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# PHOTOSYNTHETIC RECLAMATION OF AGRICULTURAL SOLID AND LIQUID WASTES

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# PHOTOSYNTHETIC RECLAMATION OF AGRICULTURAL SOLID AND LIQUID WASTES

by

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Abstracts: The overall objective of this study was to develop a system ith which a large fraction of the wastes produced by agricultural ndustries could be converted into a useful material without imposing n unacceptable burden on the environment. Specifically, the project nvolved a detailed study of the basic characteristics of an integrated naerobic fermentation and algae growth system for agricultural solid nd liquid wastes on a laboratory and small pilot plant scale, with pecial attention being devoted to the reaction kinetics of the systen, substantial degree of nutrient recovery and recycle was also attained n the system.


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## REVIEW NOTICE

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## FOREWORD

Man and his environment must be protected from the adverse effects of pesticides, radiation, noise and other forms of pollution, and the unwise management of solid waste. Efforts to protect the environment require a focus that recognizes the interplay between the components of our physical environment--air, water, and land. The National Environmental Research Centers provide this multidisciplinary focus through programs engaged in

- studies on the effects of environmental contaminants on man and the biosphere, and
- a search for ways to prevent contamination and to recycle valuable resources.

In an attempt to solve the problems involved in solid waste disposal, this study attempted to develop a partially closed system for animal waste management based on the integration of an anaerobic and aerobic phase, the recycling of water, and the reclamation of usuable products.

A. W. Breidenbach, Ph.D.
Director
National Environmental
Research Center, Cincinnati

## PREFACE

The report herein presented is of the nature of a final report covering the three year grant period, supported in part by USPHS Grant No. 5 RO1 UI 00566-03 and in part by the College of Engineering and School of Public Health, University of California, Berkeley.

The research plan on which the grant was based called for laboratory and pilot plant studies to develop a partially closed system of animal waste management based on the integration of an anaerobic and aerobic phase, the recycling of water, and the reclamation of usable products. Contained herein are summaries of the first and second progress reports, and an account of experiments performed in the final three-four-month period of the research. The first and second reports covered the first two and one-half year period of the research. The pilot plant included a poultry enclosure, a hydraulic system for handling the wastes, a heated anaerobic digester with ancillary equipment, and an algae-production pond.

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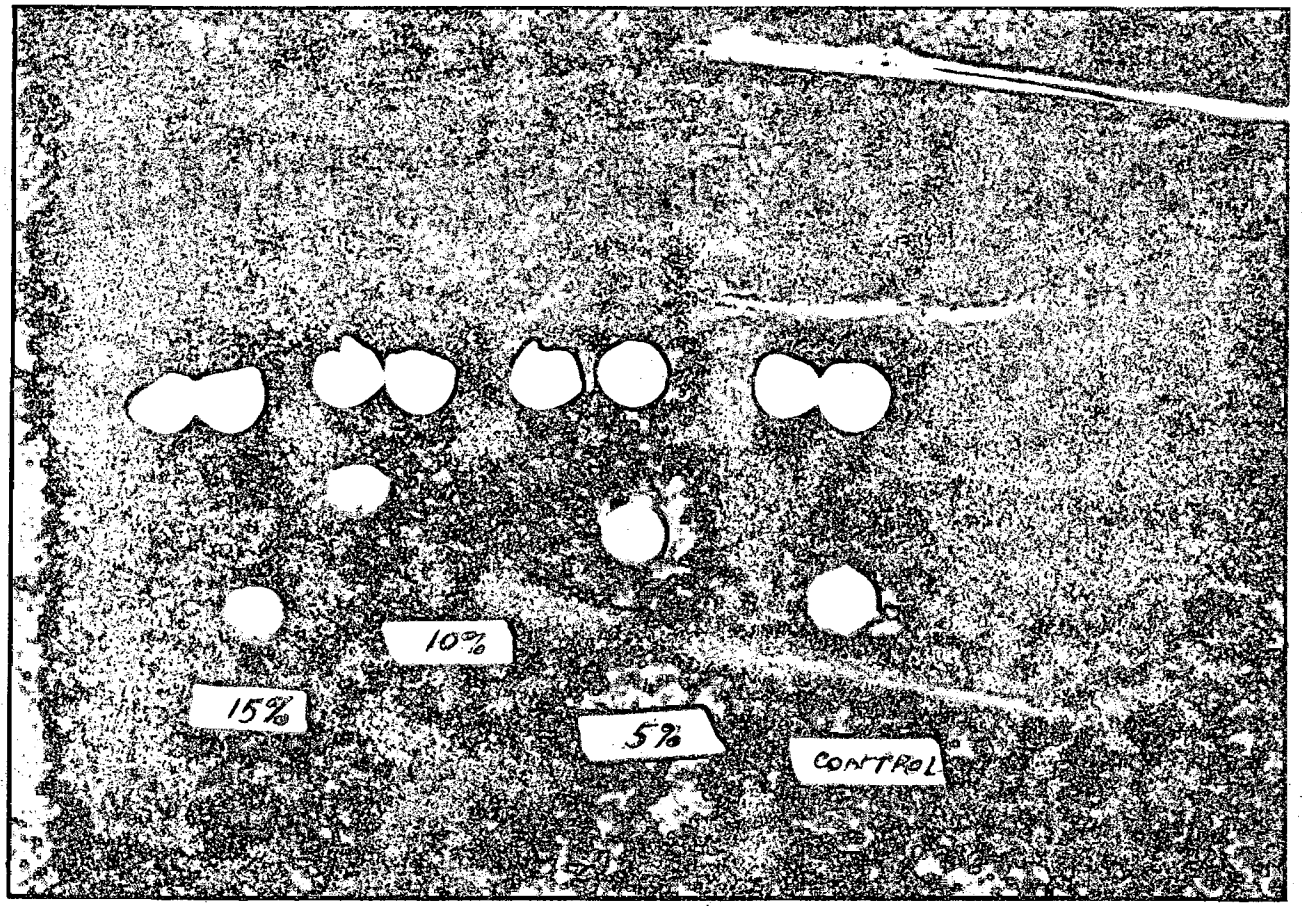



FRONTISPIECE: EGGS FROM CHICKENS FED 5 PERCENT, 10 PERCENT AND 15 PERCENT ALGAE GROWN ON CHICKEN WASTE PLUS 15 PERCENT, 10 PERCENT AND 5 PERCENT SOYBEAN OIL MEAL. THE EGG ON THE RIGHT WAS FROM A CHICKEN FED A 20 PERCENT SOYBEAN MEAL AS A CONTROL

NOT REPRODUCIBLE

# I. INTRODUCTION

## NEED FOR THE STUDY

The expanded urbanization of our population, attended as it is by a decentralization of the majority of the metropolitan areas in the United States, leads to an increasing degree of encroachment on the agricultural lands surrounding urbanized regions. In turn, the multiplication of uses and augmentation of demands for these agricultural lands inevitably is accompanied by an increase in the monetary value of the land and a concomitant increment in taxation. The result is that the agricultural enterprises located in these areas must intensify their operations if they are to survive. The intensification of agricultural industries leads to a corresponding step-up in waste production by these industries. The increase in the volume of the waste production is not the only problem, however. More serious is the fact that inasmuch as agricultural wastes generally are highly putrescible in nature, their disposal in an acceptable manner becomes an urgent necessity. The problem is aggravated by the marginal nature of the economics of the agricultural industry in general, and its consequent inability to expend the money needed to treat its wastes satisfactorily. The general unavailability of land area plus the prohibitive costs of the land make the formerly acceptable method of disposal by spreading on land no longer feasible in many areas. Thus, the sequence of events triggered by the expansion of urbanization leads to the need for research directed at developing new systems of waste management consonant with modern conditions.

The situation described above is especially typified in the poultry (shell-egg) industry. Even a relatively small operation of 10,000 chickens results in the production of at least two tons of wet manure per day. (The term "chicken manure" denotes mainly manure with small quantities of spill-feed, broken eggs, feathers, etc.). Unless carefully managed, this manure can lead to the production of very objectionable odors and the generation of huge populations of flies. The manure usually cannot be spread on land because land is either unavailable, too expensive, or too wet. It cannot be sold as a fertilizer, at least in sufficient quantities, because large-scale crop producers are unwilling to use the less convenient organic fertilizers when chemical fertilizers are at hand. Even if the operations were located so that they could be connected to municipal sewerage systems, discharging the wastes into the sewer undoubtedly would be forbidden by the affected municipality. The added burden of treating wastes from one or more large-scale poultry operations would probably overtax the capacity of the existing facilities most of which are already inadequate. Thus, in all probability, increasing the capacity of the facilities to meet the added burden would necessitate expenditures on a scale unacceptable to the taxpayers.

Since the presently practiced methods of disposal of animal wastes are becoming unsuitable, it is imperative that new methods be found which are sanitarily and aesthetically acceptable and yet economically feasible. Ideally, the method or methods should involve some type of reclamation, not only to conserve resources, but to utilize solar energy. Photosynthetic reclamation of manures in the form of algae production is one possibility.

The photosynthetic reclamation system proposed in this study

incorporates the hydraulic handling of animal wastes. A brief description of the system is as follows: The animals' wastes are trapped on a flat wet surface then quickly flushed to a holding tank in which settleable solids are separated from the liquid phase. The supernatant is pumped directly to an algae pond and the settled solids are discharged into an anaerobic digester. Digester supernatant is pumped directly to the algae pond, and the settleable, stabilized sludge (dewatered and greatly reduced in volume) is wasted. Depending upon the algae concentration, pond effluent either is recycled directly to the animal quarters for flushing, or the wastes can be processed so that the algae are removed. A portion of the supernatant from the separation process is pumped to the animal quarters for waste flushing. The algae are dried for use as an animal feedstuff. As the description indicates, the system is "closed" so far as water is concerned. However, in practice, some water is naturally lost due to product removal, evaporation and spillage. This can be compensated for by discharging the necessary overflow from the animals' drinking water supply into the system.

## OBJECTIVES OF THE STUDY

The overall objective of the study was to develop a system with which a large fraction of the wastes produced by agricultural industries can be converted into a useful material without imposing an unacceptable burden on the environment. Specifically, the project involved a detailed study of the basic characteristics of an integrated anaerobic fermentation and algae growth system for agricultural solid and liquid wastes on a laboratory and small pilot plant scale, with special attention being devoted to the reaction kinetics of the system.

As will be documented, a substantial degree of nutrient recovery and recycle was also attained in the system.

## NATURE AND RATIONALE OF THE STUDY

To attain the objectives of the study, it was not enough to establish tentative design and operational parameters of the constituents of the proposed system by way of experimentation. To complete the study, the applicability of the design and operational parameters had to be tested by way of the construction and use and operation of a pilot plant of meaningful size which incorporated these design and operational parameters.

The activities which took place in the laboratory phase included preliminary studies concerned with establishing guidelines with respect to the planning of experiments to be conducted with the pilot plant, with determining the characteristics of manure for use in laboratory experimentation in terms of source of study, and with evaluating methods for collecting and storing the manure. Pre-pilot plant activities were directed at refurbishing existing facilities to be used in the investigation, and at conducting preliminary pilot plant studies. The pilot plant phase took up the greater part of the total research period.

## ORGANIZATION FOR THE STUDY

The organization of the study is indicated by the information given in the Preface. The organization of the report is indicative of the chronology of the research effort. The laboratory studies were

conducted during the first year of the grant period. The pre-pilot plant phase of the research began toward the end of the first year and continued into the second. The pilot plant phase, naturally, began with the completion of the pre-pilot phase and continued up to the termination of the project.

ACKNOWLEDGMENTS

The research reported herein was supported in part by a Public Health Service Research Grant (UI 00566-01, 02, 03) from the Bureau of Solid Waste Management, U.S. Dept. of Health, Education, and Welfare (presently Solid Waste Research, U.S. Environmental Protection Agency) to the Regents of the University of California.

## II. SUMMARY OF RESEARCH DESCRIBED IN PROGRESS REPORTS I & II

### INTRODUCTORY REMARKS

The experimental effort in the research project "Photosynthetic Reclamation of Agricultural Solid and Liquid Wastes" was directed towards the finding of ways of converting into useful materials the agricultural wastes now causing severe environmental problems. Specifically, the work involved making determinations of the energy, organic loadings, balance of water and nutrients, and overall performance of an animal waste treatment system consisting of an anaerobic reactor preceding an algae growth pond.

b

Because the problem of manure disposal is especially critical in the poultry industry, and because at the time facilities available at the Richmond Field Station were best suited to the use of poultry manure for the study, the major effort of the research was concerned with chicken manure.

PRE-PILOT PLANT STUDIES

Because the proposed system had as one of its components, the anaerobic digestion of poultry manure, laboratory studies on the factors involved in poultry manure digestion were indicated. To develop the necessary cultures, cultures of digesting sewage (domestic) sludge were adapted to the use of manure as a nutrient source. In these studies the existence of a very critical concentration of volatile solids with respect to satisfactory digestion of poultry manure was demonstrated. This level was found to be about 50% of the dry weight of the manure as volatile solids (VS). In the experiments, the manure digested readily when its volatile solids were 62% or above; and very poorly, when the volatile solids were less than 50%.

To determine the optimum operating conditions for the algae production pond, it was necessary to know the algal growth potential (AGP) of the liquids discharged into it, namely, of the supernatant from the anaerobic reactor (digester) and the supernatant produced as a result of the incorporation of the flush method and accompanying settling tank to store the flushings. Tests showed that in terms of AGP, the optimum ratio of digester effluent supernatant to settled manure trough-flushings was from 1:10 to 1:20.

In terms of the objectives of the research, the most practical of the various methods of handling chicken manure was one based on hydraulic transport. To design the hydraulic system, information was needed on the relation between settleability and solids content of aqueous suspensions of the manure. Results of experiments involving poultry manure suspensions having solids concentration ranging from 0.1% to 30% wet weight indicated that: a) approximately 70% of the solids settle out; b) the higher the initial concentration, the slower the rate of settling; and c) that at the most probable maximum wet weight of solids concentration to be encountered in the system, namely, from 1% to 3%, most of the settling would take place in less than 15 minutes.

The experiments on the rate of settling to solids concentration of slurry were later repeated with the use of cow manure instead of poultry manure. Rates of settling of slurries of cow manure paralleled those for poultry manure slurries, and are plotted in Figure 1 as a function of solids content. The results indicate that the acceleration accompanying increase in amount of dilution also is applicable to slurries of cow manure.

