

Table 140. Effects of digested sludge and salt-simulated sludge on oil content of soybean seed. Data are reported in percent of moisture-free seed.

SIMULATED SLUDGE	DIGESTED SLUDGE				
t/ha	0	36	72	144	Average Effect
0	21.0	21.4	21.5	21.2	21.3
36	21.2	22.5	21.9	21.1	21.7
72	22.2	20.0	22.2	20.7	21.3
144	22.9	21.6	19.7	19.2	20.9
AVERAGE EFFECT	21.8	21.4	21.3	20.6	

Least significant difference
Interaction = 2.0 (99:1)
Average effect
Digested sludge = 0.6 (99:1)

nificant treatment effects as the soybean plant develops, however, these elements do show treatment responses for elemental contents of the seed.

Correlations of elemental contents among cuttings and with soil - Relationships among elements in the tissue of the first cutting are evident in Figure 80. The large number of significant relationships, most of which are negative, with Na is noteworthy. Also, the majority of interelemental relationships with Ca and Mg are negative. The highest correlation coefficient, .89, occurs between Ca and Na. Considerably fewer significant relationships occur in the second cutting (Figure 81) and among these the sense of the relationship has changed to negative for Cd and Ca contents. At harvest the number of significant relationships (Figure 82) increased substantially over that of the second cutting. More negative relationships occur and Cu-Cd and Cu-Zn correlations have changed to negative relationships.

Fewer significant correlations (Figure 83) occur in the content of elements in seed than among elemental contents of the aerial plant parts at harvest. Nitrogen and Ca are noteworthy in that all correlations associated with these elements are negative. The four correlations associated with Ni contrast with only one correlation that exists among all of the three cuttings of aerial plant parts.

Reference to Tables 141 and 142 offers an opportunity to follow the correlation of elemental content through the three cuttings, seed and with soil extract. Among the macroelements, P, K, and Mn, and Fe, to a lesser extent, exhibit significant correlations between the first harvest, subsequent cuttings, seed, and soil extract. Among the microelements, only Zn shows a similar tendency for first cutting levels to correlate with later cuttings, seed and soil. In the seed, levels of N, P, Mg, Fe, and Mn are related to contents in aerial plant parts at maturity, and among the microelements Zn, Cu and Cd in seed are related to mature plant levels. Respective seed levels correlate positively with Ca, Mg, Mn, Zn, Cu, and Cd contents extracted from soil with 0.1 N HCl.

Summary - Liquid digested sludge added to soil at rates of up to 144 t/ha will increase yields of mature plant tops and seed substantially. However, in the presence of comparable amounts of available macro-and microelements, slight depression of yields of plant tops and particularly seed are apparent at application rates of 288 t/ha of combined applications of digested and salt-simulated sludge. Digested sludge added to soil will influence the elemental composition of soybean grown in its presence, decreasing some and increasing others, sometimes as much as fourfold when compared with field-grown soybeans. Among the analyses made in this study, the levels of all elements in the seed are within limits considered desirable by animal nutritionists. Perhaps in regard to metal composition, Cd is to be considered the first limiting factor in that its content in seed is closely related to sludge

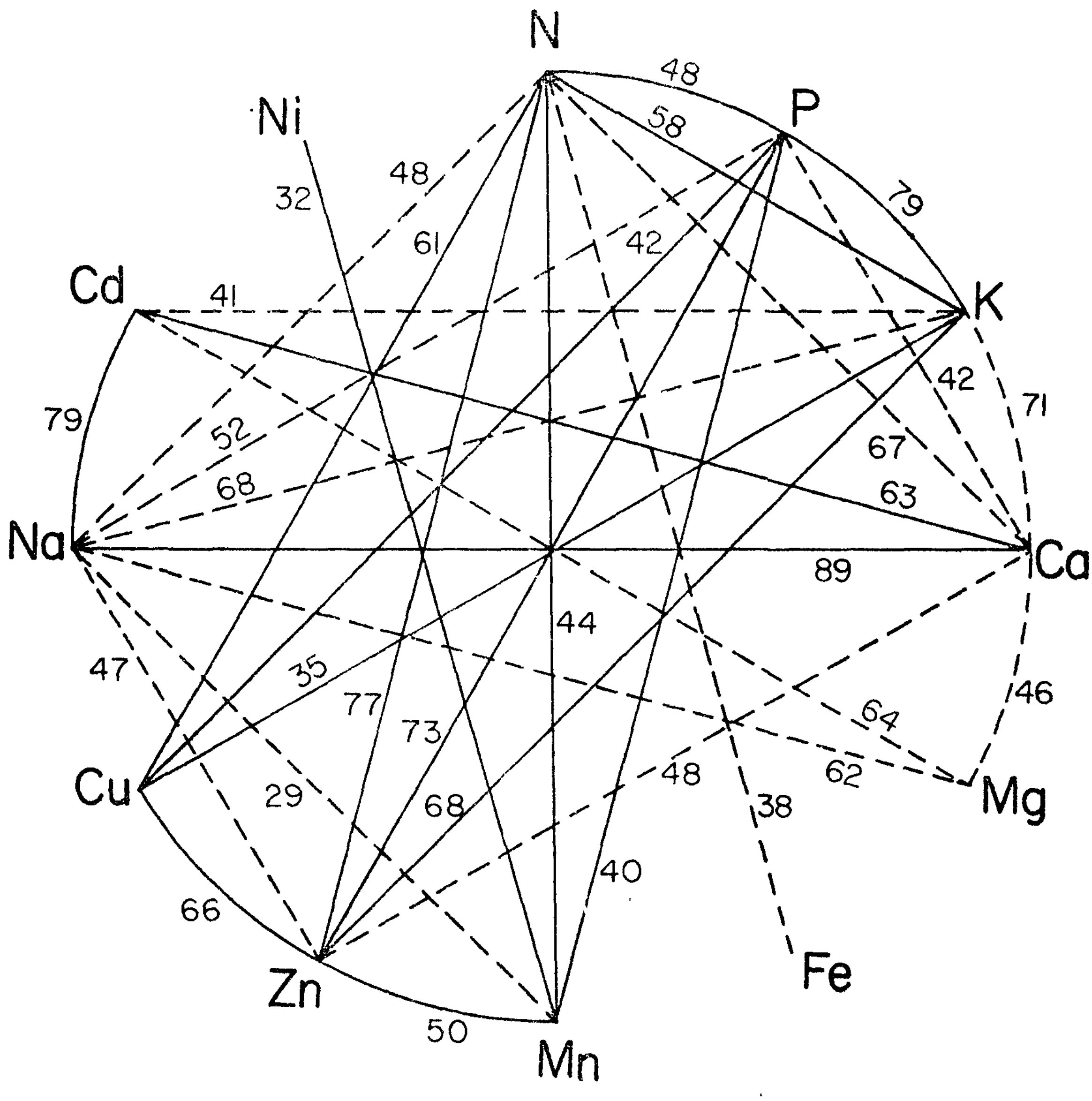


Figure 80. Graphical representation of simple linear correlations among macroelements and microelements in soybean aerial plant parts harvested at 21 days, the beginning of the rapid growth phase. Numbers are correlation coefficients multiplied by 100. Coefficients greater than 28 are significant at the .01 level. Dashed lines are negative correlations.

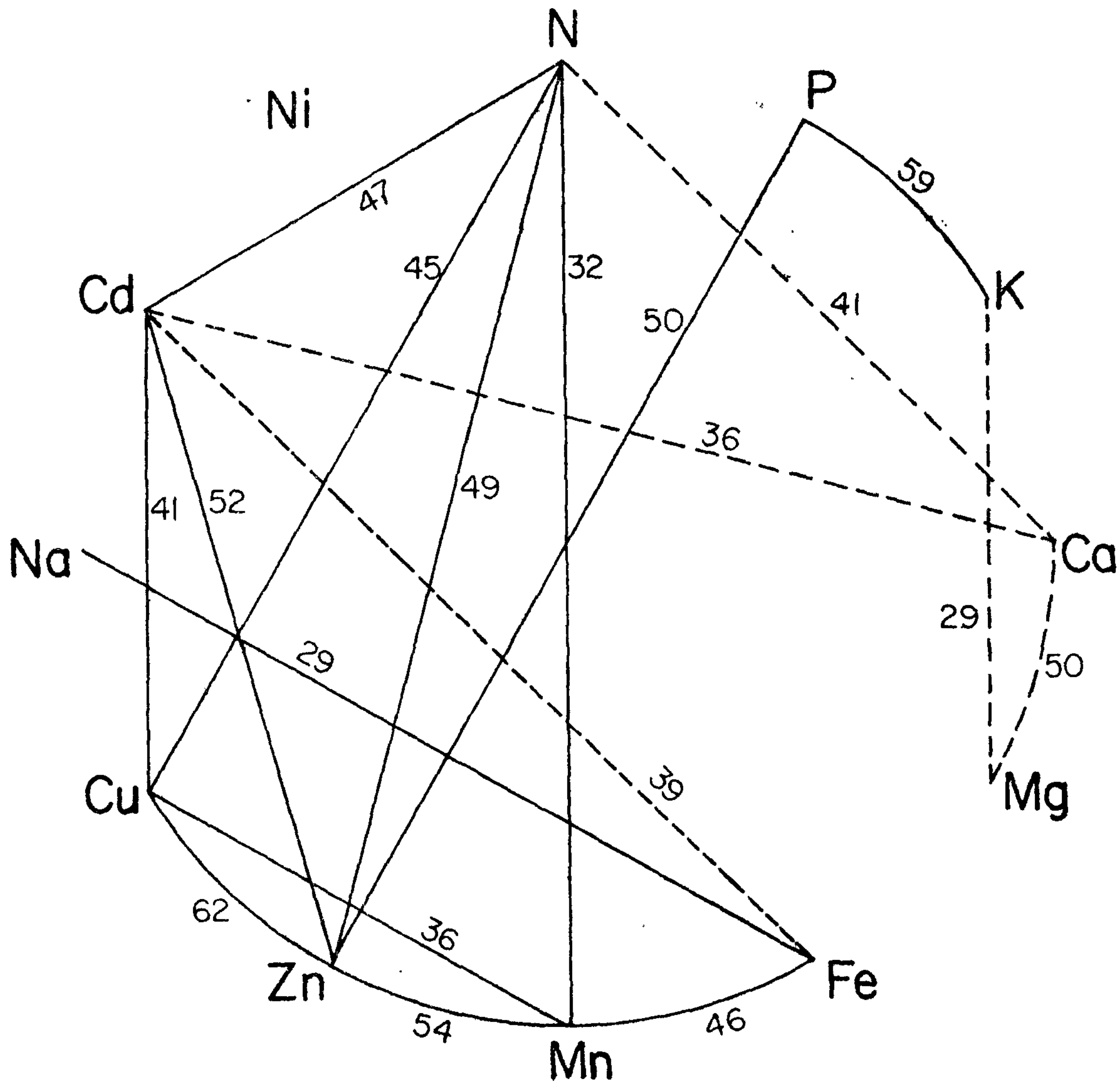


Figure 81. Graphical representation of simple linear correlations among macroelements and microelements in soybean aerial plant parts harvested at 32 days, the initiation of blooming. Numbers are correlation coefficients multiplied by 100. Coefficients greater than 28 are significant at .05 level and coefficients greater than 37 are significant at the .01 level. Dashed lines are negative correlations.

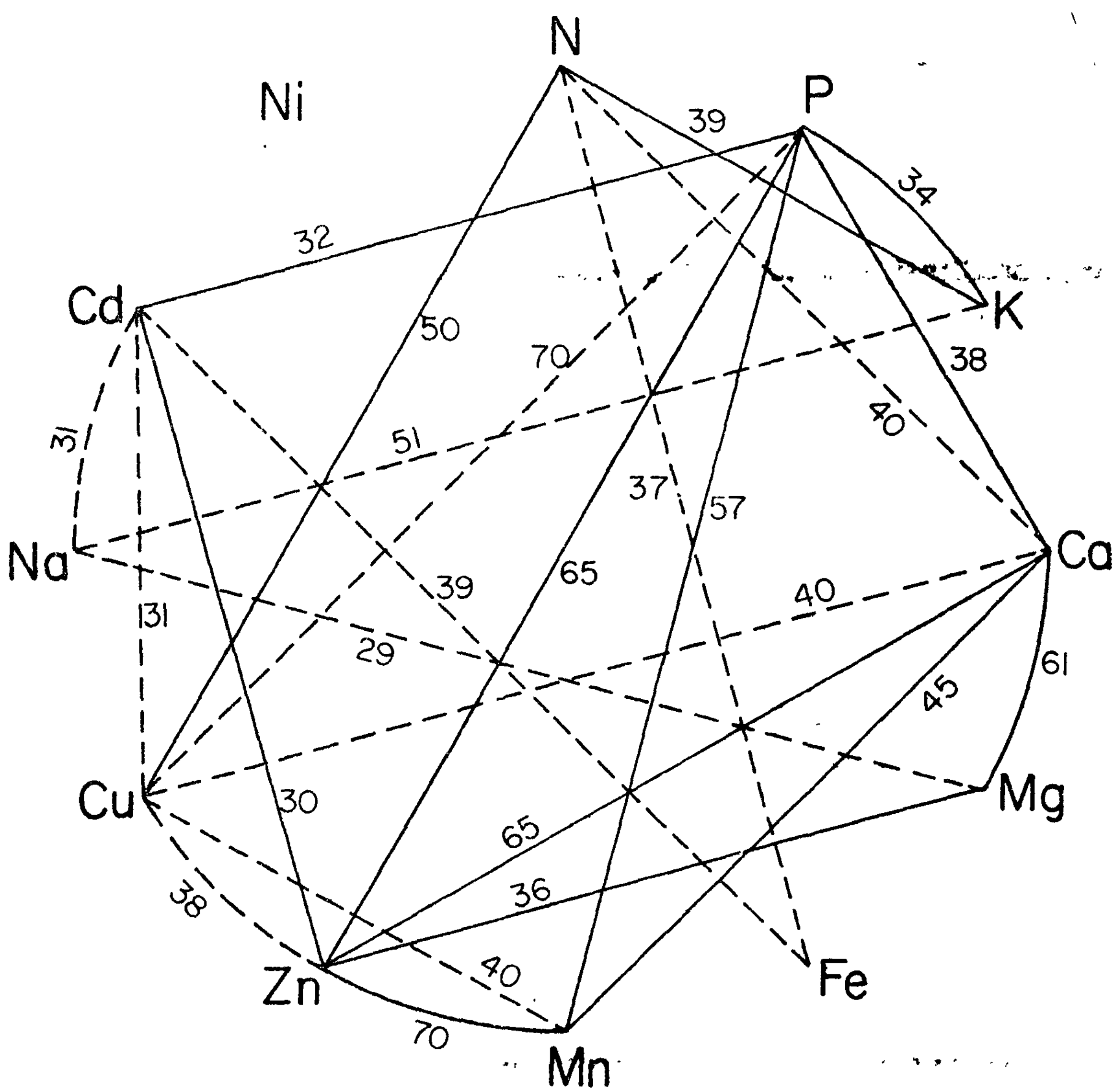


Figure 82. Graphical representation of simple linear correlations among macroelements and microelements in soybean aerial plant parts harvested at 95 days, the date of harvest. Numbers are correlation coefficients multiplied by 100. Coefficients greater than 28 are significant at .05 level and coefficients greater than 37 are significant at the .01 level. Dashed lines are negative correlations.