A hydraulic manure-handling system incorporating a program of frequent flushing appeared to be the practical solution for wastes removal and transport in the contemplated operation. To arrive at a design for the flushing system, two model tipping buckets, one having a capacity of 1 gal, and the other of 2 gal, were mounted on a trough (12 in by 96 in long) designed to be adjustable to any desired slope. The 2-gal bucket with the trough at a slope of 1:16 supplied enough flushing action to transport the manure when an equivalent of approximately two hours of manure production was placed in the trough. A critical fact brought out

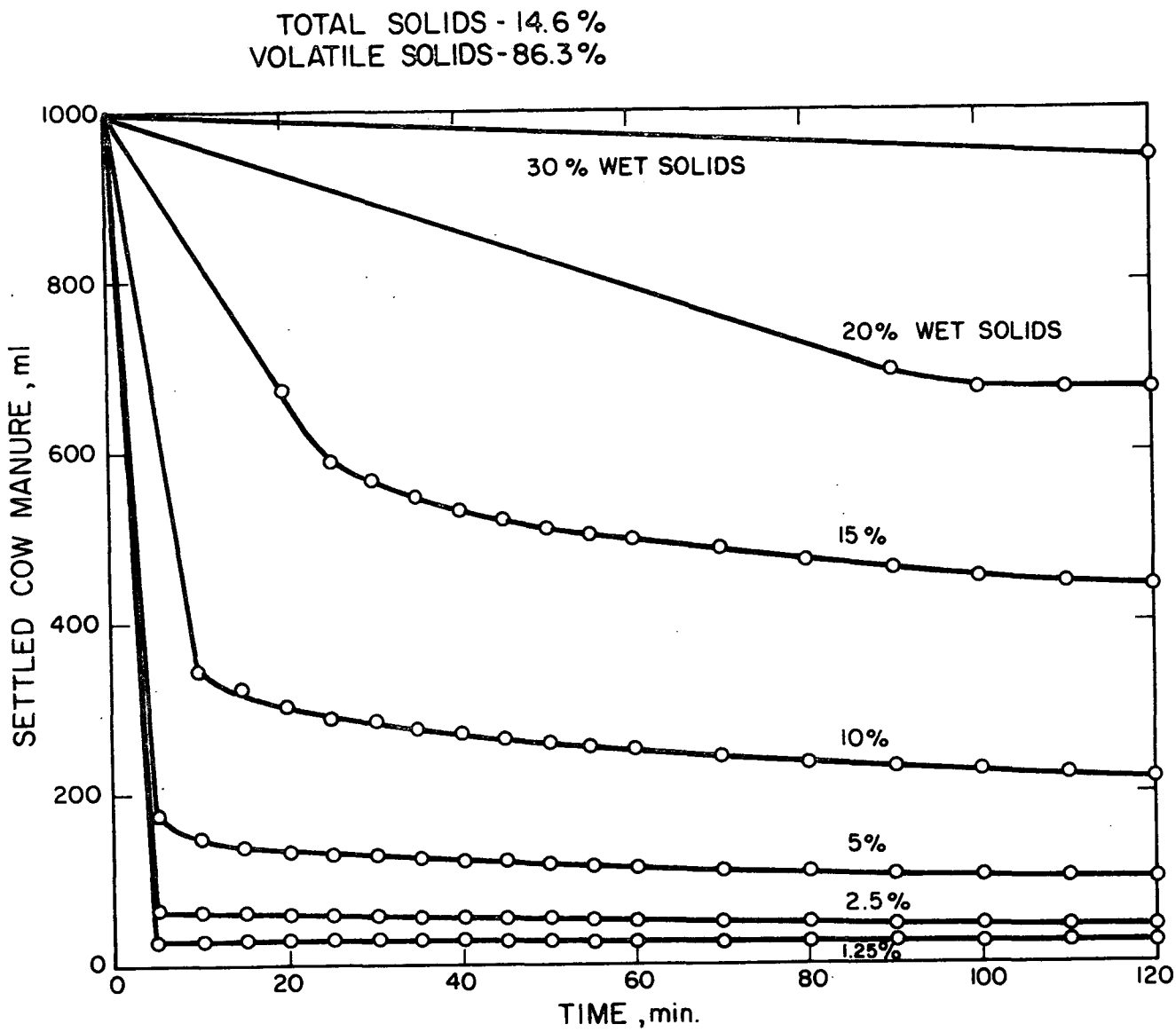



FIG. 1.-SETTLING TIME CHARACTERISTICS OF DILUTED COW MANURE[1]

1. Fecal matter only

in the test was the importance of not allowing the manure to dry on the trough surface, since it took a considerable force of water to dislodge dried material. Also drying of manure is accompanied by loss of $CO_2$ and ammonia, a factor which was decreased by prompt and frequent flushing.

Prior to the completion of the integrated pilot plant system, a closely controlled heated digester (3 ft in diameter, 8 ft in depth; culture volume 45.4 cu ft) was constructed. To arrive at parameters for later operation, a series of preliminary experiments were conducted with the use of the new digester. Operation of the digester was initiated by introducing well-digested sewage sludge into the digester and adapting it to chicken manure by starting with an initial average low loading of 0.021 lb VS cu ft-day. The pH ranged from 7.2 to 7.8. At detention times ranging from 28 to 65 days (mean 40 days), digester gas production at loading intervals of four to six days per week ranged from 5.0 to 11.25 cu ft/lb VS introduced.

The algae growth pond was in existence prior to the initiation of the present project. The planned culture depth in the pond was 7 in. At this level, the surface area of the culture was 737 sq ft, and the culture volume about 3,200 gal. Arrangement was made to mix the pond for 10-minute periods at hourly intervals by recirculating the liquid such that it acquired a velocity of 2 ft/sec. Such mixing is essential to prevent stratification and anaerobiosis in the sludge phase of the algae ponding system.

Two methods of algae harvesting were used, namely, settling and centrifugation. In harvesting by settling, pond effluent was passed through a cyclindrical settling tank having a conical bottom at a rate such that the detention time of the pond effluent in the tank was 38

hours. The settled algae were drawn off periodically. A Westphalia centrifuge and a laboratory basket-type centrifuge were used in the centrifuge operation.

Because of the rapid decomposition of volatile solids occurring under normal storage conditions and of the difficulty in procuring a constant and uniform supply of manure from nearby outside sources, the need for establishing a chicken colony at the site was demonstrated early in the study. Accordingly, a 14 ft by 14 ft poultry enclosure was designed and constructed, and was stocked with a flock of 113 twenty-week old white leghorn pullets. The birds were caged in batteries, each of which held four hens (except one cage with five hens during the initial part of the study) with a space allotment of about 0.45 sq ft/hen.

Directly beneath each row of chicken batteries was a fiberglass coated plywood trough to catch the chicken excreta. Once each hour, a tipping bucket (8.3 gal capacity) mounted at the end of each trough automatically discharged its contents into the trough to flush the excreta down the trough, through the disposal unit, and into a sedimentation tank. The working depth of the sedimentation tank was about 4 ft. A submerged sump pump moved the supernatant in the sedimentation tank to the algae pond, and a sludge pump moved the settled chicken manure hopper for calibration prior to discharge into the digester.

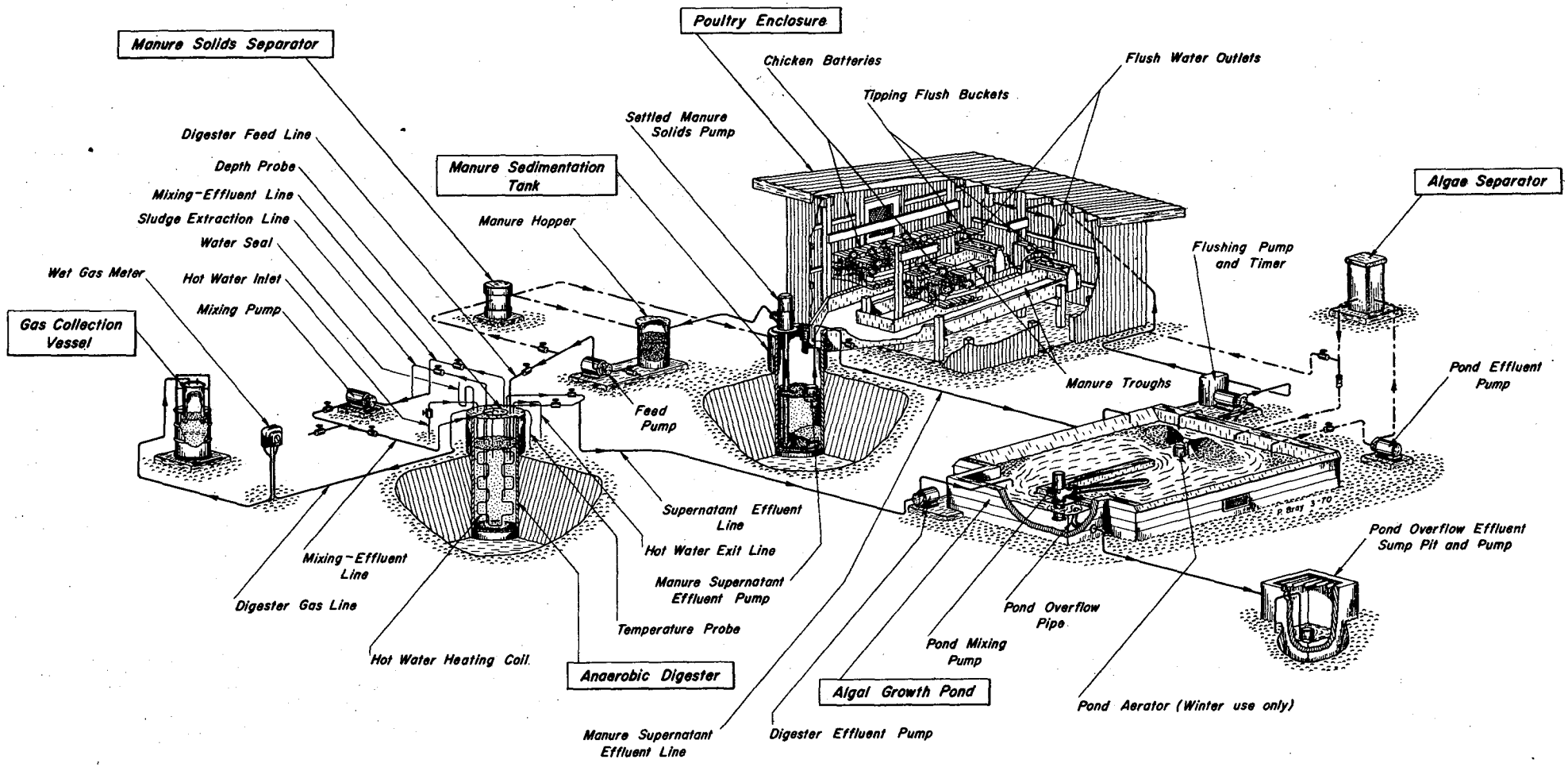
One of the more significant outcomes of the study was the success attending the functioning of the manure flushing and hydraulic transport subsystems. The success was evidenced by the practically complete lack of objectionable odors and the absence of fly breeding in the area.

A thin layer of slime did build up on the surface of the troughs. However, extensive buildup of the layer was prevented by scraping approxi-

mately every two weeks with a "squeegee". Perhaps this periodic scraping may have been unnecessary, inasmuch as the layer might have sloughed off after reaching a critical depth, much as do the slime accumulations on the media of trickling filters. The biological composition of the slime layer was much the same as that of coatings on rocks in trickling filters, except that the former had a dense population of deep pink-colored organisms--probably a species of red spirilla. A thin layer of algae developed in those portions of the troughs exposed to sunlight. Undoubtedly, the organisms in the slime layer were responsible for some decomposition of the manure in the troughs in the intervals between flushings and effected some loss of nutrients in the system.

PILOT PLANT STUDIES

Upon completion of the poultry enclosure, the enclosure, sedimentation tank, digester, and algae pond were integrated to form the pilot plant diagrammed in Figure 2. The flow arrangement may be briefly described as follows: wastes excreted by the chickens drop into manure troughs. The excreta are then flushed down the manure troughs into a sedimentation tank. Supernatant in the sedimentation tank is pumped to an algae pond, while the settled solids are measured and introduced into a digester. Digested stable sludge periodically is wasted to the environment when necessary to provide volume for newly admitted solids. Supernatant from the digester is discharged into the algae pond. Effluent from the algae pond either may be passed through an algae separation process or may be pumped directly to the manure troughs and recycled through the system. If the effluent is passed through an algae-separation process, the harvested algae are removed from the system and a portion of the supernatant is pumped to the manure troughs for use as




**Poultry Operation With An Integrated Sanitation-Waste Materials Recycling System**

flushing water, thus completing the cycle. Supernatant not diverted for flushing is returned to the pond. Physical dimensions, construction and equipment details as well as operational features of each of the units mentioned in the flow chart were given in the progress reports.

The operation of the pilot plant progressed without any major problems throughout the course of the study. Out of the 113 chickens at the start of the study, 88 remained at the end of the first run. Of the 25 chickens that were removed due to mortality or morbidity, 12 were killed by marauding dogs that gained access to the pens. Egg production ranged from 68.7% to 81.4%. Feed consumption averaged 0.258 lb/chicken/day. Manure output averaged 0.418 lb/chicken/day (189.7 gm/chicken/day) with an average solids content of 25.4% of the wet weight, and a volatile solids content of 67.7% of the dry weight. The population equivalent was 9.2 chickens per capita. No difficulties were encountered in the manure flushing operation. The quantity of flushing water (i.e. recycled pond water) was approximately 12,000 liters per week and the drinking water around 4000 liters per week.

The systems approach was used to analyze the pilot plant phase. A "balance" of the major inputs, outputs, concentration changes within the system, and $\Delta$ system changes (not accounted for in the measurements) was performed for total solids, volatile solids, total unoxidized nitrogen, and energy for the system components, chickens, sedimentation tank, digester, and algae pond. The balances were performed weekly. The digester operation was terminated at the conclusion of week 24.

Sedimentation Tank:

The sedimentation tank received 1053 kg of manure total solids from the chicken component. The supernatant was pumped directly to the

algae pond and the settled solids were pumped to the digester from the start of the study through week 24. Thereafter, the settled solids were dewatered, dried, and stored for use in feed studies. From week 5 through 24, the distribution of the sedimentation tank contents was 57% sedimentation tank supernatant to the algae pond, 34% settled solids to the digester, and 9% "grit" was removed from the settled solids before it entered the digester. The $\Delta$ outputs were: $\Delta$ total solids 24%; $\Delta$ volatile solids 28%, $\Delta$ total unoxidized nitrogen 15%; and $\Delta$ energy 32%.

The settling efficiency obtained in the sedimentation tank was not as high as might have been expected on the basis of the laboratory studies. About 57% of the supernatant discharged from the sedimentation tank was in the form of dissolved and suspended manure solids. Using a manure slurry of the same concentration (1% manure solids, wet weight) as in the sedimentation tank, only about 33% of the manure solids remained in the supernatant of slurry placed in an Imhoff cone. This difference would lead one to conclude that through better designing, the efficiency of a manure sedimentation tank could be increased from the observed 43% to highs of 60% or 65%. The desirability of increasing the settling efficiency in a practical operation would depend upon the design capacity and mode of operation of the aerobic and anaerobic constituents of the plant.

The grinder unit which was installed in the sedimentation tank to intercept and grind feathers discharged with the manure did not function as well as was expected. Because of the structure of chicken feathers and, more likely because of the spacing between the grinding surfaces of the machine, many of the feathers passed through the grinder unground. Although the obvious solution would be to reduce the spacing between the

grinding surfaces, doing so could only lead to a more serious problem, namely clogging. Since feathers float, a more likely solution would be to install some type of a skimming device at the surface level of the liquid in the sedimentation tank, or in a large operation to utilize a feather screen which is available commercially.

Digester:

The loading to the digester component ranged from 0.040 to 0.054 lb VS/cu ft-day (mean 0.043) with an average detention time of 22.9 days. Gas production was nearly 12 cu ft/lb volatile solids introduced. Digester gas composition was grouped into $CH_4$, $CO_2$, and nonidentified gas. The $CH_4$ content ranged from 13% to 45% and $CO_2$ ranged from 41% to 67%. The $CH_4$:$CO_2$ ratio increased from 0.18 in week 7 to 0.96 in week 24. Nondefined gas averaged about 20% of the total. Overall volatile solids destruction was approximately 55%. Out of the 185 kg of digest influent total solids content, 120 kg were released as digester gas.

Judging from the information given in the preceding paragraph, the digester functioned with an efficiency comparable to that characteristic of municipal digesters. Although the volume of gas produced was greater than that generally encountered in the digestion of raw sewage sludge, the methane content was correspondingly lower. It started at 12% $CH_4$, but by the end of the 24-week period in which the digester was operated it had reached 40%. In view of the steady increase, there is good reason for believing that with continued operation, the percentage of $CH_4$ would have reached the conventional 60% to 65%.

Pond:

The installation and operation of an algae pond in a photosynthetic

reclamation system such as described herein can serve many useful purposes. It can provide: 1) an effective and odor-free treatment for the solids (dissolved and suspended) coming from the sedimentation tank and for the digester supernatant; 2) a supply of water for the flushers; and 3) a highly proteinaceous plant crop. As a waste treatment device, it functions much as an activated sludge system, differing from conventional activated sludge systems in that much of the oxygen comes from algal activity rather than by mechanical induction of atmospheric oxygen.

The hydraulic detention time of the algae pond for the 36-week period ranged from 9 days to infinity. Excluding the one infinity value, the average detention time was 22 days. The algae pond received a total solids loading of 1151 kg from the sedimentation tank supernatant (including recycled flushing water) and digester supernatant. The measured total solids outputs as percentages of input were: centrifuged harvested algae, 3%; settled harvested algae, 5%; sump output, 17%; and recycled flushing water, 55%. The ratios pertaining to the other parameters, namely, volatile solids, total unoxidized nitrogen, and energy were quite similar to those for total solids. The $\Delta$ outputs were: total solids, 20%; volatile solids, 21%; total unoxidized nitrogen, 17%; and energy 12%. The $\Delta$ energy did not include solar energy input directly. Photosynthetic efficiency as measured by the harvested algae and the visible solar energy input averaged 0.64%, with a high of 2.8%. Low efficiency was primarily due to long detention time in the reactor. A study of the diurnal changes in pH, alkalinity, dissolved oxygen, and calculated inorganic carbon uptake produced results that were within 1% of values calculated according to the classical photosynthesis formula.

An interesting development in the study was the apparent absence

of a detectable accumulation of dissolved solids in the algae pond. When the operation was initiated, it was taken for granted that the dissolved solids concentration would increase because the system was closed to some extent. Under such a circumstance, with salts coming in as manure solids and the small amount with the tap water, coupled with loss of water through evaporation should have led to some concentrating of dissolved solids. Although there was some effluent from the pond by way of sump overflow, the amount of solids discharged in this manner was only 18% of the input of solids into the system. Solids removed as algae from the system amounted to only an additional 8%. About the only explanation for the lack of accumulation would be one involving a biological conversion of a large fraction of the incoming solids to gaseous volatile substances. This apparently occurred. In view of the absence of accumulation, it would seem feasible to operate the system as a closed one or a nearly closed one until the salt buildup reached a level at which algae and bacteria would be seriously affected. Of course, there remains the possibility that in a closed system an accumulation of some toxic metabolite of one of the microbial groups may reach inhibitory proportions long before that from an inorganic salt buildup, but such information can only be obtained through a more sustained operation of a pilot plant than was possible in this study.

The Integrated System:

An analysis of the integrated system reveals that biological activity in the sedimentation tank, digester, and algae pond decreased the total solids by 60%; the volatile solids by 62%; the total unoxidized nitrogen by 45%; and the energy by 56%. The extent of the biological destruction of incoming solids was substantial. An overall analysis

of the integrated system for a 31-week period revealed a loss of about 60% of the incoming total solids. The most likely explanation for the loss is conversion to gas through biological decomposition, both aerobic and anaerobic. In addition to the loss attributable to gasification, settled solids accounted for 13%; sump outflow, 18%; harvested algae, 8%; and grit, 7%. (Settled solids were those solids routinely removed from the sedimentation tank after the digester was discontinued.) Had the digester been continued throughout the run, the loss attributable to settled solids would have been correspondingly higher. The grit is mainly the inert material found in chicken feed. Since it is inert, disposal of it would pose only a mechanical problem. If a more effective algae harvesting system had been available during the run, the solid conversion to algae would have been appreciably greater. The conversion of solids to algal cells is the reclamation feature of the integrated system, since waste solids are converted to algae which can be used as a highly proteinaceous feedstuff.

## III. STUDIES CONDUCTED FOLLOWING PUBLICATION OF THE PROGRESS REPORTS

### INTRODUCTORY REMARKS

At the time the second progress report was written, the authors had good reason for assuming that the project would be funded for an additional year or two so that certain questions concerning the present and future performance of the system could be explored. Consequently, it was planned to follow the solids that must surely accompany the continued operation of the plant as a "closed system", and thus determine the rates required for repletion of necessary elements and removal of undesirable elements to assure the continued successful operation of the

system. Another question to be answered was that of the minimum surface area of pond per bird. As was pointed out in the second progress report, the pond area could be reduced to 2 sq ft/bird without any adverse effects. Although intuition would indicate a limitation somewhere between 1.5 and 2 sq ft, this important question requires an answer more firmly based than intuition. Another projected study was to determine the utility of the dewatered sedimentation tank settled solids as a feedstuff for ruminants. Unfortunately, plans to study these important questions had to be laid aside when it was learned that the needed funding would not be available. The few months remaining until the imposed termination of the project did not allow the time needed for a meaningful exploration of the questions.

Inasmuch as a study on the use of mechanical aeration to supply the oxygen needed for oxidation of the organic wastes in the pond was in progress at the time the second progress report was written, it was decided to continue that study. The successful utilization of artificial aeration would expand the geographical application of the integrated system by providing a means of oxygenation during those seasons when environmental factors were not conducive to algal growth and oxygen production. In addition, the pilot plant studies included two special investigations. They were: 1. a study of inorganic carbon metabolism in the pond as related to the photosynthesis reaction in the pond portion of the system; and 2. a determination of the lytic properties of photosynthetic bacteria in the algae pond. The scope of the overall study was broadened to examine a vegetable solid waste by including a study on the treatability of potato wastes through algal culture. The study was made a part of continuing effort to apply photosynthesis to the treatment of agricultural wastes--

both plant and animal. Potato wastes have the high carbon content characteristic of most cannery wastes and are of special interest in the projected integrated systems for communities.

## PILOT PLANT STUDIES

### Mechanical Aeration of the Pond:

The method of supplying aeration and preliminary results were described in the second progress report (p. 134) as follows:

"Inasmuch as the season (i.e. winter) was patently unfavorable for growing algae in the concentration needed to meet the increased loading, it was decided to try mechanical aeration as a means of supplying the oxygen needed in stabilizing the organic loading. Accordingly, three float-activated 1/3 hp sump pumps were pressed into service to act as aerators by installing a 2-ft vertical pipe into the discharge point of the pump and capping the pipe with a "T" fitting such that the liquid was discharged horizontally into the air and cascaded back into the pond. This closely simulated the type of aeration attained with floating surface aerators.

With continuous operation of 3 aeration pumps, the dissolved oxygen concentration of the pond remained >7 mg/liter. Reducing the number of pumps to two resulted in a small decrease in the average dissolved oxygen concentration to ~6.5 mg/liter. A further reduction to one pump resulted in an average dissolved oxygen value of slightly less than 6 mg/liter. The power cost of an aeration system equivalent to one pump operated continuously on a year-round basis would be about 0.1¢/doz eggs."

The research reported herein was concentrated principally on the solids build-up and the carbon and nitrogen relationships during the time

the pond was mechanically aerated. The time period elapsed during the study was from Feb. 4 to April 11--a period of about 63 days. The time period encompassed the transition from the San Francisco Bay Area winter; slightly frosty, foggy or rainy season to spring.

An important feature of the run was the fact that no water was wasted from the system, excepting that lost through spillage, evaporation, etc., indicating the aerators were effective in enhancing evaporation. The total and suspended solid concentration of the pond culture during the course of the run are plotted in Figure 3. According to the slope of the curves in the figure, the solids content of the pond increased gradually during the run. The exception was the very sharp increase during the last couple of weeks of the run. Judging from the difference between the numerical value of the total solids and those of the suspended solids, the sharp increase in the suspended solids concentration was not accompanied by a comparably sharp rise in total dissolved solids. The variation was within the range of 613 mg to 783 mg/liter or about 21% of the highest concentration of suspended solids. Consequently, the increase in total solids was solely a function of increase in suspended solids concentration.

Reasons for the buildup in suspended solids concentration could have been an increase in bacterial population--or one in algal numbers. The buildup in bacterial population could be expected because of the increased oxygen supply and the heavy concentration of nutrient. The algal buildup would come as an accompaniment of the rising temperature and increase in solar irradiation attendant upon the change in season from winter to spring. An examination of the bottom sludge showed it to have a biota characteristic of conventional activated sludge.