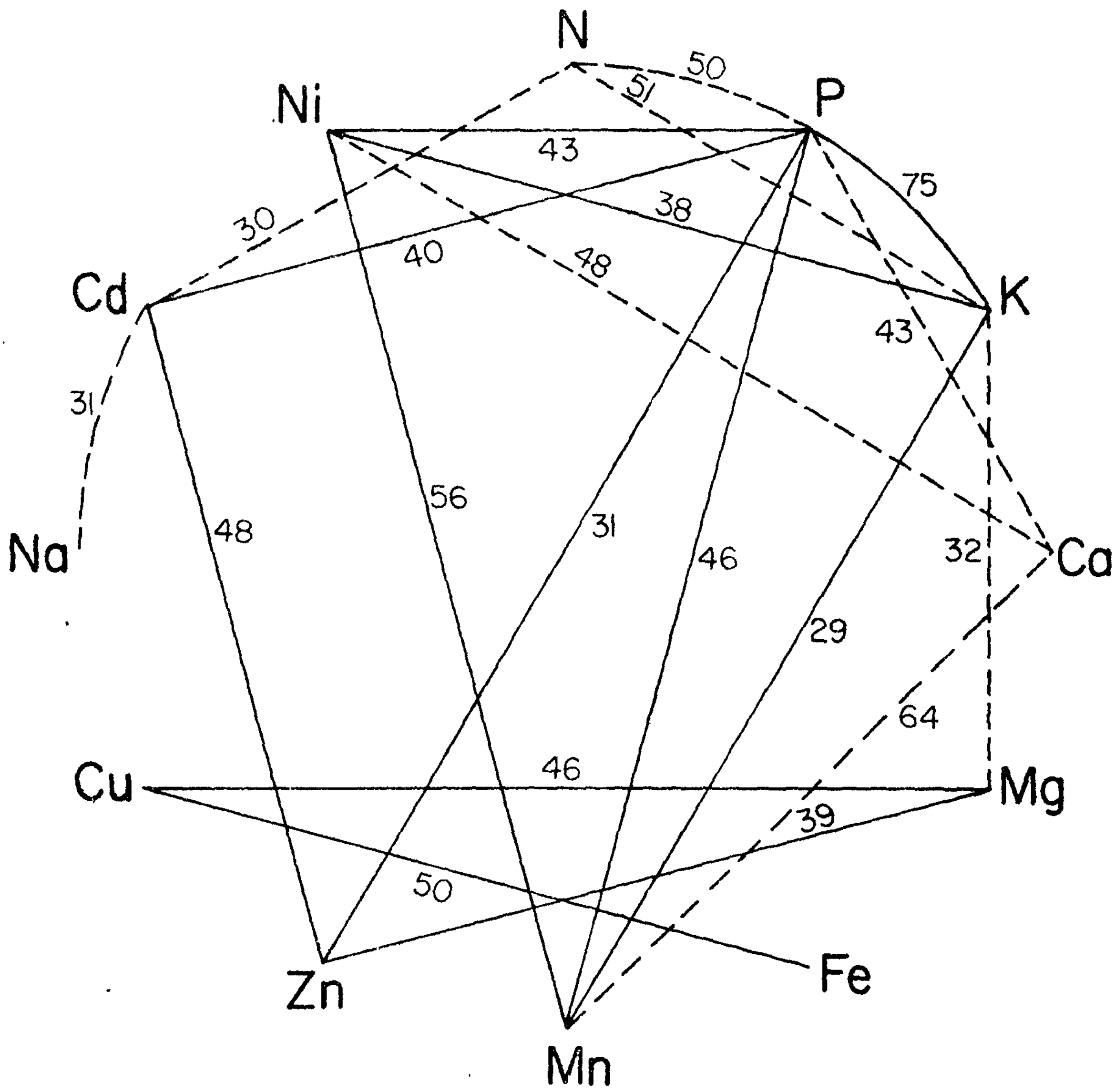


Figure 83. Graphical representation of simple linear correlations among macroelements and microelements in soybean seeds. Numbers are correlation coefficients multiplied by 100. Coefficients greater than 28 are significant at .05 level and coefficients greater than 37 are significant at the .01 level. Dashed lines are negative correlations.

Table 141. Simple linear correlation matrices for several elements among analyses of tissues from three successive cuttings of soybean, soybean seed, and 0.1 N HCl extract of soil in which plants were grown. The correlations are based on 48 observations. Asterisks have their usual meaning.

	CUTTINGS			
	1st	2nd	3rd	Seed
NITROGEN				
2nd Cutting	.038			
3rd Cutting	.547**	.164		
Seed	.069	.120	.419**	
Soil				
PHOSPHORUS				
2nd Cutting	-.299*			
3rd Cutting	.665**	-.009		
Seed	.826**	-.112	.802**	
Soil	.300*	-.220	-.118	.021
POTASSIUM				
2nd Cutting	-.265			
3rd Cutting	.376**	-.069		
Seed	.458**	.045	.199	
Soil	.616**	-.476**	.440**	-.142
CALCIUM				
2nd Cutting	-.140			
3rd Cutting	.169	.023		
Seed	.250	.050	.065	
Soil	-.755**	.421**	-.082	-.350*

	CUTTINGS			
	1st	2nd	3rd	Seed
MAGNESIUM				
2nd Cutting	.027			
3rd Cutting	.161	-.043		
Seed	-.042	.029	.411**	
Soil	-.101	.181	.368*	.617**
IRON				
2nd Cutting	.127			
3rd Cutting	.524**	.019		
Seed	-.155	.014	-.296*	
Soil	-.443**	-.191	-.504**	.067
MANGANESE				
2nd Cutting	-.322*			
3rd Cutting	.818**	-.212		
Seed	.790**	-.211	.895**	
Soil	.326*	-.205	.428**	.470**

Table 142. Simple linear correlation matrices for several elements among analyses of tissues from three successive cuttings of soybean, soybean seed, and 0.1 N HCl extract of soil in which plants were grown. The correlations are based on 48 observations. Asterisks have their usual meaning.

	Cuttings 1st	Cuttings 2nd	Cuttings 3rd	Seed		Cuttings 1st	Cuttings 2nd	Cuttings 3rd	Seed
ZINC					NICKEL				
2nd Cutting	-.036				2nd Cutting	-.071			
3rd Cutting	.678**	-.044			3rd Cutting	-.083	-.063		
Seed	.629**	-.323*	.368*		Seed	.347*	.212	-.048	
Soil	.634**	.013	.296*	.705**	Soil	-.097	.072	.148	.281
COPPER					CADMIUM				
2nd Cutting	.082				2nd Cutting	-.052			
3rd Cutting	-.116	-.228			3rd Cutting	.292*	-.149		
Seed	.271	-.170	.417**		Seed	.109	.000	.827**	
Soil	.659**	-.030	.161	.613**	Soil	.338*	-.004	.637**	.645**
SODIUM					ASH				
2nd Cutting	.078				2nd Cutting	.000			
3rd Cutting	-.084	.040			3rd Cutting	.128	.078		
Seed	.290*	.020	.066		Seed	.378**	.098	.225	
Soil	-.129	-.267	-.085	-.029	Soil				

loading rate and this element is the source of adverse physiological and pathological effects. The highest levels observed in the seed are below the 5 to 10 ppm of diet that Weber and Reid (168) found reduced bone citric-acid levels in mice. Assuming the concentration effect in processing beans for meal the level of Cd remains below the considered detrimental to livestock especially in view of the fact that soybeans grown in sludge amended soil will be blended or mixed with beans of low, native Cd content originating from usual agricultural production. Of course, the important aspect of availability of Cd in meal to absorption in the animal is not considered here. Additionally, the ameliorative effects of increasing amounts of Zn, creating a similar Zn to Cd ratio, should be of benefit to any ultimate animal use of the bean. Except for K which occurs in small amounts, digested sludge used in the study is a ready source for plant uptake of all major elements and minor elements determined in this study. Also, the lack of toxicity symptoms and the positive results for yield suggest that the ratios of elements taken up by soybean under the conditions of this experiment are in favorable ranges for soybean growth and development although the data suggest that application rates in excess of 144 t/ha, especially over short time intervals, may be associated with yield decline.

SECTION VIII

SUPPLEMENTAL FIELD STUDIES

Plant Responses to Applications of Digested Sludge in Field Studies

Introduction - Yields of both corn and soybeans have been significantly increased during the last four growing seasons by furrow irrigation with digested sludge drawn from the heated anaerobic digestor at the Metropolitan Sanitary District of Chicago's Southwest Plant. Sludge, soils, and plant tissue samples were analyzed to determine nutrient and certain nonnutrient chemical element accumulations in soils and uptake by various grain, forage, and fiber crop plants. Absorption of some chemical elements by grain and forage crop plants has been increased by sludge applications but concentrations in plant tissues are considerably below levels considered to be harmful to animals consuming the crops.

Experimental procedure: corn - The continuous corn experiment was conducted on 6 x 12 m plots and was replicated four times. The soil was Blount silt loam which had been cropped to alfalfa in 1965 and 1966.

The check plots were not fertilized in 1968 but 269 kg of N and 302 kg of P (P_2O_5 equivalent) per hectare were applied annually during three following years. All plots received a broadcast application of 224 kg of P per hectare (K_2O equivalent) during the last three years. All inorganic fertilizer was applied in the spring before plowing and ridging the soil. After the ridges and furrows were established, corn was planted on the ridge tops 76 cm apart at a row spacing to give a plant population at harvest of about 44,440 in 1968 and 61,700 plants per hectare in following years. The first application of sludge after planting was usually made when the corn plants had reached a height of about 15 cm. The treatments were 0-, 0.64-, 1.3-, and 2.5-cm applications applied as frequently during the growing season as drying conditions of the sludge would permit. Preemergent herbicides were used to control weeds and the plots were never cultivated during the growing season.

Results of continuous corn study - Average corn yields for three rates of sludge irrigation, as compared with no sludge, are given in Table 143. Also given are the total liquid and dry solids applied each year.

Table 143. Corn yield obtained with sludge treatments and sludge treatment levels.

	Rate of Application				Greatest Application Rate	
Year	0 cm	0.64 cm	1.3 cm	2.5 cm	Total liquid	Total dry solids
	yield ton per hectare				cm/yr	t/ha
1968	4.16	6.03	7.16	7.02	17.14	51.5
1969	8.96	9.34	9.42	9.44	25.4	47.3
1970	5.53	7.48	7.62	8.63	22.86	69.0
1971	6.06	6.50	6.92	7.88	25.4*	96.5
4 yr ave.	6.18	7.34	7.78	8.24	90.80	264.3

* An additional 12.7 cm were applied after the growing season containing 3.38 percent solids or on a dry weight basis, 41.2 tons per hectare.

In Table 143 it may seem that with regard to 1969 yields the plots treated with one-fourth maximum applications were as great as those from plots receiving higher rates. Considering the carry-over of nutrients from sludge application made during the previous year, plus those made during the growing season, the lack of response to higher applications of sludge was probably due to the fact that over 33 cm of well-distributed rainfall occurred during the months of June and July (Table 144). It is somewhat remarkable though, that 25.4 cm of additional water supplied as sludge did not cause a decrease in yields for the highest treatment. One might well expect a decrease during a season of such high amounts of rainfall, because Blount silt loam is poorly drained. It appears that yields were substantially increased as a result of the additional water supplied by the sludge irrigation treatments in 1970, a year in which 17.8 cm of rain fell in June and July. While a favorable response was obtained with sludge applications in 1971, the increase in yields was less than expected in a year during which only 11.2 cm of rain fell in June and July.

In some similar sludge-treated plots on the same field in 1971 the conductivities of saturated extracts of soils were found to be greater than 5 mmhos/cm. Thus, under the extremely dry conditions experienced during the 1971 growing season, high soluble salt contents may have adversely affected corn yields on sludge-treated plots.

Table 144. Monthly rainfall during the growing season, as cm.

Year	April	May	June	July	August	September	Totals
1968	6.35	5.38	13.97	6.43	3.43	9.35	44.91
1969	9.12	13.00	17.93	15.67	1.78	3.22	60.73
1970	10.39	13.18	11.00	6.81	2.54	22.07	65.99
1971	1.52	6.73	4.19	6.98	4.95	5.46	29.84

Although most of the water applied by sludge irrigation is lost by evaporation because the suspended solids seal the surface of the soil, this sealing apparently serves to conserve available water stored in the root zone. On the other hand, when rainfall is insufficient to leach soluble salts to lower depths, the increase in osmotic pressure in soil solutions with higher rates of sludge applications may off-set some of the water conserving advantage. Nevertheless, it is noteworthy that neither during the relatively wet growing season of 1969 nor the dry season of 1971 did yields decrease with increased sludge applications.

Digested sludge on a dry weight basis is a low grade fertilizer, but when frequently applied as a liquid by irrigation methods large amounts of almost all plant nutrients are supplied. The total amounts of plant macronutrients added to the soil as a result of maximum irrigation of corn with digested sludge are given in Table 145. These amounts have been incrementally applied as constituents of the applied digested sludge on maximum-treated plots during four years. The aggregate of 103.4 cm of digested sludge applied during four years is equivalent to a total solids loading of 305.5 dry tons per hectare.

Table 145. Total plant macronutrients in kilograms per hectare applied as a constituent of sludge on corn plots receiving maximum treatments during four years.

Total N	NH_4-N	P	K	Ca	Mg	S
14,426	6,462	8,445	1,277	9,990	2,890	1,131

Digested sludge nearly always contains large quantities of plant essential micronutrients. The amounts applied on the maximum-treated corn plots as a constituent of sludge are presented in Table 146.

Table 146. Total plant essential micronutrients in kilograms per hectare applied as a constituent of sludge on corn plots receiving maximum treatments during four years.

Fe	Zn	Cu	Mn	Mo	B	Cl
14,717	2,072	538	179	0.4	18	2,262

Plants are the source of certain minor elements that are essential to the growth of animals but not for the plant itself. In addition to most of the elements listed in Table 146, animals require small amounts of the elements listed in Table 147. The data in Table 147 indicate that relatively large amounts of essential elements for animals have been added on the maximum-treated plots.

Table 147. Minor elements (kilograms per hectare) applied as a constituent of sludge on crop plots receiving maximum treatments during four years. These elements are considered essential for animals but not for plants.

Na	Cr	Co	Se	Ni
605	1288	1.2	1.6	130

Plants absorb many minor elements which are not presently considered to be essential for either plants or animals. Like most natural products, digested sludge contains nonessential trace elements. The elements listed in Table 148 are those most frequently mentioned as having potential for producing a detrimental effect on animals which have consumed feed containing some concentration in excess of the critical level. The total amounts of four nonessential minor elements applied during a four year period as a constituent of sludge on maximum-treated corn plots are given in Table 148.

Table 148. Additional total trace elements (in kilograms per hectare) applied as a constituent of sludge on corn plots receiving maximum treatments during four years. These elements are not considered essential for either plants or animals.

Pb	Hg	Cd	Sn
459	0.16	146	18.4

Some changes in soil chemical parameters with digested sludge applications are of interest. First, in the absence of a continuous liming program, frequent sludge applications will result in a lowering of the pH in the soil surface as evident from the data presented in Table 149. After the application of 42.5 cm during a two-year period the soil was reduced from a pH value of 5.6 to 4.9. The depression of soil pH values probably was caused by the large amounts of nitrogen applied as a constituent of digested sludge. The pH values were allowed to reach much lower values than would be permitted under a normal soil management program, because we wanted to see how soil pH would effect the absorption of trace elements by corn plants, which is discussed later on. In the fall of 1970, limestone was applied on the plots at rates calculated to raise the soil pH to a value of at least 6. As much as 11.2 tons per hectare of limestone were applied on the maximum sludge treated plots. When the soils were sampled in the latter part of April, a few weeks before planting the 1971 crop, only slight increases in soil pH values were noted as a result of lime applications. Another item to be noted from Table 149 is the marked increase in plant available phosphorus (P_1) with increasingly greater sludge applications. After two years of digested sludge application the concentration levels of phosphorus (P_2) which are slowly available to plants from precipitants in soils were greater than could be read from the standard laboratory calibration curve, even at the lowest sludge application rate. The apparent decrease in plant available potassium from 1969 to 1971 may be normal variation related to soil conditions at time of sampling, sample handling, etc., but the trend is of interest. Available K appears to have been reduced in all plots, even though 224 kg per hectare of (K_2O equivalent) K fertilizer were applied annually over all plots. Soil contents of plant available K do not appear to be influenced by sludge loading rates.

After the first three years of applying digested sludge at the annual rates given in Table 143, it was possible to detect significant changes in the total concentrations of some elements in the surface of the soil. As shown in Table 150, the concentration increases in soils of total P, Cu, Cd, and Hg with increased loading rates were significant at the

Table 149. Averages of soil test results from samples collected in April 1969, 1970 and 1971 from corn plots after the 1st, 2nd and 3rd years of digested sludge application.

	Kilograms per hectare											
	pH			P_1			P_2			K		
cm/ appl.	1969	1970	1971	1969	1970	1971	1969	1970	1971	1969	1970	1971
0	5.2	5.6	5.4	62	38	49	83	49	60	396	420	262
1/4	5.7	5.4	5.8	65	151	110	102	140+	140+	358	311	246
1/2	5.5	5.1	5.4	92	180	214	120	140+	140+	339	319	259
Max	5.3	4.9	5.2	132	253	302	140+	140+	140+	394	315	289

Table 150. Total contents of chemical elements in the (0-to 15.2-cm depth) surface horizon soil samples collected from corn plots in April 1971 after a total of 65.4 cm or 167.8 dry tons per hectare of digested sludge was applied on the maximum-treated plots. Data are reported on oven-dry basis (110°C).

Sludge appl.	P**	K	Ca	Mg	Fe	Mn	Na	Cr*	Cu**	Pb*	Ni	Zn*	Cd**	Hg**
Rate	- - - - - - - - - percent - - - - - - -							- - - - parts per million - - - -						part per billion
0	0.0429	1.80	0.30	0.27	1.83	0.12	0.59	29	19	31	23	72	1.1	44
1/4 Max	0.0522	1.91	0.36	0.30	1.85	0.13	0.60	43	22	39	30	110	3.0	87
1/2 Max	0.0750	1.78	0.31	0.28	1.80	0.14	0.59	61	34	44	25	163	5.1	154
Max	0.1152	1.82	0.31	0.25	1.85	0.15	0.60	86	52	60	28	260	8.5	273

* Treatment effects significant at 5% level
** Treatment effects significant at 1% level

1-percent level. Concentration increases of Cr, Pb and Zn in the soil surface horizon were found to be significant at the 5-percent level. Changes in concentration levels were not noted for all elements for several reasons. Digested sludge may contain concentrations that are about the same as those in soils, the chemical elements have migrated with percolating water to lower depths, or the amount of a particular chemical species in soils is so large that the amounts added as a constituent of sludge cannot yet be detected above the normal variation between soil samples.