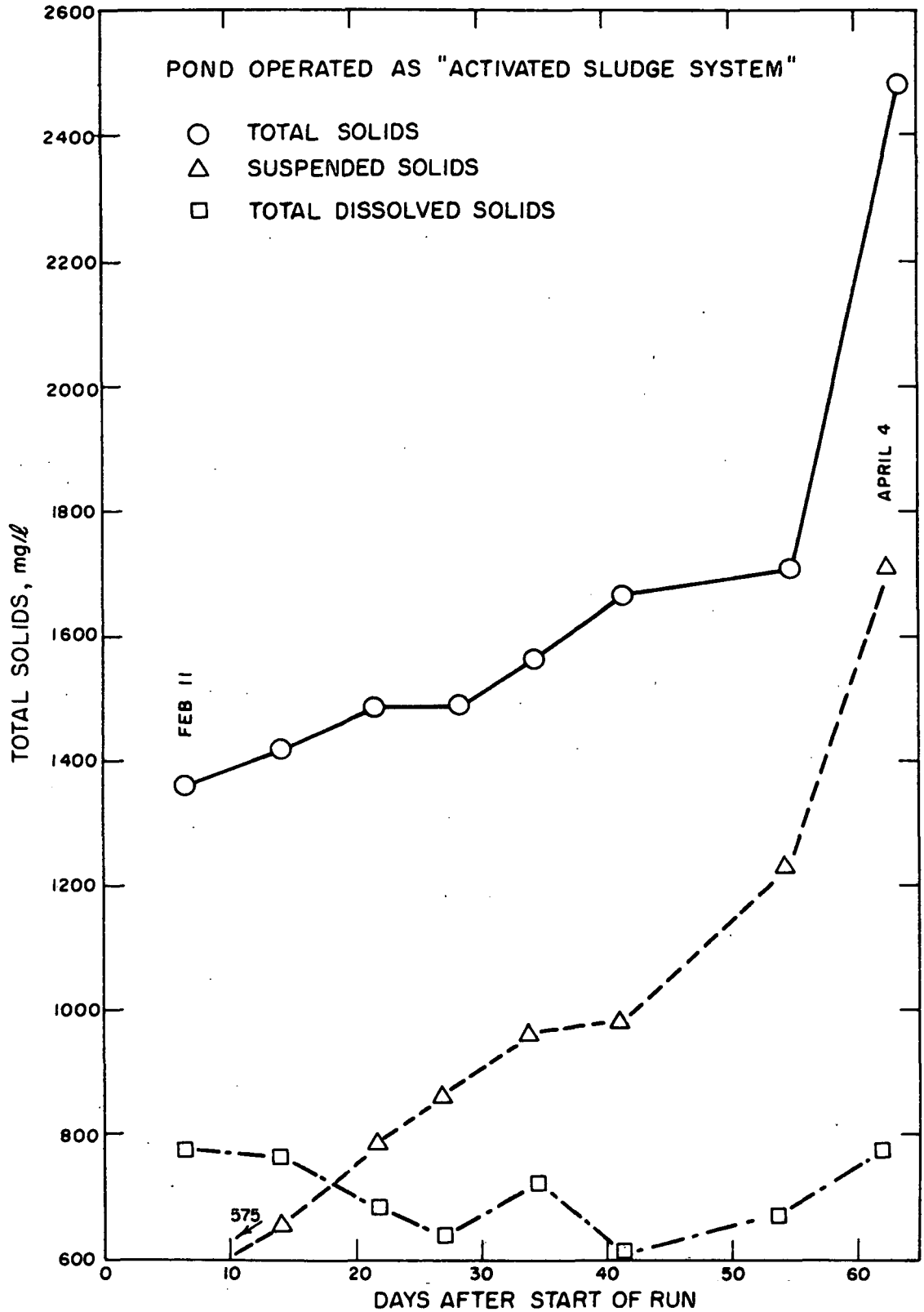



FIGURE 3. TOTAL, SUSPENDED, AND DISSOLVED SOLIDS OF POND CULTURE DURING MECHANICAL AERATION

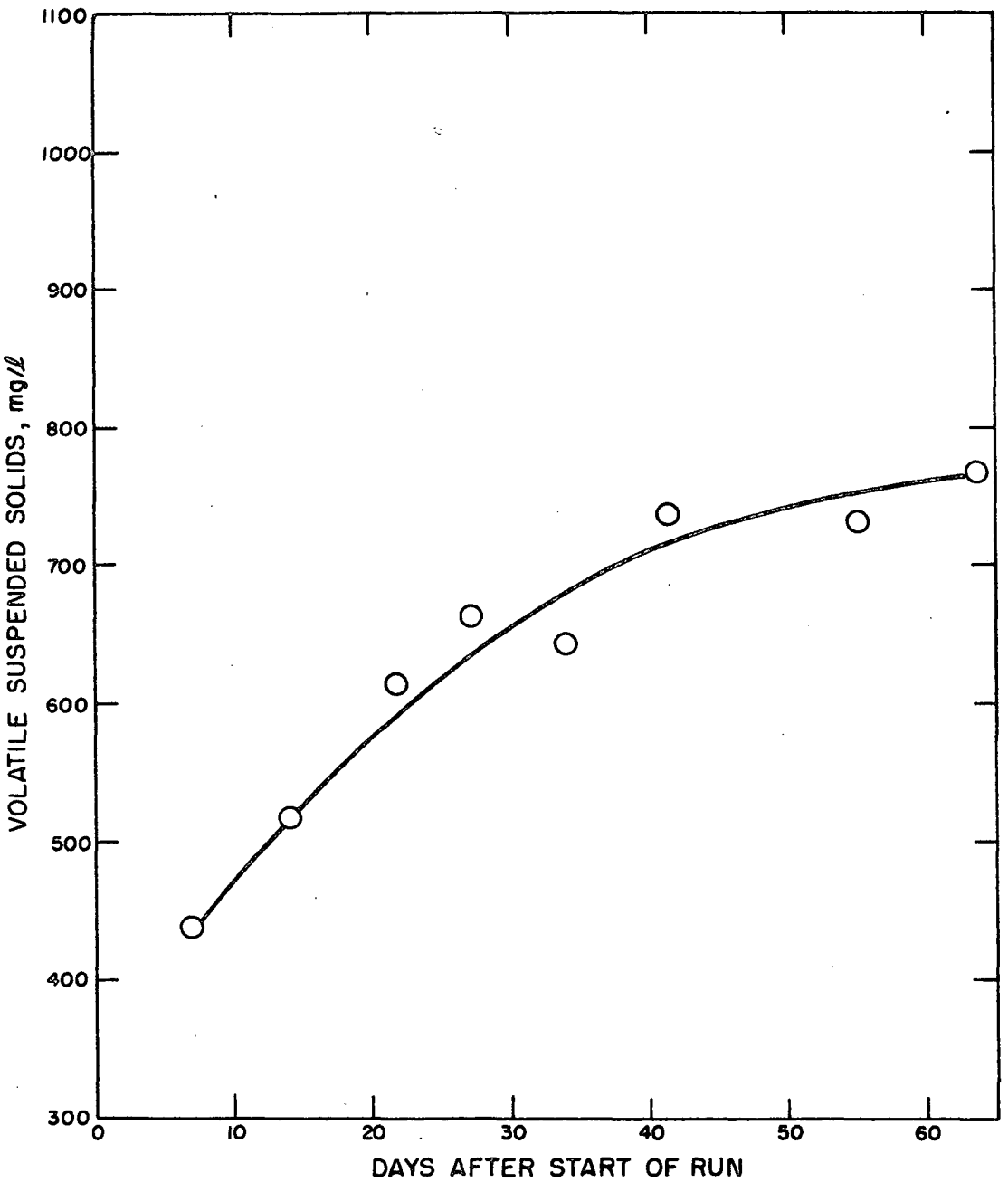



FIGURE 4. CONCENTRATION OF VOLATILE SUSPENDED SOLIDS DURING MECHANICAL AERATION

The final abrupt rise in total and suspended solids cannot be attributed solely to increase in cell mass--algal or bacterial--because the percentage of the suspended solids in a volatile form dropped equally abruptly. Whereas throughout the run, the volatile fraction of the suspended solids ranged between 70 and 79%, on the 63rd day it dropped to only 45%. However, as is shown in Figure 4, the actual concentration of volatile suspended solids rose from about 436 mg/liter to about 767 mg/liter. On the last day of the run (63rd day), it was 767 mg/liter. It is unlikely that the final spurt in total solids is an artifact arising from experimental or analytical error in that the total and suspended solids and nitrogen all showed an abrupt increase at the same time. The phenomenon was probably caused by a burst of algal growth with the coming of spring. Algal growth was followed by a rise in pH and precipitation of soluble complexes accumulated during the long period of aeration.

The slope of the curves for unoxidized nitrogen concentration and $NH_4$-N concentration of the pond culture, as plotted respectively in Figures 5 and 6 fairly closely approximate those for total and suspended solids in the pond. Although the total concentration of unoxidixed nitrogen in the pond rose quite steeply in the last 20 or more days of the run, the percentage of total solids in the form of unoxidized nitrogen dropped sharply. This drop in percentage nitrogen is shown in Figure 7. At the start of the run, unoxidized nitrogen accounted for slightly more than 7.5% of the total solids. This percentage soon dropped to 4 to 5% and remained there until the end of the run. The drop in percentage probably is symptomatic of the shift from algal to bacterial predominance in the biota that began with the application of mechanical aeration.

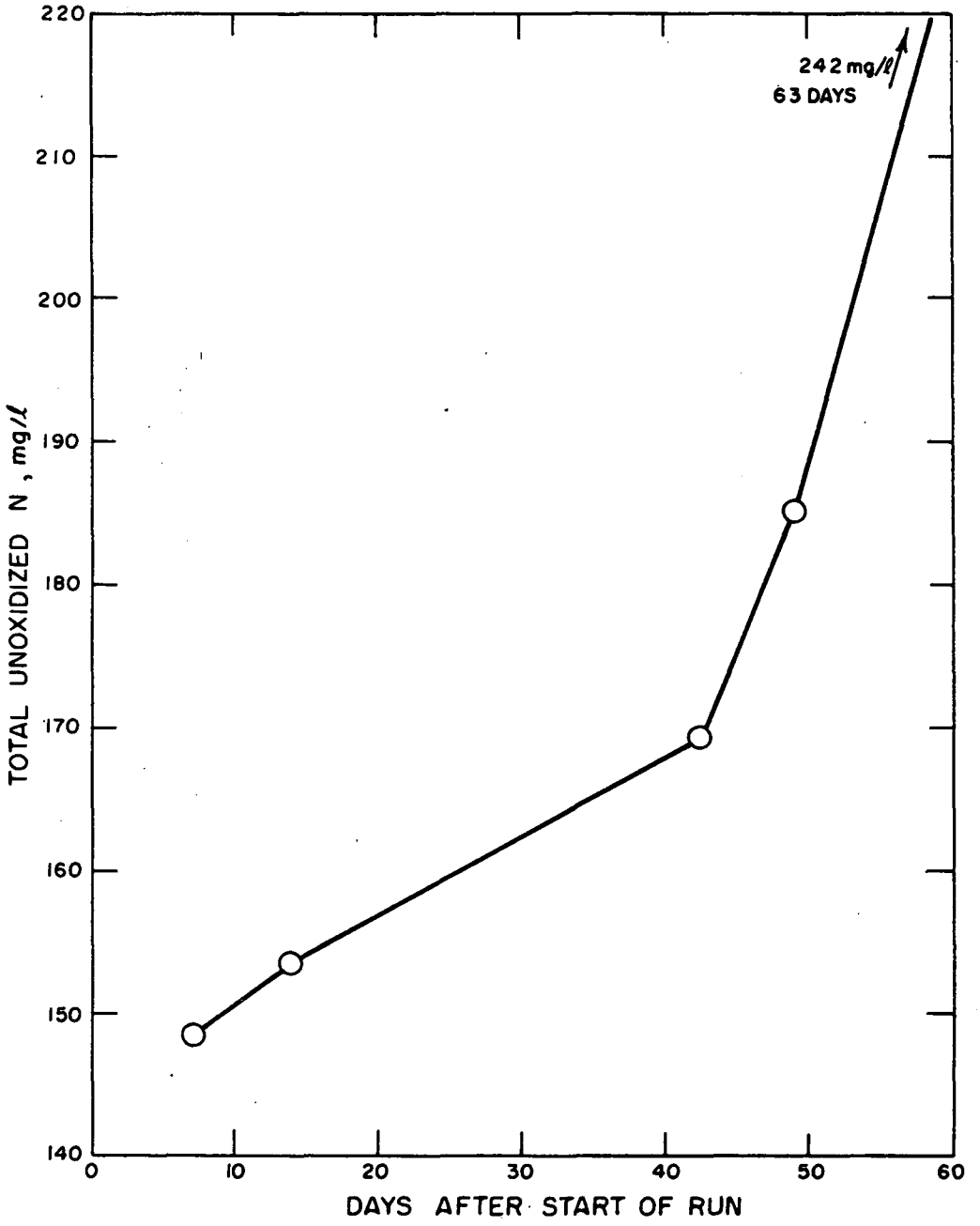



FIGURE 5. THE CONCENTRATION OF UNOXIDIZED NITROGEN IN THE POND DURING MECHANICAL AERATION

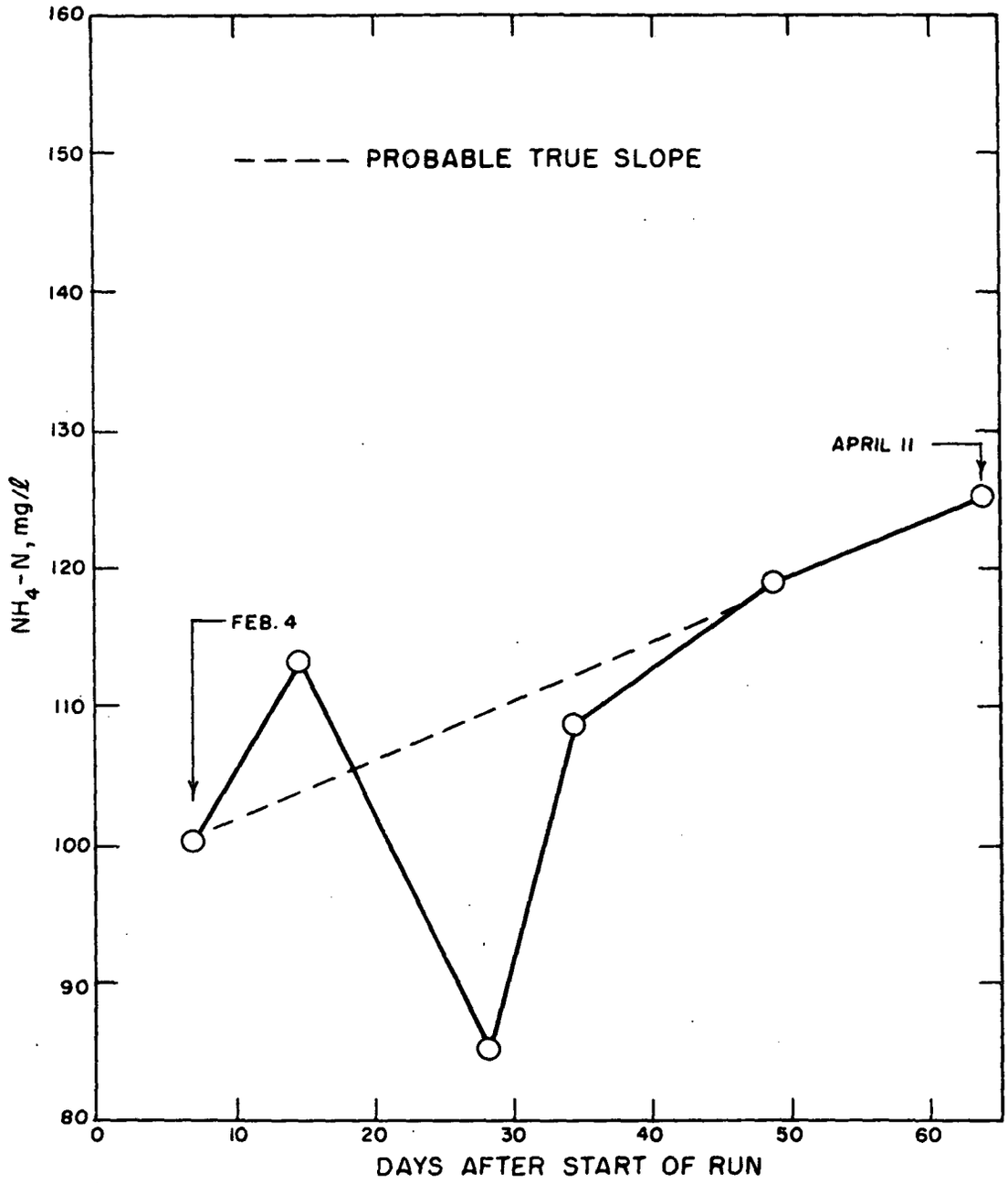



FIGURE 6. CONCENTRATION OF $NH_4$-N IN THE POND DURING MECHANICAL AERATION

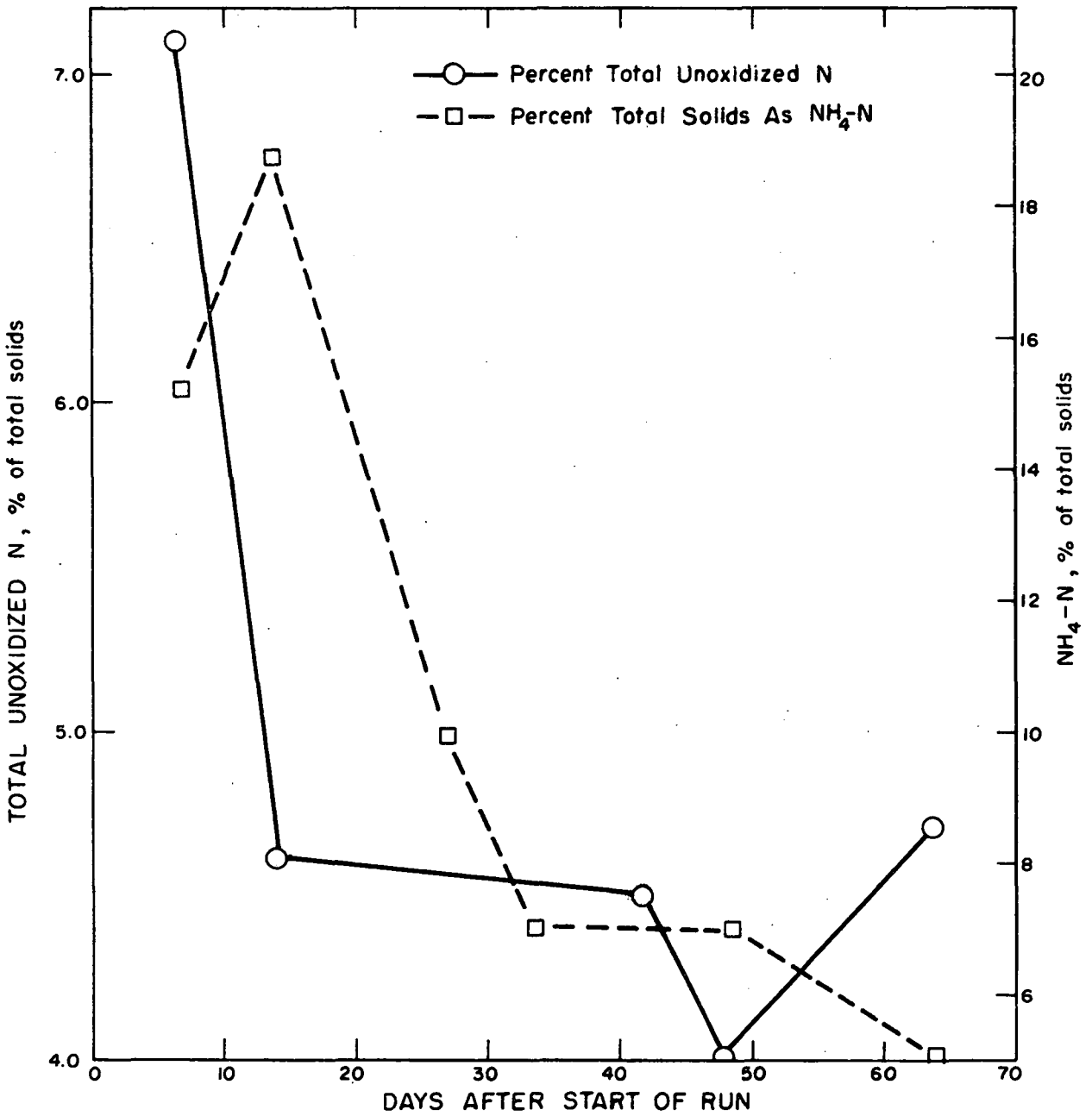



FIGURE 7. PERCENTAGE OF TOTAL SOLIDS IN THE FORM OF UNOXIDIZED NITROGEN AND OF $NH_4$-N

The type of unicellular alga generally encountered in ponds almost always has a nitrogen content of at least 6%. Visual and random observation with a microscope confirmed the decline in number of algae.