Determinations of chemical concentrations in 0.1 N HCl soil extracts provides an estimation of the plant availability or mobility of the several species of elements. As shown in Table 151, digested sludge applications have increased the mobility of P, Na, Cr, Cu, Pb, Ni, Zn, and Cd in the zero to 15.2-cm depth of Blount silt loam. In the 30.5-to 45.7-cm depth increased mobilities of Na, Cr, Cu, Zn and Cd with sludge applications are highly significant, although actual change in concentration are very small for some species. The increased mobilities of Mn and Pb with sludge application are significant at the 5-percent level. An important observation that may be made from the data in Table 151 is that whereas there is little doubt that available phosphorus increases with sludge applications in the plow layer, increases in concentration levels in deeper soil horizons are highly variable.

Data are gathered into Table 152 to show the relative proportion or percent of the several total chemical elements extractable with 0.1 N HCl. Except for K, Ca, Mg, Fe, and Mn, extractability increases for all elements with increased sludge applications.

As can be seen in Table 153, sludge applications were highly correlated with increased contents of N, P and Zn in corn leaf tissue. In the case of Mg, sludge applications were highly correlated with a decreased content of the element in the plant leaves. Concentrations of the elements Ca, Mn, Cd, and B as found in corn leaf tissue correlated with sludge applications at the 5-percent level of significance. Only Zn and Cd concentrations in corn grain correlated at the 1-percent level with sludge application. The content of K in corn grain significantly correlated with sludge application at the 5-percent level.

To summarize, it is evident that a favorable yield response can be expected from relatively large sludge applications in a year of normal weather conditions. However, corn yields were not decreased by sludge applications during a very wet growing season. Trace elements added as constituents of sludge have not presented a problem, even though the Blount silt loam is a poorly drained soil and soil pH was permitted to decrease to a low value with respect to crop production. Since trace elements would be most mobile or available to plants in poorly drained, acid soils, the concentrations of trace element in corn tissue samples

Table 151. Contents of chemical elements extractable by 0.1 N HCl in soil samples collected in April 1971 from corn plots at two depths after a total of 65.4 cm or 167.8 dry tons per hectare of digested sludge was applied on the maximum-treated plots.

Sludge appl. Rate	P**	K	Ca	Mg	Fe	Mn	Na**	Cr**	Cu**	Pb**	Ni**	Zn**	Cd**
	0-to 15.2-cm depth												
	Parts per million												
0	16	16	1200	400	499	304	14	0.94	3.9	6.6	2.3	13	0.22
1/4 Max	51	224	2100	536	536	306	24	3.3	8.4	11.0	3.5	41	1.50
1/2 Max	148	229	1200	413	792	428	27	11.0	19.0	17.0	5.3	98	3.80
Max	376	268	1500	410	775	402	32	19.0	32.0	30.0	7.0	181	7.00
	30.5-to 45.7-cm depth												
0	15	283	1600	1300	741	45	18	0.64	3.5	2.0	2.6	7.8	0.61
1/4 Max	25	258	1500	1200	671	63	27	0.82	4.9	2.7	3.6	12	0.68
1/2 Max	27	272	1600	1200	706	57	38	1.30	5.9	4.2	3.6	16	0.82
Max	49	247	1600	1200	791	61	52	1.60	6.4	5.3	3.6	18	0.88

* Treatment effects significant at 5% level
** Treatment effects significant at 1% level

Table 152. Percent of the total concentration of chemical elements extractable with 0.1 N HCl from surface soil (0-15.2 cm) samples collected in April 1971 from corn plots treated with digested sludge.

Sludge appl.	P	K	Ca	Mg	Fe	Mn	Na	Cr	Cu	Pb	Ni	Zn	Cd
Rate	Percent												
0	3.8	1.3	40.1	15.3	2.7	24.1	0.24	3.3	22.9	trace	9.9	17.6	22.5
1/4 Max	9.9	1.2	51.2	17.3	3.0	24.6	0.45	7.9	40.1	6.9	13.2	37.5	49.3
1/2 Max	19.4	1.3	33.7	15.0	4.4	31.9	0.46	17.1	56.4	21.1	21.5	59.6	75.0
Max	30.7	1.5	49.9	17.6	4.2	28.2	0.56	21.1	60.9	35.3	24.6	68.4	82.8

Table 153. Total contents of chemical elements in corn tissue samples collected in 1970. Data are reported on oven-dry basis (60°C).

Sludge appl Rate	N**	P**	K	Ca*	Mg**	Fe	Mn*	Na	Cr	Cu	Pb	Ni	Zn**	Cd*	B	Hg1/
	Corn leaf at tasseling stage															
	Percent					Parts per million										ppb
0	2.43	0.27	2.12	0.51	0.30	106.8	81.3	96.0	4.1	8.9	7.1	2.8	58.0	3.3	26.3	27.3
1/4 Max	2.71	0.29	2.25	0.49	0.25	84.0	83.3	121.8	3.9	9.0	7.4	1.3	85.0	3.0	31.8	17.5
1/2 Max	3.07	0.29	2.12	0.57	0.27	101.0	92.5	111.0	4.9	10.2	7.3	2.6	137.8	5.3	34.6	29.6
Max	3.14	0.33	2.11	0.55	0.18	111.5	116.3	94.0	4.5	8.7	6.3	4.3	212.0	11.6	43.6	37.9

Sludge appl Rate	N	P	K*	Mg	Ca	Fe	Mn	Na	Cr	Cu	Pb	Ni	Zn**	Cd**	B	Hg
	Corn grain															
	Percent				Parts per million											ppb
0	1.65	0.50	0.63	0.26	29.6	100.0	18.0	146.0	0.28	5.2	0.025	2.28	88.8	0.30	7.1	5.2
1/4 Max	1.60	0.46	0.73	0.23	30.7	85.0	13.5	206.8	0.35	6.3	0.025	3.03	93.0	0.60	6.2	4.3
1/2 Max	1.74	0.49	0.65	0.25	25.1	95.3	11.3	98.8	0.34	5.2	0.035	2.18	127.0	0.79	5.4	4.5
Max	1.82	0.56	0.91	0.26	40.7	105.5	18.3	232.3	0.38	5.6	0.028	3.08	152.3	1.03	6.6	3.6

1/ Mercury values are average results from 2 or 3 treatment replications
* Treatment effects significant at 5% level
** Treatment effects significant at 1% level

are higher than would be expected where internal soil drainage is better and soil pH is maintained at a value of 6 or greater. Thus, corn plants did not accumulate toxic levels of trace elements even under soil conditions which should favor such a development.

Experimental procedure: soybeans - The continuous soybean experiment was established to test, under field conditions, the availability to crop plants of phosphorus in digested sludge. However, because of what we now know about the rapid build-up of available phosphorus with sludge applications, our present interest in continuing this study is to see how the high soil contents of available phosphorus will effect soybean nutrition. Soybeans were chosen for this study in order to eliminate the effects that the nitrogen in the sludge might have on a non-leguminous plant.

Three replications of 12 x 12 m plots were established on Blount silt loam in the fall of 1968 for the following treatments: (1) zero or control, (2) maximum, (3) one-half maximum, (4) one-fourth maximum application rates of sludge, and (5) well water supplied at the same time and rate as the maximum sludge application. The plots were split and superphosphate was applied by broadcasting on one-half of each plot at a rate to provide 269 kilograms per hectare of P_2O_5 equivalent each year. Also, all plots received a broadcast application of potassium chloride to provide 269 kilotrams per hectare of K_2O equivalent. After fertilizer was applied, the Blount silt loam plots were fall plowed. During the following years all inorganic fertilizer was applied before spring plowing. The soybeans were planted on the ridges and furrow-irrigated with sludge and water in the same manner as was used for the corn study described earlier.

Results and discussion - Soybean yields for phosphate, sludge and water treatments are given in Table 154. A significant increase in yields in response to additional phosphorus applications had not been observed in three years. Evidently the soil has sufficient available P to meet the demands of soybean plants, even though the Blount silt loam study site was selected because this soil type generally is somewhat deficient in available P. In all three years yields have been significantly ($P > .01$) increased with increased sludge applications. During the first two years, water alone significantly increased yields, but not in 1971. Except for the first year, 1969, the maximum sludge treatment produced an increased yield over an equivalent amount of applied well water. However, the increased soybean yields for the maximum application of sludge were better than the equivalent water treatment only at the 5 percent level of significance in 1970. While it is probably too soon to speculate, it can be seen in Table 154 that there is a trend toward decreased soybean yield by years for all treatments. The failure to obtain a favorable yield response to the 20.3 cm of irrigation water applied during the very dry season of 1971 (see Table 144) cannot be explained.

Table 154. Soybean yield responses to phosphorus, sludge, and water applications.

Year	P_2O_5 kg/ha	Rate of sludge application 0 cm	0.64 cm	1.3 cm	2.5 cm	Water[a/]	Total sludge applied
		- - - - - Metric ton per hectare - - - -					cm
1969	0	2.28	3.02	3.24	3.36	2.92	20.3
	269	2.53	3.00	3.16	3.50	3.48	20.3
1970	0	1.93	2.76	2.98	2.84	2.57	22.9
	269	1.89	2.57	2.84	3.19	2.59	22.9
1971	0	1.77	1.93	2.10	2.13	1.50	33.0[b/]
	269	1.53	1.87	2.08	2.12	1.74	33.0[b/]

a/ Water was applied at the same rate and time as the maximum sludge application.

b/ 20.32 of the 33.02 cm of sludge were applied during the growing season.

Several chemical elements (Table 155) were significantly increased in the surface horizon of Blount silt loam by applications of digested sludge. However, no significant increases in soil contents of total K, Mg, Mn, Na, or Ni were detected. Failure to observe a significant increase in total amounts of these elements is probably due to the fact that sludge applications have not supplied amounts of the elements in sufficient quantities to exceed amounts removed in grain and leaching to lower soil depths. Also, the amounts of these elements added are small compared to levels that occur naturally. Applications of inorganic P affected only the accumulation of total Fe in the surface of the soil. Increases in surface-soil Fe contents with increased sludge applications were significantly greater ($P>0.05$) in the absence of the additional inorganic P. Perhaps the inorganic P increased the mobility of the iron, resulting in a greater movement of the element to lower soil depths, although no differences in extractable Fe content were observed in the 30.5-to 45.7-cm depth (Table 156). Applications of inorganic P fertilizer did not significantly affect the total P content in the soil surface, which is to be expected because the levels added correspond to about ten percent of the naturally occurring amount.

Concentration levels of chemical elements extractable with 0.1 N HCl from samples of soil collected from soybean plots are presented in Table 156. As may be seen in Table 156, the extractability of all

Table 155. Total contents of chemical elements in the (0-15.2 centimeter depth) surface horizon soil samples collected from soybean plots in April 1971 after a total of 43.18 cm or 121.9 dry tons per hectare of digested sludge was applied on the maximum treated plots. Data are reported on oven-dry (110°) basis.

Sludge or water appl. Rate	P**	K	Ca**	Mg	Fe**	Mn	Na	Cr**	Cu**	Pb**	Ni	Zn**	Cd**
	Additional phosphorus fertilizer												
	Percent							Parts per million					
0	0.054	1.91	0.40	0.40	1.89	0.13	0.81	25.7	13.3	5.0	30.3	50.0	1.53
1/4 Max	0.099	1.94	0.44	0.41	2.06	0.13	0.80	61.0	37.4	26.0	33.5	146.8	3.93
1/2 Max	0.144	1.91	0.47	0.43	1.91	0.15	0.81	96.5	60.1	41.6	40.1	234.7	8.07
Max	0.199	1.85	0.52	0.45	2.27	0.17	0.77	137.0	87.0	61.1	39.2	336.9	12.45
H_2O	0.054	1.95	0.40	0.43	2.16	0.14	0.81	27.9	12.4	7.4	31.7	45.7	1.62
Sludge or water appl. Rate	No additional phosphorus fertilizer												
0	0.054	1.90	0.41	0.40	2.03	0.16	0.82	31.3	14.6	7.0	33.5	55.4	1.67
1/4 Max	0.078	1.96	0.43	0.44	2.11	0.13	0.80	56.1	27.9	16.2	35.3	116.9	3.28
1/2 Max	0.142	1.92	0.46	0.44	2.18	0.13	0.78	100.7	64.8	42.8	39.7	243.5	7.68
Max	0.186	1.92	0.52	0.49	2.27	0.13	0.75	135.7	89.8	59.2	43.0	335.6	12.13
H_2O	0.060	1.99	0.39	0.53	2.48	0.11	0.76	30.2	13.2	6.6	34.0	50.8	1.53

* Treatment effects significant at 5% level

** Treatment effects significant at 1% level

Table 156. Contents of chemical elements extractable with 0.1 N HCl in soil samples collected in April 1971 from soybean plots at two depths after a total of 43.18 centimeters or 121.86 dry tons per hectare of digested sludge was applied on the maximum treated plots.

Sludge or water appl. Rate	P**	K**	Ca**	Mg**	Fe	Mn*	Na**	Cr**	Cu**	Pb**	Ni**	Zn**	Cd**
	Additional phosphorus fertilizer								0 - 15.2 centimeter				
	Parts per million												
0	45.4	239.3	1524	502.7	148.7	132.7	13.0	0.36	4.66	5.16	3.97	13.0	0.29
1/4 Max	255.4	259.7	1746	585.7	691.0	258.0	24.0	10.92	26.31	18.36	6.82	49.3	3.75
1/2 Max	571.9	292.7	2089	670.3	558.7	219.3	24.3	24.23	49.09	34.61	9.15	142.4	8.15
Max	945.1	334.7	2785	863.0	966.7	303.7	30.7	44.64	72.75	49.63	11.72	294.9	13.49
H_2O	95.2	220.0	1512	583.0	622.3	195.0	124.7	0.71	5.56	4.39	4.23	13.2	0.15
	No additional phosphorus fertilizer												
0	42.5	266.7	1652	651.7	188.3	102.7	16.7	0.81	5.12	5.42	5.49	13.2	0.23
1/4 Max	199.1	259.7	1706	566.0	284.7	153.7	19.7	7.53	20.68	14.35	7.43	32.2	2.56
1/2 Max	541.0	343.0	2088	748.0	977.0	305.0	25.7	29.48	50.68	37.33	10.20	153.6	8.50
Max	846.7	368.0	2842	914.7	1046.3	291.3	35.0	44.53	75.25	54.12	13.84	301.1	13.61
H_2O	58.5	232.0	1588	651.7	691.0	220.0	113.3	0.56	4.01	3.94	2.67	13.1	0.22

* Treatment effects significant at 5% level

** Treatment effects significant at 1% level

Table 156. (Cont'd) Contents of chemical elements extractable with 0.1 N HCl in soil samples collected in April 1971 from soybean plots at two depths after a total of 43.18 centimeters of 121.86 dry tons per hectare of digested sludge was applied on the maximum treated plots.

Sludge or water appl. Rate	P	K**	Ca	Mg**	Fe*	Mn**	Na**	Cr*	Cu	Pb	Ni	Zn	Cd
	Additional phosphorus fertilizer								30.5 to 45.7 centimeter depth				
	- - - - - - - - - -						Parts per million	- - - - - - - - - -					
0	12.6	241.7	1010	566	1435	133.7	24.7	1.30	3.37	4.60	4.46	7.4	0.24
1/4 Max	15.0	331.3	1376	917	1939	123.7	37.0	1.79	4.47	6.76	5.30	10.9	0.20
1/2 Max	16.4	354.0	1402	936	1612	96.0	39.3	1.69	4.63	5.03	3.86	10.9	0.27
Max	18.7	336.7	1242	1310	1806	135.0	57.3	2.00	4.79	6.33	5.56	13.2	0.23
H_2O	10.8	387.0	1640	1253	1750	99.0	66.7	1.69	3.79	6.59	5.60	10.4	0.39
	No additional phosphorus fertilizer												
0	14.6	213.3	1285	488	1892	304.0	24.7	1.30	5.14	4.69	9.44	13.1	0.19
1/4 Max	15.4	307.3	1509	869	2014	204.0	35.7	1.82	4.53	4.95	6.24	13.0	0.20
1/2 Max	16.0	300.3	1399	961	1733	146.0	41.3	1.95	5.15	5.21	6.35	13.9	0.51
Max	17.4	361.0	1743	1379	1776	145.7	59.0	2.29	5.00	6.67	6.45	14.5	0.34
H_2O	17.1	367.0	3359	2299	1584	180.3	61.7	2.05	3.74	6.24	8.36	11.4	0.35

*Treatment effects significant at 5% level

**Treatment effects significant at 1% level elements except Fe was significantly increased in the soil surface horizon (0 to 15.2 cm) by digested sludge applications. But the application of inorganic P did not affect the extractability of any of the elements in the surface horizon. It did, however, have a significant affect on the extractability of several elements from subsoil samples (30.5 to 45.7 cm). Applications of inorganic P fertilizer significantly decreased the levels of extractable Mn, Ni and Zn from subsoil samples. Digested sludge applications significantly increased the levels of extractable K, Mg, Fe, Na, and Cr from subsoil samples. In the absence of inorganic P fertilizer applications, the application of digested sludge decreased the extractability of Mn from subsoil samples. Where inorganic P fertilizer was applied, digested sludge applications did not affect the extractability of Mn. Thus, for subsoil extractable Mn, a highly significant ($P>0.01$) interaction effect between P fertilizer and sludge applications was found.