Another piece of evidence, or even a symptom of decline in the algal population, was the high concentration of $NH_4$-N which prevailed in the pond. As is shown by the curve in Figure 6, in which is plotted the change in concentration of $NH_4$-N, the $NH_4$-N concentration remained above 100 mg/liter throughout the run excepting for an apparent drop to about 85 mg/liter on one occasion. Inasmuch as a rule, algae preferentially use $NH_4$-N as a nitrogen source, the high $NH_4$-N content would indicate either a sparse or an inactive algal population. Interestingly, the slope of the curve showing percentage of total solids as $NH_4$-N (cf Figure 7) fairly well reflects that for percentage unoxidized-N. The concentration began with a high of 6.8% and ended at 5%. The high ammonia content could--and probably did--have a significant effect on amount and rate of nitrogen loss from the system. The concentration was at a level at which it was toxic for fish. Carp ("goldfish") placed in the pond expired in less than a day.

The percentage carbon content remained quite constant throughout the run, ranging from 34 to 39% of the total solids. The percentage did not change drastically even during the final surge in solids concentration. The fact that the percentage carbon never exceeded 39% would indicate that some of the solids were not organic in nature, since most organic material averages about 50% carbon.

It is unfortunate that the study could not have been extended long enough to determine at what level the buildup of solids would continue, or whether the final sharp surge was a single event or would have

continued. All in all, the pond functioned much as a conventional activated sludge or extended aeration unit during the run. Tests did show that under the constant turbidity prevailing in the pond because of the mechanical aeration, algal growth was greatly inhibited except possibly during the final weeks when spring weather occurred.

Inorganic Carbon as Related to Photosynthesis in the Algae Pond:

Introduction: The purpose of this phase of the study was to ascertain with reference to the algae pond, the relationship of photosynthesis with the diurnal parameters of pH, alkalinity, dissolved oxygen (DO), water temperature, and inorganic carbon uptake; inasmuch as the extent of diurnal variation is an excellent indicator of intensity of photosynthetic activity in an outdoor algal culture. With information gained by measuring the diurnal shift, it should be possible to evaluate the performance of the algae pond. Alkalinity measurements could be used to establish the amount of carbon available to the algae from alkalinity.

To determine the diurnal variation in photosynthetic related factors, ideally a 24-hour period of monitoring is needed. Less ideally, the period of observation could be confined to that time of the day during which photosynthesis occurs. Because of a manpower shortage and the lack of a mechanical sampling system, only one extended observation was made of the diurnal variations in the study discussed herein although several short-term observations were also made.

Methods: The mixing usually applied to the pond was suspended during the time in which the actual studies were made. Although mixing was held in abeyance during the period of observation, some minimal disturbance of the pond contents did take place when the flush water was discharged into the pond each hour, since recycling was maintained at all times.

Alkalinity, pH, temperature and dissolved oxygen were analyzed at hourly intervals immediately after sampling during the extended period of observation of the diurnal shift. Analyses were made according to Standard Methods (1). Inorganic carbon content was determined from a table compiled by Saunders, et al. 1962 (2). Saunders' table is based on equilibrium calculations for the different carbon chemical species. The period of observation was begun at 0500 hours. It was terminated at 2400 hours because the dissolved oxygen content had dropped to zero at 2000 hours and remained at that level thereafter. Moreover, during that same time period (2000-2400 hrs), pH and alkalinity levels also remained almost constant.

Results: According to Table 1 in which are listed data obtained in the run, the solar energy input was at its maximum at 1300 hours. The pond temperature rose from 9.0°C at 0700 hours to 27°C at 1500 hours. Judging from the solar energy level and the high pond temperature, the day was an excellent one as far as algal growth was concerned. The period of highest pH level (pH 10.85), came about two hours after the peak of the solar energy input had been reached. The level of pH at 0500 hours was 7.82; and at 2400 hours, 8.40. As would be expected, the high pH levels were accompanied by high phenolphthalein alkalinities, namely, 160 and 165 mg/liter as $CaCO_3$. Maximum variation in total alkalinity was only a matter of 65 mg/liter as $CaCO_3$. The lowest level of total alkalinity, 268 mg/liter as $CaCO_3$, occurred at 1500 hours; and the highest, 333 mg/liter, at 0500 hours. The concentration of dissolved oxygen was greatest (37.5 mg/liter) at 1100 hours. On the other hand, it was zero from 0500 and 0600 hours, and from 2100 hours to 2400 hours.

At pH levels from 5.0 to 9.4 the inorganic carbon uptake was a

TABLE 1. DIURNAL CHANGES IN pH, ALKALINITY, DISSOLVED OXYGEN, AND INORGANIC CARBON UPTAKE[a]

| Time (hr) | Solar[b] (amp-hr) | Temperature | | pH | Alkalinity[c] | | Dissolved Oxygen (mg/1) | Calculated Inorganic Carbon (mg/1) | Carbon (mg/1) | Remarks |
|---|---|---|---|---|---|---|---|---|---|---|
| | | Ambient (°C) | Pond (°C) | | Phenol-phthalein (mg/1) | Total (mg/1) | | | | |
| 0500 | Base | 6.0 | 9.0 | 7.82 | 0 | 333 | 0.00 | 83.3 | Base | Mild north wind |
| 0600 | 0.00 | 6.0 | 9.0 | 8.07 | 0 | 328 | 0.00 | 78.7 | - 4.6 | Mild north wind |
| 0700 | 0.03 | 8.5 | 9.0 | 8.07 | 0 | 330 | 0.95 | 79.2 | + 0.5 | Mild north wind |
| 0800 | 0.11 | 10.5 | 11.0 | 9.27 | 60 | 315 | 11.55 | 72.5 | - 6.8 | Mild north wind |
| 0900 | 0.18 | 12.0 | 13.0 | 9.10 | 48 | 298 | 10.25 | 68.5 | - 4.0 | Mild north wind |
| 1000 | 0.34 | 18.0 | 19.5 | 9.57 | 75 | 288 | 20.50 | 51.1[d] | -17.4 | Calm |
| 1100 | 0.32 | 19.0 | 23.0 | 9.80 | 98 | 278 | 37.50 | 42.2[d] | - 7.9 | Calm |
| 1200 | 0.33 | 18.0 | 18.0 | 9.04 | 45 | 300 | 12.80 | 66.0 | +22.8 | Moderate west wind |
| 1300 | 0.43 | 19.0 | 22.0 | 9.59 | 80 | 290 | 22.30 | 50.4[d] | -15.6 | Calm |
| 1400 | 0.34 | 21.0 | 24.0 | 9.88 | 95 | 278 | 33.10 | 43.9[d] | - 6.5 | Calm |
| 1500 | 0.29 | 20.0 | 27.0 | 10.85 | 160 | 268 | 33.20 | 25.9[d] | -18.0 | Mild west wind |
| 1600 | 0.25 | 20.0 | 26.0 | 10.85 | 165 | 273 | 31.90 | 25.9[d] | 0.0 | Mild west wind |
| 1700 | 0.20 | 19.0 | 22.0 | 9.21 | 58 | 298 | 4.10 | 68.5 | +42.6 | Mild west wind |
| 1800 | 0.08 | 16.0 | 20.0 | --- | -- | 300 | 2.25 | 72.0 | + 3.5 | Mild west wind |
| 1900 | 0.00 | 14.0 | 18.5 | 8.95 | 28 | 285 | --- | 65.6 | - 6.4 | Mild west wind |
| 2000 | 0.00 | 12.2 | 17.5 | 8.74 | 18 | 290 | 2.60 | 69.6 | + 4.0 | Mild west wind |
| 2100 | 0.00 | 12.0 | 17.0 | 8.52 | 8 | 295 | 0.00 | 70.8 | + 1.2 | Mild west wind |
| 2200 | 0.00 | 12.8 | 16.0 | 8.49 | 8 | 303 | 0.00 | 72.7 | + 1.9 | Mild west wind |
| 2300 | 0.00 | 12.5 | 15.0 | 8.45 | 3 | 295 | 0.00 | 70.8 | - 1.9 | Mild west wind |
| 2400 | 0.00 | 12.0 | 14.5 | 8.40 | 3 | 308 | 0.00 | 73.9 | + 3.1 | Mild west wind |

[a]Run made on April 25, 1969.

[b]Solar meter calibrated for 1.0 amp-hr to equal about 0.190 k-cal/cm$^2$ hr.

[c]Phenolphthalein alkalinity titrated to pH 8.3. Total alkalinity to pH 4.3.

[d]Determined as follows: Inorganic carbon = 0.24 (total alkalinity-phenolphthalein alkalinity) when the pH level is above 9.4.

function of temperature and total alkalinity. At pH levels above 9.4, the inorganic carbon uptake was simply the difference between the total and the phenolphthalein alkalinities multiplied by 0.24 (Saunders, et al. 1962). Although these calculations are dependent on the assumption that equilibria exist in a system having relatively high concentrations of only $CO_2$, $HCO_3^-$, $CO_3^=$, and $OH^-$, this assumption would not lead to erroneous conclusions because the conclusions are based on relative values or on differences, thus cancelling out any errors with respect to inorganic carbon.

In arriving at the values given in Table 1, it was assumed that there was no interchange of $O_2$ and $CO_2$ between the pond and the atmosphere and this appeared quite probable because of the persistence of a noticeable but thin scum observed on the surface of the pond which would probably impede significant gas transfer unless the scum were disturbed. This assumption appeared reasonable because the great decreases in dissolved oxygen levels were observed only after a wind had occurred and disturbed the water surface.

Discussion: The interrelationships between the photosynthesis-related phenomena are shown in Figure 8, in which values for the different variables are plotted as functions of time of day. The interrelationships are demonstrated by the simultaneity of the sharp responses of the four variables which took place at 0700 hours, again at 1200 hours, and finally at 1600 hours. The nature of the response in terms of inorganic carbon was the inverse of those for pH, phenolphthalein alkalinity, and dissolved oxygen.

The initial rise in level of response for pH, alkalinity, and DO, and the dip in inorganic carbon corresponded with the beginning of

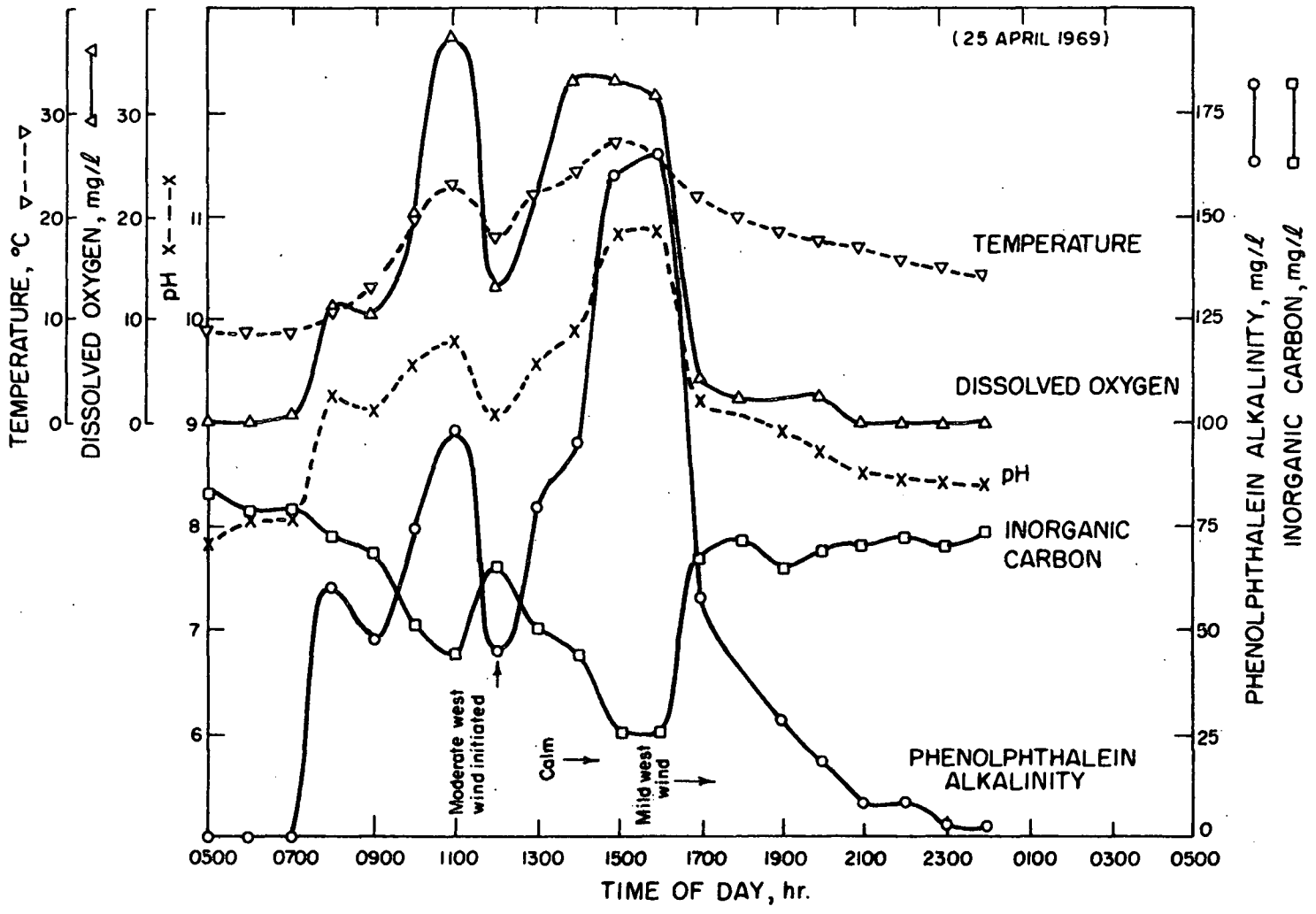



FIGURE 8. INTERRELATIONSHIP BETWEEN PHOTOSYNTHESIS AND RELATED PHENOMENA

the daylight period, i.e., the beginning of the input of solar energy. Thus, as would be expected, all of the responses mark the beginning of photosynthetic activity in the pond. This relationship between the four parameters and photosynthetic activity continued throughout the daylight period.

The dip in pond temperature as well as in the levels of pH, phenolphthalein alkalinity, and DO which took place at 1200 hours coincided with a brief period of moderate west wind. The sharpness of the response of the three parameters seems to have been out of proportion to the extent of the temperature drop. Perhaps the temperature-related effects may have been enhanced by those due to wind action. For example, the rate of $O_2$ loss to the atmosphere may have been increased, inasmuch as the pond culture was supersaturated with $O_2$ at the time. Similarly, some $CO_2$ may also have been lost.

The abruptness of the change in all four parameters which took place at 1600 to 1700 hours was out of proportion to the drop in pond temperature and to the amount of solar energy available to the pond culture at the time, if the two factors are taken <u>individually</u>. On the other hand, if temperature and solar energy are considered together and added to the effects from wind action which took place at that time, then the responses would have been cumulative and hence abrupt in their expression.

<u>Inorganic Carbon and Dissolved Oxygen Responses:</u> This analysis is based upon the classical formula for photosynthesis, namely:

$$CO_2 + 2H_2O \xrightarrow{\text{energy}} (CH_2O)_x + O_2 + H_2O$$

The quantity of oxygen produced during the period of observation, as determined by integrating the area under the DO curve from Figure 8

amounted to 235.6 mg/l-day. Accordingly, the total amount produced by the entire algae pond culture for the day was 2.85 kg of $O_2$ (41.6 g $O_2/m^2 \cdot$ day). The inorganic carbon uptake was considered to be the summation of the negative values given in the $\Delta$ carbon column in Table 1. Calculated on this basis, the inorganic uptake was 89.0 mg/l as C or 326.3 mg/liter as $CO_2$ which equals 3.95 kg of $CO_2$ (57.7 g $CO_2/m^2 \cdot$ day) for the entire algae pond culture.

According to the formula for photosynthesis, for every gram of $CO_2$ reduced in the photosynthetic process, 1.375 g of $O_2$ are produced. If this be true, the total $CO_2$ uptake by the pond during the day should have been 3.92 kg (1.375 x 2.85 kg of $CO_2$). Thus, the difference between the observed amount (3.95 kg) and the expected theoretical value (3.92 kg) was only 0.03 kg, or less than 1 percent.

The small difference between the observed and the theoretical values does not necessarily mean that the observed value reflected reality equally closely. Two possible sources of error, other than experimental and mathematical, may have influenced results. The first could have resulted from making the assumption that no net loss or gain in $CO_2$ or $O_2$ took place between the pond culture and the atmosphere. Perhaps there may have been a loss or gain. A second source could stem from the assumption that the $CO_2$ uptake was only the summation of the negative values for inorganic carbon. Some $CO_2$ uptake undoubtedly occurred during the hours when the net change in inorganic carbon may have had a positive value. Nevertheless, the extent of the error, if any, introduced from these sources must be small, since a high degree of fortunate coincidence would have to have been involved in arriving at a set of observed values so closely identical with the theoretical values.

The correlation between theoretical and measured carbon utilization in the system was not unexpected (e.g., Odum - References 3 and 4). Similar cycles have been described by Bartsch and Allum (5). Not only are relatively copious amounts of carbon available for algal photosynthesis from carbon sources in alkalinity but dissolved oxygen can be accumulated to relatively high concentrations (>30 mg/l) without inhibiting the photosynthetic reactions.

Although this method of determining respiration-photosynthesis may not be simply applicable to lakes or larger ponds, by dealing with entire systems it has considerable value over previous methods based on enclosed polyethylene bags (e.g., Goldman, Ref. 6) or open cylinders (Kemmerer and Newhold, Ref. 7). In the system described herein, planktonic and benthic production are measured and wall interference caused by confinement are avoided. Measurements are simple and easy to make and diurnal measurements can be replicated as necessary to provide valid statistical analysis. Also, in a pond system such as this, the production rates are quite high, a factor of 100 greater than for a eutrophic lake and changes are more easily measured. However, more attention should be devoted to gas diffusion effects, even though they probably are minor in relative magnitude in an algae pond. (In lakes, diffusion would be extremely important [e.g., Hutchinson, Ref. 8].) Thus, it is a valuable method for analyzing the balanced operation of a pond system in waste treatment.

<u>Summary and Conclusions</u>: 1) Agreement between the theoretical carbon change and measured carbon change in the algae pond was within 1 percent. Oxygen production was calculated to be 41.6 g $O_2/m^2 \cdot$ day and carbon dioxide uptake as 57.7 g $CO_2/m^2 \cdot$ day. 2) Sufficient carbon was avail-

able for algal growth, almost all of which was derived from carbon chemical species measured in alkalinity. 3) The rather simple measurement of alkalinity, pH, temperature, and dissolved oxygen provided information, which could be of considerable value in interpreting the operation of algae ponds.

Lytic Properties of Photosynthetic Bacteria in the Algae Pond

Introduction: Considerable research has been focused on the role of algae in natural and artificial ecosystems where large quantities of organic compounds are decomposed. In contrast, the involvement of photosynthetic bacteria in heterotrophic environments is poorly understood. Cooper and others (9, 10) have noted that ponds receiving domestic, industrial or agricultural wastes frequently develop conspicuous populations of photosynthetic bacteria. Other reports attest to the ubiquity of such bacteria in eutrophic environments.

During the course of observations on the algae pond, the characteristic pink color of photosynthetic bacteria was noted in samples of algae harvested from the pond. Several species were isolated from the pond, and their physiological properties were examined in the laboratory. Herein are summarized the results of studies concerned with lytic properties of facultative heterotrophic photosynthetic bacteria that relate to the use of algae as a protein supplement.

Materials and Methods: For laboratory studies, samples of the pilot chicken waste pond material were collected from several sites in the hydraulic transport system, from the sides of the pond, and from algae harvested by centrifugation from the pilot plant. These samples served as inoculum for preparing enrichment cultures and for direct plating experiments. The media, general procedures and conditions for establishing

enrichment cultures of photosynthetic bacteria have been described by van Niel (11).

Pure cultures were obtained by means of the shake-tube technique previously described (12). In experiments concerned with lysin production, the bacterial strains were streaked on sheep-blood agar and incubated under either anaerobic conditions using the gaspak technique, or aerobic conditions. Plates were stored at room temperature in the light unless otherwise indicated.

Results: Enrichment cultures incubated under anaerobic conditions with incandescent lamps as a source of illumination developed turbidity and pigmentation characteristic of the non-sulfur photosynthetic bacteria (Athiorhodaceae) within 3-5 days. Although no experiments were undertaken to specifically determine the numbers of photosynthetic bacteria in the algae pond at the time of sampling, from serial dilution of shake-tube cultures it is possible to make an estimate of approximately $10^6$-$10^7$/ml. Using the shake-tube method, several strains of Rhodopseudomonas and Rhodspirillum were established in pure culture.

Experiments with sheep-blood agar plates streaked with inoculum taken directly from pond samples prior to establishing enrichment cultures yielded large numbers of colonies ringed with a clear colorless zone. When such cleared areas were examined under the microscope, no red blood cells were detected. These results suggested that microorganisms in the algae pond were producing a diffusible agent capable of lysing red blood cells. The lytic reaction was characteristic of BETA-hemolysis.

Pure cultures of Rhodopseudomonas and Rhodospirillum isolated from the algae pond were tested for hemolytic activity on sheep-blood

agar plates. All strains tested were found to exhibit hemolytic activity when cells were grown under aerobic conditions. No detectable activity was observed when growth occurred under anaerobic conditions. These results provide evidence that lysis of red blood cells is strictly dependent upon the presence of oxygen.

In subsequent studies, strains of Rhodopseudomonas and Rhodospirillum isolated from the algae pond were compared with reference strains obtained from the University of California Department of Bacteriology culture collection. These experiments served to confirm earlier findings that hemolytic activity is a common feature of the facultative strains of Rhodoseudomonas and Rhodospirillum, and that the production of the lytic agent requires molecular oxygen.

Other experiments were performed to obtain additional information concerning the process of lysin production, and to study some properties of the lysin. The results can be summarized as follows:

1. Using sheep-blood agar plates to assay for hemolytic activity, it was noted that the lytic reaction was detectable after two days at 30°C and reached a maximum between three to four days after incubation.

2. When plates containing fully grown colonies of photosynthetic bacteria were shifted from anaerobic to aerobic conditions, a lag was detected in the appearance of lytic activity. These observations are consistent with the hypothesis that synthesis of the lysin is inducible by oxygen. The lag period that precedes the detection of lytic activity argues against the idea that the lytic agent preexists in the bacterial cells and requires oxygen for activation.

3. The regulation of lysin synthesis is coordinated in a negative fashion with the formation of photosynthetic pigments such as

bacteriochlorophyll and spirillaxanthin. Oxygen appears to serve two functions in regulation: a) As a corepressor to induce transcription of a specific messenger RNA coding for lysin synthesis; and 2) As a repressor of photosynthetic pigment synthesis.

4. On the basis of dialysis experiments, a rough estimate of the molecular weight of the lysin has been obtained. The lysin appears to be of the order of 40,000 M.W.; it is apparently excreted from the bacterial cells into the surrounding medium, and is readily diffusible in agar.

Discussion: The discovery that non-sulfur photosynthetic bacteria such as *Rhodopseudomonas* and *Rhodospirillum* produce a lytic agent capable of destroying red blood cells raises a number of questions concerning the ecological importance of such lysins in eutrophic environments. In this paper, the discussion is limited to the significance of this finding for algae production in photosynthetic reclamation systems.

If the properties of the lysin produced by photosynthetic bacteria are compared with other bacterial lysins, it becomes apparent they have much in common. The lytic agent produced by *Rhodopseudomonas* or *Rhodospirillum* is a high molecular weight compound that is excreted into the medium--non-dialyzable but diffusible in agar. The hemolytic exotoxins of certain Streptococci and Clostridia are soluble enzymes that hydrolize phospholipids of cell membranes. Such enzymes are capable of lysing other types of cells besides red blood cells.

The inducibility of lysin synthesis by oxygen represents the first case of a non-constitutitive lytic factor. Either the inducibility of lysins in other bacteria has been overlooked, or lysin synthesis in non-photosynthetic bacteria is normally constituitive.

Approaches to Inactivation and Control of Lytic Activity: If the large molecular weight compound is a protein, then lytic activity should be abolished by thermal denaturation. Although we have not as yet determined thermal decay kinetics for the lysin, preliminary experiments indicate that virtually all activity is destroyed within 5 minutes at $100^{\circ}$ C. Accordingly, one solution to the problem of lysins in algae to be used as a protein supplement in animal feed is to pasteurize the algal product. Alternatively, there may be a decay, albeit less rapid, of lytic activity during storage at ambient temperatures.

The potential contamination of algae by hemolytic photosynthetic bacteria poses the question: Do hemolytic strains represent possible pathogens for man or domestic animals? Again, if it is assumed that the lysin is a protein, then it is likely that it will be degraded by proteolytic enzymes in the digestive tract.

It seems important to keep in mind that photosynthetic bacteria are not the only potentially hemolytic microorganisms in algae ponds. It is highly likely that a large number of non-photosynthetic bacteria are also present in such ponds, and that a fraction of these may also produce lysins.

One way of looking at the problem of photosynthetic bacteria in algae ponds is to consider the pond as an enrichment culture. The high concentrations of organic hydrogen donors and ammonium nitrogen favors the growth of photosynthetic bacteria over other microorganisms in the pond, bacterial photosynthesis competes with algal photosynthesis for $CO_2$; finally, the growth rate for photosynthetic bacteria is probably two or three fold more rapid than that of most algae.

Cooper (9) has noted that the population of photosynthetic

bacteria in waste treatment ponds seems to depend upon the organic loading rate. At high loading rates, the ponds become anaerobic and the photosynthetic bacteria become conspicuous; at low loading rates, the algae are favored, the pond becomes aerobic, and the photosynthetic bacteria are suppressed. Cooper has commented that regardless of load rate, the ponds that he studied functioned well. These observations suggest that the photosynthetic bacterial population in algae ponds can be controlled by maintaining the loading rate below the level that favors the enrichment of these bacteria over the algae.

Summary: Non-sulfur photosynthetic bacteria were isolated from the algae pond. When pure cultures were isolated, and tested on sheep-blood agar plates, all strains of *Rhodopseudomonas* and *Rhodospirillum* were capable of lysing red blood cells. The lytic reaction is produced only in the presence of oxygen. Such lytic activity may have a bearing on the feed value of recycled algae from the system.

## Discussion of Pilot Plant Studies:

Design Considerations: Pond area required per bird is a determining factor with respect to the economic feasibility of incorporating an algae production pond into a system of the type described in the study reported herein. In the experiments, a pond surface area of 7 sq ft was originally alloted per bird. However, the loading to the pond resulting from this large surface area allotment proved to be far less than the capacity of the pond to treat the wastes. Through successive partitioning of the pond, it was found that a surface area of $\simeq$ 2 sq ft/bird was as adequate as 7 sq ft per bird. As stated in the opening paragraphs of this report, the project unfortunately had to be terminated before the minimum permissible area per bird could be determined. However,

the closer the minimum surface area is approached, the less the safety factor becomes in terms of satisfactory waste treatment. There is the possibility, or even likelihood, that during seasons of the year unfavorable to algae growth, the 2 sq ft/bird may be less than adequate. However, as the studies showed, the deficiency can be easily met by resorting to a simple form of mechanical aeration. An economic analysis would have to be made to determine the trade-off point at which cost of land area balances cost of purchase and operation of equipment for mechanical aeration. All things considered, the 2 sq ft of pond area/bird dimension probably will prove to be generally applicable.