Among samples from the surface horizon some of the concentration values (Table 156) for 0.1 N HCl extractable Cd are greater from surface soil samples than are found for total Cd as presented in Table 155. These discrepancies are due in part to experimental error and to the variability among samples. Even more important though is the fact that nearly all of the Cd added in sludge is extracted by the acid. Comparison of the data given in Tables 155 and 156 for increasing sludge treatments bears out this relationship. Cadmium supplied to soils as a constituent of digested sludge exhibits a high degree of availability for absorption by crop plants as indicated by the fact that extractable amounts are comparable to amounts added.

Contents (Table 157) of P, Mg, Mn, Na, Zn, and B in soybean leaves collected at the early bloom stage were significantly increased by digested sludge applications. Increased concentrations of these six elements in soybean leaves with sludge treatment were significant at the one percent level. Analyses for Hg contents in soybean leaves from selected plots were made and the results are also shown in Table 157. Although the Hg concentration data were not statistically analyzed, they do show a trend toward an increased relationship between leaf contents of the element and quantities of sludge applied. It has been observed from other studies that the Hg content in plant tissues is decreased by digested sludge applications. The additional inorganic P fertilizer apparently increased the content of Mg in soybean leaves. The effect of phosphorus fertilizer on Mg levels in soybean leaves was significant at the 5-percent level.

Increased concentrations of K, Ca, Mn, Na, Zn, and Cd were found to be significantly ($P>0.01$) increased in mature soybean grain with the application of digested sludge. Although significant at only the 5 percent level, it can be seen in Table 158 that P and Mg concentrations are also increased in soybean seeds with increased sludge applications.

Table 157. Total contents of chemical elements in soybean tissue samples collected in 1970. Data are reported on oven-dry (60°C) basis.

SOYBEAN LEAVES

Sludge or water appl. Rate	P**	K	Ca	Mg**	Fe	Mn**	Na**	Cu	Zn**	B	Hg
	Additional phosphorus fertilizer										
	- - - - - Percent - - - - -				- - - - - - Parts per million - - - - - -						ppb
0	0.29	0.96	2.39	0.69	147.3	137.0	667	16.2	75.7	90.9	200
1/4 Max	0.33	1.05	3.06	0.80	140.7	250.0	906	16.6	183.3	75.1	182
1/2 Max	0.35	1.23	3.21	0.77	153.3	429.0	1037	16.5	248.7	70.9	
Max	0.38	1.45	3.03	0.66	143.3	526.7	1350	16.5	368.0	91.4	165
H_2O	0.29	1.78	1.87	0.44	136.0	146.7	738	16.5	59.7	138.7	
	No additional phosphorus fertilizer										
0	0.28	1.42	2.22	0.54	147.0	131.0	596	18.1	77.6	64.5	
1/4 Max	0.32	1.20	3.06	0.75	142.0	242.0	918	17.2	188.7	62.2	186
1/2 Max	0.35	1.21	3.12	0.69	153.0	349.0	1190	16.3	305.3	56.0	
Max	0.37	1.42	2.90	0.63	154.3	569.0	1356	17.5	344.0	90.0	198
H_2O	0.28	1.99	1.71	0.44	148.3	127.0	678	16.8	71.3	126.9	

* Treatment effects significant at 5% level
** Treatment effects significant at 1% level

Table 158. Total contents of chemical elements in soybean tissue samples collected in 1970. Data are reported on oven-dry (60°C) basis.

SOYBEAN GRAIN

Sludge or water appl Rate	P*	K**	Ca**	Mg*	Fe	Mn**	Na**	Cu	Zn**	Cd**	B
	Additional phosphorus fertilizer										
	- - - - - Percent - - - - -				- - - - - - - - - Parts per million - - - - - - - - -						
0	0.89	4.18	0.50	0.45	162.0	77.3	410	30.0	153.3	0.44	60.1
1/4 Max	0.93	4.57	0.45	0.48	160.7	78.3	587	29.4	164.7	0.51	54.1
1/2 Max	0.90	4.61	0.43	0.46	144.0	96.0	517	27.3	174.7	0.71	54.6
Max	0.98	4.93	0.45	0.48	148.7	121.7	638	28.9	178.7	0.93	53.3
H_2O	0.89	4.53	0.50	0.43	177.7	77.3	394	29.3	139.3	0.39	65.2
	No additional phosphorus fertilizer										
0	0.86	4.29	0.51	0.43	173.7	79.3	481	30.7	154.0	0.43	60.1
1/4 Max	0.96	4.74	0.45	0.44	185.3	75.7	492	30.9	166.3	0.64	53.5
1/2 Max	1.00	4.68	0.41	0.47	166.0	89.0	591	29.5	174.3	0.86	55.5
Max	1.00	4.88	0.41	0.44	203.7	93.0	507	29.4	175.3	1.24	50.9
H_2O	0.73	4.19	0.47	0.41	156.7	64.3	365	29.6	138.3	0.30	57.0

* Treatment effects significant at 5% level

** Treatment effects significant at 1% level

Furthermore, the increases in concentrations of Mn, Mg and Ca in soybean grain observed with applications of inorganic P fertilizer are significant at the 5-percent level.

Contents of Cr, Pb and Ni in soybean leaves and grain were below detectable limits for the method used. These elements are present at the hundreds of part per billion level or less.

In view of the fact that concentrations of both plant available and slowly-available forms of P are increased in soils at a rapid rate following applications of digested sludge, the split-plot soybean study has afforded an excellent opportunity to make some observation regarding the effect of heavy phosphorus fertilizations on the mobility and plant availability of other elements. It appears that the P build-up in soils may affect the mobility of Fe and Mn and may affect the absorption by plants and/or translocation within the plant of Mn, Mg and Ca.

Experimental procedure: Kenaf - Kenaf (Hibiscus cannabinus) is grown in several tropical countries as a source of textile and cordage fiber. Fibers of the plant have characteristics or properties that make it comparable to most softwoods and superior to hardwoods as a raw material for making paper products. It can also be used as a blend to improve the pulping quality of less satisfactory materials used in the production of paper.

Although very little was known about the fertility requirements or adaptability of kenaf to northern Illinois climatic conditions, it seemed in 1968 to be an ideal crop for use in a digested sludge utilization study. Thus, a field study was established to determine the yield response of several varieties of kenaf to digested sludge application. During the first year, 1968, only one variety was planted in 102 cm spaced rows in 4-row plots, 12-m long. Each of the four rates (same as for the corn study) of sludge application on kenaf plots was replicated four times. In 1969 and 1970, the plots used for kenaf in 1968 were split to accommodate two varieties which were planted with a grain drill in 61-cm row spacing by May 15 each year. In 1969 each treatment for each variety was again replicated four times but in 1970 the treatments were replicated only twice since the east one-half of the area was used to establish an alfalfa study. Digested sludge was applied between the rows of kenaf during the growing seasons beginning each year when the plants had reached a height of 20 to 25 cm.

Results and discussion of the kenaf study - The kenaf yields are reported in Table 159 with respect to variety, year, and treatment. The maximum treated plots received a total of 36.3 to 51.5 tons per hectare of sludge on a dry weight basis each year and during the latter two years the control or check plots were treated with 269 kilograms of N, 305 kilograms of P (P_2O_5), and 134 kilograms of K (K_2O)per hectare.

Table 159. **Kenaf yields obtained with digested sludge treatments. Yields are in tons per hectare adjusted to 20% moisture.**

		Rate of Application in Centimeters				Greatest Application Rate	
Variety	Year	0 1/	0.64	1.3	2.5	Liquid cm/yr	Solids tons/ha
Everglade 71	1968	4.7	8.1	8.3	8.3	17.8	51.5
Cuba 2032	1969	11.2	10.3	10.8	11.9	20.3	36.3
Guatemala 4	1969	10.3	11.4	11.6	12.1	20.3	36.3
Cuba 2032	1970	7.6	8.1	7.2	9.2	17.8	51.7
Guatemala 4	1970	6.3	7.8	7.6	7.6	17.8	51.7

1/ 0 - Received only basic application of 134 Kg/ha of K_2O in 1968 but fertilized with 269-305-134 Kg/ha in 1969, and 1970

2/ All yields reported are averages of those obtained from 4 treatment replications except in 1970 when one-half of the plots were seeded to alfalfa leaving only 2 replications per treatment.

However, kenaf yields were not significantly increased by the added fertility. Either kenaf has a very low fertility requirement or the varieties available are not well adapted to climatic conditions in Illinois or both. At any rate, the yields obtained from kenaf are insufficient for it to compete with corn or soybeans for land on an economical return basis.