Water requirements for flushing need not be the limiting factor in reducing areal requirements, since only about two-thirds of a gallon (about 2.5 liters) of water per chicken per day is required to clean the troughs and provide a 1% (wet weight) manure slurry. This water could be supplied to the system by way of overflow from the chickens' drinking water troughs.

Roof Ponds: A portion of the land area required for the algae pond could be reduced by constructing an algae pond on the roof of the poultry enclosure. (By coincidence in the study reported herein, the surface area of the partitioned (i.e. active) portion of the pond proved to be about the same as the floor area of the poultry enclosure.) The design load of the substructure of the poultry enclosure would have to be increased to accommodate the added weight load of the pond and its contents (62.4 lb/sq ft-12 in water), as well as the impulse loads attending the recirculation of the algal culture. Moreover, the roof also would have to be curbed and waterproofed to a greater degree than is practiced in normal construction. However, the increased design loading would be

no greater than that generally made for snow and wind in many parts of the U.S. The head provided by the elevated culture would permit flushing of the manure by simple systems, as for example electrically activated valves connected to timers. The pond would serve as an insulation for the poultry enclosure, thus providing some degree of cooling in the summer and reducing heat loss during the winter. Heat lost into the culture from the poultry enclosure during winter would raise the temperature of the pond culture somewhat, and thus the efficiency of the culture. Heat exchange between the enclosure and the pond might be sufficient to diminish or eliminate freezing in areas where sustained freezing normally occurs.

Harvesting Algae: There is no need for an extensive dissertation on the uses of harvested algae in this report, since the use of algae as a feedstuff is well documented in the literature. According to Grau and Klein (13) and Hintz and Heitman (14), algae in either the dried state or as a dewatered paste can be used with good results as a protein supplement in the diet of chickens, ruminants, and swine. If a market for the algae as a feedstuff should fail to develop, the product could be used as a high-grade fertilizer. If used as a fertilizer, the algae could be pumped to the fields directly after the initial removal, thereby eliminating the cost of dewatering and drying steps. Yet another fate for the harvested algae could be digestion in the enclosed digester. Regardless of the ultimate use of algae removed from the pond, some harvesting will probably have to be done in a practical operation to provide a detention period for the algal population. Experience with algal cultures, as well as with any microbial culture, has shown that once an optimum population density of the organism in question is

exceeded, the activity of the population as a whole declines quite rapidly. Consequently to maintain the pond at its peak oxygen production efficiency, some algae would have to be harvested.

Neither the need for algae removal nor the amounts needed to be removed were fully explored in the present study. Obviously, the amount to be removed would depend upon the rate of algal growth. Hence, more would have to be harvested in the summer than in the winter. The study showed that algae could be removed from the pond culture either by centrifugation or by natural settling. The slurry resulting from either method of separation could be easily air dried. The authors estimate that in a large-scale operation, dried sewage-grown algae could be produced for about $0.04/lb. Feeding the algae as a wet paste, i.e. directly after the dewatering step, would lower the cost of production by an estimated 3¢/lb.

If the algae product is to be used as a fertilizer, then undoubtedly separation by natural settling would be indicated. Nonalgal solids also could be removed in this manner by operating the pond mixing pumps while the pond effluent is being discharged to the harvesting settling tank. An alternative to the use of a separate settling tank for harvesting algae through natural settling would be to construct in the algae pond a narrow, deep pit having a cross-sectional design comparable to that of an Imhoff tank. Algal and nonalgal solids settled in this pit could be periodically or continuously removed and pumped directly to the digester, or to the fields, or to dewatering and drying units. The settling tank as well as the anaerobic digester could be located below the chicken house, resulting in a saving of land.

Practical Scale Systems: Findings made thus far are sufficiently

advanced to make it possible to directly adapt portions of the pilot plant system to a large-scale egg-laying operation without undue difficulty. The manure troughs and flushing units could be easily formed of sheet metal and be fabricated into a conventional poultry battery arrangement. The manure trough scheme would be especially suitable in a multi-storied operation, in which at present a barrier has to be interposed between each story of poultry batteries to shield the birds in the lower cages from the droppings of those in the upper cages. The manure trough would serve that function.

The sedimentation tank could be a conventional open tank. A substantial part of the required culture volume could be ponded on the roof of the poultry shelter. Depending upon the market, algae either could be harvested by flocculation (or centrifugation) or by simple settling.

If the system is to include an anaerobic phase, this phase could consist either of a closed digester or of an anaerobic lagoon system. The utilization of an enclosed heated digester probably would be justified only in an extremely large enterprise, or where land use must be kept at an irreducible minimum because of high land costs, or where even a low level of odor emanation would not be tolerated. Although the gas wasted from the digester in the experiments had some odor, odor problems would not plague a practical operation because the waste gases would be combusted. In cases where a digester would not be warranted, an anaerobic or facultative lagoon might be used to receive the settled solids from the sedimentation tank. This would be especially attractive in situations in which the solids could be discharged into irrigation pipes during the crop-growing season. The settled solid could also be

dewatered, dried, and used as a low-protein feedstuff for ruminants.

One might feel that even in operations somewhat smaller than the type named above, the use of a closed digester would be justified if the gas were burned and the heat used to heat the digester and to dry algae. The applicability of this assumption would depend on the market value of combustible gases, i.e. would the savings in gas purchase outweigh the expenses involved in constructing and operating the digester.

Outflow from the sump (i.e. discharge to the environment) can be set at any desired level, even to the point of closing the system. Should an extensive prolongation of the operation of the integrated system as a closed system eventually prove to be unfeasible, even then effluent discharged to the environment would require no further treatment. The reason is that the sump outflow would be passed through the algae harvesting subcomponent of the system, with the result that algae would be removed, and the effluent, now devoid of algae and most of the nutrients conducive to algal regrowth, would have received the equivalent of secondary and tertiary treatment.

Economics: While it would be somewhat presumptious to attribute a high degree of accuracy to projections made at this time on the economics of the process, one should be made so that a basis for a judgment can be had with respect to the order of magnitude of the economic feasibility of such a system. Moreover, by being aware of the factors which limit the firmness of the analysis, one can arrive at a reliable conclusion as to the degree of the accuracy of the analysis. One factor which would detract from the firmness of an economic analysis at this time is the fact that the present study had not progressed far enough to supply the firm data needed in making an analysis. Insufficiently known variables

are the actual pond area requirements, the actual market value of the algal product, the food and market value of manure processed for use as a feedstuff for other animals, and certain of the operating conditions. Other variables, unknown for specific situations, but for which ranges of values can be set up are climatological factors (temperature, solar radiation, wind, rainfall, etc.), land use, and costs, market value of the products other than algae and processed manure (e.g. eggs, fryers, etc.), economics of manure use, disposal, labor, and equipment costs.

An analysis made as a part of this study for a 100,000-bird operation is summarized in the information given in Table 2. The analysis was made in order to arrive at the maximum practical cost of a system which would include a heated digester, hard-surfaced algae pond, and algae-removal equipment. If desired, the digester could be replaced by a facultative pond. If this were done, no hard surface would be applied to the facultative portion. Algae removal could be by settling rather than by centrifugation.

The poultry enclosure additions under the heading "Construction and Equipment Costs" in Table 2 include those required for the hydraulic system, namely, manure troughs, flushing mechanisms, etc. The incorporation of the manure troughs, flushing mechanisms, etc., into the chicken battery assemblies at the time of fabrication would undoubtedly save a considerable portion of the $5,000 allotted for these items. The capacity of the sedimentation tank was designed for a 2-hour detention time. For practical purposes two tanks should be used, so that one can be by-passed for maintenance. The estimate was based on two rectangular shaped reinforced concrete tanks with sloping bottoms. Sludge and supernatant pumps, as well as the necessary plumbing were included in the estimate.

TABLE 2

Estimated Capital and Maintenance Costs of the Waste Handling Facilities of a 100,000 Egg-Laying Poultry Operation Utilizing Photosynthetic Reclamation and Hydraulic Transport

| Construction and Equipment Costs: | Estimated Costs[a] |
|---|---|
| Poultry Enclosure Additions | $ 5,000 |
| Sedimentation Tank + Pumps, etc | 5,000 |
| Digester(s) + Appurtenances | 50,000 |
| Algae Pond (including land, pumps, etc.) | 100,000 |
| Algae Removal Equipment (Batch Centrifuge and Drying Facilities) | 10,000 |
| TOTAL | $170,000 |
| **Fixed Costs:** | |
| Depreciation (10 yr - straight line) | $ 17,000 |
| Working Capital - 7% | 11,900 |
| TOTAL | $ 28,900 |
| **Utilities and Maintenance:** | |
| Utilities and Repair of Equipment | $ 5,000 |
| Additional Labor | 5,000 |
| TOTAL | $ 10,000 |
| Total Estimated Annual Cost | $ 38,900 |
| Total Estimated Cost per Chicken per Year | .39 |
| Total Estimated Cost per Dozen Eggs | .02 |

[a]"1970-dollars"

The digester cost was based on unit prices developed by Taiganides et al. (15) for animal manures with the capacity based on the approximate organic loading of 0.09 lb VS/cu ft. The design utilized for developing the unit prices was based on the fairly sophisticated type of digester typically found in municipal sewage treatment plants. It was assumed that the digester gas would be utilized as the fuel for heating the digester contents.

Although it is not considered likely that expensive surfacing would be required, nevertheless the cost of a hard-surfaced algae pond (i.e. asphalt, gunite, etc.) including medium-priced agricultural and mixing equipment, transfer pumps, etc., is estimated to be $10,000/acre. At the pilot plant loading based on approximately 7 sq ft/chicken, 16 acres would be required. However, since the performance of the pilot plant pond indicated that the area required per chicken could be safely reduced to 2 sq ft, it would be excessively conservative for evaluation purposes to extrapolate from the full 7 sq ft/chicken. On the basis of 2 sq ft/bird, the pond area would be 5 acres. Therefore, in making the evaluation, the range of indicated areas was taken into consideration and the safe operational pond area was assumed to be 10 acres (4.36 sq ft/bird). The unit price for the pond area would be the same regardless of whether the pond was installed on the roof of the poultry enclosure or on the ground.

The $10,000 allotted for algae removal leaves considerable latitude in the size and type of batch centrifuge and drying facilities used. The production of saleable high-protein algae suitable for animal feed supplements is an ancillary benefit that was not included in the economic analysis given in Table 2. Depreciation was calculated on a straight

line for a period of ten years. Working capital was assumed to be 7%. Utilities and repair of equipment will vary with the region, however, $5,000 should cover most regions for the anticipated type of equipment. The $5,000 allotted for additional labor was assumed to cover the wages of a laborer on a one-half time basis.

The total estimated annual cost was $38,900 or 39¢/chicken for the 100,000-bird operation. Based on the expected average egg production, this would amount to 2¢/dozen eggs. If the cost benefits of algae production were taken into consideration, the net annual cost would be reduced by at least $9,800. This figure is based on a value derived from only that fraction of the algae actually harvested by centrifugation in the experiments, and hence is much lower than would be the case in an operation in which algae production was emphasized. The bases for the $9,800 estimate are on algal yield of 6 tons/acre-yr and a market value of $100/ton. (The harvesting costs were included in the overall $38,900 estimate--see Table 2). The quoted market value is based on a compromise between the two demonstrated potential uses of the algal product, namely as a substitute for fish meal or for soybean oil meal. Fish meal sells for about $150/ton, and soybean oil meal for approximately $80/ton (16). As was stated earlier, in the experimental run, harvesting was carried on only for about 6 hrs/day and 5 days/week. Experience with algae production on waste waters indicates that the annual yield could well be 20 tons/acre-yr. Assuming a more realistic and yet conservative yield of 12/tons acre-yr, the market value of the algal crop then would be $19,600/100,000 hens, or an annual cost of about 20¢/bird-yr. This reduces the wastes handling costs to approximately one cent per dozen eggs.

Linton (17) reported in 1966 for two rural New York counties an average manure handling cost of 0.75¢/dozen eggs for a six months storage and later spreading in addition to 35¢/chicken for the initial installation. Calculated by the same method as the proposed system, this would amount to approximately one cent per dozen eggs. This does not include the difference between the present prices and the pre-1966 prices. Therefore it is obvious that the two cents per dozen eggs for manure handling in the proposed system (excluding algae recovery), even though based on the higher cost components and current construction costs, compares quite favorably with the approximately one cent per dozen eggs for a rural area where manure spreading can still be carried out within a reasonable distance.

Overall Evaluation of the System: The little, if any, increase in costs incurred in using the photosynthetic reclamation system described herein over the more adequate of the conventional systems for handling and treating animal wastes is offset by the benefits of providing a sanitary and efficient method of handling wastes with low adverse environmental impact. Its reclamation aspects and potential have much to offer in that its by-products are a high-protein feedstuff and where desired, an effluent water low not only in plant macronutrients, but also in the micronutrients. Of course the system is by no means a panacea for all organic waste problems. For instance, its main value is for large-scale operations, and it perhaps is limited to certain areas of the country (perhaps 40 to 50%) because of climatological factors. However, there is a multitude of situations in which the system, or portions of it, could be applied with great advantage in the protection of the environment. In view of the emerging national policy to develop systems for recycling critical resources, and in view of the emphasis on environ-

mental enhancement, closed systems for conveyance, treatment and regeneration of animal manure should be considered as a major alternative to other methods in the design of wastes systems for the future.

## TREATMENT OF POTATO WASTES THROUGH ALGAL CULTURE

### Introduction

The possibility that micro-algae could be produced on potato processing wastes had been suggested during a presentation before the potato industry at Sun Valley in 1963 (18). At that time, there was no information to indicate whether or not algal growth on potato wastes was a feasible concept; and indeed it was unknown whether algae cultivation could be practiced economically with any waste but sewage. In the intervening years, however, algae culture studies have been extended to include beet sugar flume wastes, Steffens waste, and chicken manure. Moreover, two studies have been made on growing algae in potato wastes. The first involved the potential of potato wastes to support algae in an aerobic system, and the second involved anaerobic fermentation of potato wastes as a step toward algal growth.

The literature on potato wastes reports little concern to date for the problem of plant nutrients (19, 20). However, analyses showing the major characteristics of the wastes do exist. The data listed in Table 3 give an idea of the nutritional quality of potato wastes as compared to that of other wastes. As is now well known, carbonates, nitrates, phosphates, and other oxides are produced in the bacteriological decomposition of all wastes, including those from potato processes.