Near the end of the first growing season for kenaf (1968), but before frost occurred, samples of leaf tissue were collected from the top most part of the 1.8-to 2.1-meter tall plants. The kenaf leaf samples were analyzed for total N and some metals. Average results are presented in Table 160, but the data were not statistically analyzed. Nevertheless, the increase in leaf N content and decrease in leaf Mg content with increasingly greater sludge applications may be real differences. Both Zn and Mn concentration levels appear to be increased with increasingly greater sludge applications. On the basis of the amount of Zn applied as a constituent of digested sludge, one might expect an increased Zn uptake by plants, but not of Mn. The digested sludge contained Zn concentrations that ranged from about 150 to 200 parts per million, while Mn concentration levels were most often in the range of 3-5 parts per million and seldom exceeded 10 parts per million on a wet weight basis. The amounts of Mn supplied on the plots as a constituent of sludge were very small compared to native amounts in the soil. None of the concentration levels of Cr, Pb or Ni were high enough to be detected by the method used.

Table 160. Total nitrogen and selected metal contents of kenaf leaf tissues after the first growing season (1968) during which 17.8 cm of liquid digested sludge (equivalent to 51.5 tons of solids per hectare) were applied on the maximum treated plots.

Sludge appl. Rate	N	Mg	Zn	Mn
	- - Percent - - -		- - - ppm - - -	
0	1.87	0.53	44.0	62.5
1/4 Max	3.51	0.51	77.5	100.0
1/2 Max	3.60	0.50	130.0	310.0
Max	3.77	0.44	123.5	312.5

Experimental procedure: alfalfa - As mentioned above, one-half of the plots formerly planted each year to kenaf were seeded to alfalfa in the spring of 1970. The alfalfa was established in the absence of a nurse crop by the use of a herbicide. The first cutting of alfalfa was made on July 7, 1970. Only one additional application of sludge was made on the alfalfa plots in 1970 prior to taking the first cutting. From Table 159, it may be seen that a total of 20.3 cm of digested sludge was previously applied on the maximum-treated plots when the area was in kenaf. Alfalfa yields are given in Table 161 for the first cutting of alfalfa.

Table 161. Alfalfa yield obtained during the first cutting after establishment in 1970. Maximum-treated plots had received a total of 20.3 cm of digested sludge (36.3 dry tons/ha) before the establishment of alfalfa and 2.54 cm (36.3 dry tons/ha) after establishment.

Sludge appl Rate	Yields Tons/ha
0	3.78
1/4 Max	5.11
1/2 Max	4.93
Max	3.72

Results and discussion of alfalfa study - The first cutting yields did not significantly differ as a result of treatment and while alfalfa was clipped again in August and September 1970 the yields were not recorded. After each cutting of forage additional digested sludge was applied. The maximum-treated plots received 15.2 cm in July, 2.5 cm in August and 2.5 cm in September of additional sludge for a total application in 1970 of 20.2 cm.

Soil samples were collected from the alfalfa plots in April 1971 after a total of 40.5 cm of sludge had been applied on the maximum-treated plots, during a period of three years. Total contents of selected chemical elements in surface and subsoil-samples are given in Table 162. Digested sludge applications have resulted in greater total concentration levels of several metals in the surface-soil samples, but levels in subsoil samples have been changed very little, if at all. In Table 163 the concentration levels of extractable chemical elements in both surface- and subsurface soil samples are given. The extractability of P, Fe, Cu

Table 162. Total contents of selected elements in the 0- to 15.2-centimeter depth and 30.5-to 45.7-centimeter depth of the alfalfa experiment. Soils were sampled in April 1971 after 40.5 centimeters of sludge equivalent to 73.3 t/ha solids had been applied on the maximum-treated plots.

Sludge Appl. Rate	P	K	Ca	Mg	Fe	Mn	Na	Cr	Cu	Pb	Ni	Zn	Cd	B	Hg
	ppm	- - - -	%	- - - -		ppm	%	- - - -	- - - -	- - - -	- - ppm	- - - -	- - - -	- - - -	- -
0-to 15.2-Centimeter Depth															
0	378	1.86	.26	.20	1.61	732	.59	32	17	25	20	78	1.8	26	0.04
1/4	558	1.91	.27	.20	1.70	1028	.61	56	27	38	22	112	3.7	26	0.09
1/2	616	1.89	.28	.24	1.38	696	.64	57	29	33	23	131	4.3	22	0.11
Max	1000	1.93	.28	.20	1.81	1322	.60	90	39	50	26	201	7.6	25	0.21
30.5-to 45.7-Centimeter Depth															
0	375	2.09	.15	.33	3.49	448	.37	53	28	24	40	98	2.2	n.d.	n.d.
1/4	392	2.11	.14	.34	3.61	430	.35	56	30	28	41	100	2.3	n.d.	n.d.
1/2	371	1.83	.18	.33	3.08	386	.42	46	25	24	32	88	1.9	n.d.	n.d.
Max	394	2.07	.16	.34	3.42	406	.41	54	30	28	38	100	2.1	n.d.	n.d.

n.d. - not determined

Table 163. Extractable contents of elements in the 0-to 15.2-centimeter and 30.5-to 45.7-centimeter soil depth of the alfalfa experiment. Soils were sampled in April 1971 after 40.5 centimeters of sludge equivalent to 73.3 t/ha solids had been applied on the maximum-treated plots.

Sludge Appl. Rate	P*	K	Ca	Mg	Fe*	Mn	Na**	Cr	Cu*	Pb**	Ni	Zn*	Cd	B
	- ppm -		%	- - -	- - -	- - -	- - -	- - -	ppm	- - -	- - -	- - -	- - -	- - -
0-to 15.2-Centimeter Depth														
0	28	186	.21	732	506	288	13	n.d.	4.0	8.4	n.d.	16	.38	.47
1/4	93	206	.24	788	672	366	38	n.d.	14.0	11.0	n.d.	51	1.8	.80
1/2	174	202	.18	582	678	282	28	n.d.	16.0	10.0	n.d.	69	2.4	.68
Max	332	263	.24	797	855	458	22	n.d.	24.0	18.0	n.d.	115	4.0	1.01
30.5-to 45.7-Centimeter Depth														
0	8	363	.14	.13	834	78	24	n.d.	4.1	6.3	n.d.	7.4	.09	n.d.
1/4	10	368	.15	.12	890	77	42	n.d.	3.9	5.6	n.d.	8.2	.07	n.d.
1/2	19	308	.13	.12	913	68	48	n.d.	5.6	3.1	n.d.	10.0	.05	n.d.
Max	13	342	.14	.12	907	64	36	n.d.	6.6	5.8	n.d.	13.0	.34	n.d.

* Treatment effects significant at 5% level

** Treatment effects significant at 1% level

n.d. - not determined

and Zn was significantly increased at the 5 percent levels in the soil surface by sludge applications. Only extractable Na concentrations in surface-soils showed a highly significant increase as a result of sludge treatments. Furthermore, only the extractability of Na was significantly (P>0.05) increased in subsurface-soil samples by sludge treatments.

The total contents of selected chemical elements in whole alfalfa plant samples are given in Table 164. The plant samples were taken from the first cutting in 1970. The data were not statistically analyzed, but the trend for increased concentration levels of Zn and Mn in alfalfa with increased sludge applications is similar to findings from analyses of other plant leaf tissue samples.

Table 164. Total contents of selected chemical elements in alfalfa whole plant samples collected in 1970. Data are reported on oven-dry (60°C) basis.

Plot Number	Sludge appl Rate	N	P	K	Ca	Mg	Fe	Mn	Na	Cr	Cu	Pb	Ni	Zn	Cd	Ash
		- - - - - - -	- - - -	%	- - - -	- - - -	- - - -	- - - -	- - - -	- - - -	ppm	- - - -	- - - -	- - - -	- - - -	%
7	0	2.91	.20	1.87	1.29	.34	428	38	970	4.2	11.0	n.d.	3.6	53	3.0	7.91
13	0	2.89	.21	2.30	1.11	.26	232	47	800	3.9	15.6	n.d.	4.0	37	2.5	8.66
6	1/4 Max	2.95	.29	2.05	1.26	.36	171	42	1354	3.5	10.1	n.d.	5.2	87	6.2	8.33
14	1/4 Max	2.89	.19	2.33	1.20	.26	151	52	984	3.7	15.2	n.d.	3.6	68	4.0	8.50
8	1/2 Max	2.92	.19	2.03	1.26	.31	144	118	984	2.8	7.2	n.d.	5.6	150	6.2	8.90
15	1/2 Max	2.95	.26	2.80	1.45	.33	149	365	866	3.5	15.6	n.d.	10.4	200	9.0	10.24
5	Max	3.15	.24	2.42	1.30	.33	157	134	784	4.8	7.1	n.d.	7.5	215	7.6	9.55
16	Max	3.14	.25	2.53	1.73	.45	186	529	810	3.7	15.2	n.d.	15.6	376	11.8	11.23

(NOTE: These references have not been verified by the Office of Solid Waste Management Programs.)

SECTION IX

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SECTION X

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