Based upon the nitrogen content, the algal growth potential (AGP) of the potato waste listed in Table 3 is 1,800 mg per liter. As based on phosphorus, it is 2,000 mg per liter. Using BOD as a basis, there

TABLE 3

Comparison of Domestic Sewage, Beet Flume Water, and Potato Waste With Standard Algae Nutrient

| Characteristics | Domestic Sewage | Beet Sugar Wastes | Standard Algae Pilot Plant (inorganic) | Potato Wastes |
|---|---|---|---|---|
| Total Solids | 800 | 3000 | 6250 | 4000 |
| Volatile | 500 | 1500 | -- | 2500 |
| Total N | 60 | 50 | 315 | 180 |
| Organic N | 28 | 20 | -- | 170 |
| Ammonia N | 30 | 30 | -- | 10 |
| Nitrate N | 2 | | 315 | 1 |
| Phosphorus (P) | 20 | 5 | 300 | 20 |
| Sulfur | 8 | 100 | 300 | -- |
| Potassium | 12 | 80 | 1690 | 100 |
| Alk. mg/liter as $CaCO_3$ | 200 | 500 | -- | |
| 5-day 20°C BOD | 350 | 1000 | 00 | 2500 |
| pH | 7-8 | 9-11 | 6 | 9-11 |
| Magnesium | 18 | -- | 492 | |
| Calcium | 7 | -- | | |
| Trace Minerals | Pres. | Prob. pres. | Must be add. | Prob. pres. |

is sufficient carbon in the waste to yield an algal crop of about 1,500 mg per liter of waste. Thus, it appears that carbon may limit algal growth in some potato waste. Since potato wastes are of organic origin, it is logical to assume that all trace elements are present although this could be erroneous if precipitation of polyvalent metals were to occur due to the high pH of the wastes.

Experimental:

To determine how much of the algal nutrients in potato wastes actually are available for algal growth, experimental work was initiated which involved the use of bioassays as the major experimental procedure.

The assays were carried out in accordance with procedures for AGP determinations, which are currently under standardization by EPA (21). Essentially, the tests involved serial dilutions of the waste with water and inoculation of the dilutions with bacteria and algae. The inoculated samples were then incubated in the light and at temperatures such that bacterial and algal growth occurred freely. The tubes were mixed and sniffed daily for odor. If found to be odorous, the indication was that algae growth in the dilution at hand was insufficient to meet the oxygen demand of the bacteria at that time. In samples that were fresh-smelling, the algae were evidently meeting the oxygen demand of the bacteria. At the end of the incubation period indicated by no further increase in algae growth, and in this case in 10 days, the dry weight of algae was determined and its oxygen equivalent compared with the BOD. The oxygen equivalent or theoretical oxygen production (TOP) was computed by multiplying the algae concentration ($C_c$) by 1.6.

Results of a typical experiment are listed in Table 4. Judging from the amount of nitrogen and phosphorus removed from the waste, the

TABLE 4

Photosynthetic Oxygen Production in Potato Waste

| No. | Algae ml | Water ml | Potato Waste ml | BOD[b] mg/l | $C_c$[c] mg/l | TOP mg/l | TOP BOD |
|---|---|---|---|---|---|---|---|
| 1 | 2 | 0 | 100 | 1980 | -- | -- | -- |
| 2 | 2 | 50 | 50 | 990 | 875 | 1400 | 1.42 |
| 3 | 2 | 66 | 33 | 660 | 800 | 1270 | 1.89 |
| 4 | 2 | 85 | 16 | 396 | 437 | 700 | 1.76 |
| 5 | 2 | 91 | 9 | 200 | 138 | 220 | 1.10 |
| 6 | 2 | 98 | 2 | 40 | 50 | 80 | 2.00 |

[a]N = 150 mg/l; P = 12 mg/l

[b]Based on 60% depletion of $O_2$

[c]Concentration of algae in mg/l

[d]TOP - theoretical oxygen production

AGP of the waste was 1500 mg/liter for N and 1200 mg/liter for P. Based on the actual AGP attained, the 50% dilution produced 875 mg/liter of algae. Thus, it appears that following bacterial action, most of the nitrogen and phosphorus became available to algae, and in addition algae growth was stimulated to an extent wherein the actual concentration of nitrogen in the cells fell below 10% and the concentration of phosphorus in the cells fell below 1%. For example, assuming that phosphorus was a limiting factor in the test, the ratio of phosphorus in the cells must have been 600/875 x .01 = .0069%. A shortage of phosphorus in the waste would account for the decreased TOP/BOD ratio in the 10% dilution.

It is significant that in all cases where growth occurred the TOP/BOD was greater than 1. Failure of algae to grow in the undiluted waste in this test was attributed to pH level in excess of that favorable to bacterial action. However, at the end of 10 days, some green became apparent in the undiluted samples; and after 20 days, they too became green. It was concluded that a stronger inoculum of algae adapted to potato wastes might grow rapidly in pure potato waste with adjusted pH. This was found to be the case. Accordingly, algal cell material from previous growth experiments was conserved and used for inoculum in subsequent experiments. This proved to be practical since adapted algal inoculum was found to grow very rapidly in full-strength potato waste.

The possibility that phosphorus or pH limited the growth of algae on potato waste was tested by assays similar to those described above. Growth in pure waste following phosphate addition and carbonation was to be compared with growth in the pure waste attained in a 1:1 dilution. The pH of potato waste was lowered from pH 10 to 6 by bubbling $CO_2$ through the waste for two minutes. The rate of bubbling was about 0.2 SCUF pure

$CO_2$ per minute in a 1-liter vessel of waste. After the waste had attained a pH of 6.0, triplicate flasks were inoculated with 5 ml of adapted inoculum and incubated for 8 days. The mean growth is indicated by the data in Table 5.

Similarly in testing the effects of phosphate addition, $H_3PO_4$ was added to give a final concentration increment of 8 mg per liter of available phosphorus as P. Again, triplicate samples were incubated and the concentrations of algae determined as a function of time. Mean growths are listed in Table 5. To further test the efficiency of 1:1 dilution of the potato wastes in tap water, triplicate samples were made up and incubated as above. The concentrations of algae are shown in Table 5. The results show that algae grow vigorously in raw undiluted potato waste if either phosphate is added at the 8 mg/liter level, if carbonation is applied, or if the waste is diluted 1:1. In each case the 8-day growth represented a theoretical oxygen production greater than the BOD of the waste. Early in the test some odors were noted but they became negligible by the third or fourth day. The TOP/BOD ratio was 1.3 for phosphate addition, 1.23 for carbonation, and 1.46 for dilution. Thus, although carbonation and phosphate addition resulted in higher growth rates, simple dilution resulted in superior oxygenation and a shorter period of odor production.

A study of odor emission was made concurrently with all other tests. A more precise experiment was subsequently conducted. To assure that odor-forming organisms would be present, a weak domestic sewage having a BOD less than 100 mg/liter was used as dilution water. As is evident from the data on odor production given in Table 6, odors persisted in the concentrated wastes up to 7 or 8 days, whereas the 1:1 diluted

TABLE 5

Concentration of Algae ($C_c$) and Theoretical Oxygen Production (TOP)[a] Under Phosphate Addition, Carbonation, and Dilution[b]

| Time Days | 100 ml PW 8 mg/1 P $C_c$ (mg/1) | 100 ml PW $CO_2$ to pH 6 $C_c$ (mg/1) | 50 ml PW 50 ml water $C_c$ (mg/1) |
|---|---|---|---|
| 1 | 490 | 250 | 210 |
| 2 | 840 | 512 | 420 |
| 3 | 1090 | 700 | 615 |
| 4 | 1340 | 870 | 755 |
| 5 | 1470 | 1060 | 850 |
| 6 | 1540 | 1230 | 880 |
| 7 | 1580 | 1400 | 910 |
| 8 | 1600 | 1520 | 910 |

[a]To obtain theoretical oxygen production, multiply $C_c$ x 1.6

[b]Inoculum - 5 ml algal suspension

TABLE 6

Odor Endurance at Various Dilutions of Potato Wastes in Domestic Sewage

| Sewage ml | Potato Waste ml | Algae Seed ml | Days | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 |
| 90 | 5 | 5 | + | + | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| 85 | 10 | 5 | + | + | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| 80 | 15 | 5 | + | + | - | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| 75 | 20 | 5 | + | + | + | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| 70 | 25 | 5 | + | + | + | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| 65 | 30 | 5 | + | + | + | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| 63 | 35 | 2 | + | + | + | - | 0 | 0 | 0 | 0 | 0 | 0 |
| 58 | 40 | 2 | + | + | + | + | 0 | 0 | 0 | 0 | 0 | 0 |
| 53 | 45 | 2 | + | + | + | + | 0 | 0 | 0 | 0 | 0 | 0 |
| 48 | 50 | 2 | + | + | + | + | - | 0 | 0 | 0 | 0 | 0 |
| 43 | 55 | 2 | + | + | + | + | 0 | 0 | 0 | 0 | 0 | 0 |
| 38 | 60 | 2 | + | + | + | + | 0 | 0 | 0 | 0 | 0 | 0 |
| 33 | 65 | 2 | + | + | + | + | + | 0 | 0 | 0 | 0 | 0 |
| 28 | 70 | 2 | + | + | + | + | + | 0 | 0 | 0 | 0 | 0 |
| 23 | 75 | 2 | + | + | + | + | + | - | 0 | 0 | 0 | 0 |
| 18 | 80 | 2 | + | + | + | + | + | + | 0 | 0 | 0 | 0 |
| 13 | 85 | 2 | + | + | + | + | + | + | + | 0 | 0 | 0 |
| 8 | 90 | 2 | + | + | + | + | + | + | + | 0 | 0 | 0 |
| 3 | 95 | 2 | + | + | + | + | + | + | + | - | 0 | 0 |
| 0 | 100 | 2 | + | + | + | + | + | + | + | 0 | 0 | 0 |

waste became odorless in about 4 days. The indication here is that in any system, detention periods of at least 5 days should be used, and treated waste should be recycled at least 1:1 in order to avoid odors.

Preliminary studies on the anaerobic fermentation of potato wastes indicate that although potato wastes are subject to methane fermentation, extended continuous fermentation will not sustain itself without provision for pH and odor control. Thus, in any system in which anaerobic fermentation is applied to pretreat potato wastes for subsequent algal growth, recirculation and dilution of the waste with buffered final effluent water would be desirable.

Discussion:

From the preliminary data presented herein, it is evident that the potato wastes contain the nutrients needed for algal culture, and that a system can be designed in which algae provide all of the oxygen required for oxygenation and complete treatment. If the entire algal growth potential of the waste were exerted, production would be on the order of 1,200 mg of algae per liter, or about 5 tons per million gallons of waste processed. At 5 cents per pound, the value of the algae would be on the order of $500 per million gallons of potato waste.

To obtain such quantities of algae, a completely controlled system would be required and could be obtained only with a substantial capital investment. The problems of inclement weather would have to be compensated and complete mixing of the system would be required. However, it seems likely that if a suitable market could be found for the algal product, such an investment would be justified. There is a new system for growing algae in greenhouse fashion, but it remains to be seen whether such a complex system would be justified for by-product recovery

from potato wastes. One such system is the algatron (22) in which cultures of algae as dense as 6 grams per liter can be produced in as little as 12 hours' detention period. If such a system could be justified economically, there is little doubt that complete in-house treatment and reclamation of potato wastes could be attained essentially independently of weather.

The beneficial effects one might hope to attain from the utilization of algae culture in treating potato wastes would be an improved effluent, reduced aeration costs resulting from economic production of oxygen, reduction in odors of holding ponds, production of by-product algae, and nutrient stripping. Theoretically at least, use of a waste for algae production is comparable to use of wastes for irrigation of crops with two exceptions. Algae production per unit of area is 10- to 20-fold that of crop production; and with algae production, most of the water is not consumed and hence can be used for irrigation directly, or reused in the plant following algae removal. From a production standpoint, intensive algae culture on potato waste would yield algae aesthetically suitable for human consumption.

A major deterrent to growth of algae on potato wastes is climate. Potato production in northern latitudes from mid-September to early May bridges the winter months when algae growth is most limited. Thus algae culture on potato wastes must be applied during early fall and late spring, with alternate methods used during the winter. The alternative is to store the wastes for processing during the summer. Another alternative is greenhouse culture applied the year around. The latter would be expensive and hence only economically feasible if algae could be solid in large volumes at moderately high prices. The cost of greenhouse algae

production would most likely be 20 to 30 cents per pound--a cost greatly exceeding that of production in outdoor ponds. Because the greenhouse-produced algae would be a pure by-product of a vegetable processing system, the product might command a price of 50 cents per lb on the open market, which would offset the high production cost. The benefits of waste treatment, nutrient stripping, and water recovery would also accrue to the industry if greenhouse culture were practiced.

### Conclusions

The results reported herein indicate that large quantities of edible algae can be produced from potato wastes concurrently with complete oxidation of the waste and removal of algae nutrients. More intensive investigation of the kinetics of algae growth in potato wastes should be carried out in the near future. Studies should also be undertaken to examine the benefits and disadvantages of combining vegetable and animal wastes in integrated regenerative systems.

## IV. POTENTIAL OF PHOTOSYNTHESIS IN WASTE TREATMENT SYSTEMS

### GENERAL CHARACTERISTICS

As is well known today, conventional waste treatment systems which were suitable a very few years ago to the preservation of a healthful and aesthetically satisfactory environment, no longer are adequate to meet the burden imposed upon them under present-day conditions, nor would they be able to do so under the forecasted future conditions. Conventional waste treatment processes traditionally have been designed around the concept of the biostabilization of degradable organic wastes to decrease their oxygen consumption capacity (BOD). In general, these processes have been concerned mainly with providing some means of

oxidizing organic products, and very little with the products of this oxidation or of the minor nutrients not affected by the treatment process.

The situation now has changed, and attention is being devoted not only to the problem of oxidizing the organic wastes, but also to that of preventing from entering receiving waters those substances in municipal and agricultural wastes which could support undesirable aquatic growth in the receiving waters. As a consequence, more and more waste water treatment plants are being modified or new ones constructed to have the capacity not only of decreasing the BOD of the waste waters but also of reducing the concentration of nitrogen and phosphorus in the waters. The difficulty is that although carbon, nitrogen, and phosphorus are essential to aquatic growths, merely removing even a large percentage of the substances from organic wastes does not always sufficiently decrease the algal growth potential (AGP) of the waste waters so treated (23). Apparently, in organic wastes an unassessed concentration of vitamins, certain amino acids, and trace elements, remain in the effluent in quantities sufficient to stimulate a leisurely growth of algae in the natural environment.

It is in the removal not only of oxygen-demanding substances and of nitrogen, phosphorus, and carbon, but also of the biostimulants that treatment systems involving the culture of algae are especially useful. Hence, the wastes management system described herein is especially valuable in those situations in which complete (i.e., primary, secondary, and tertiary treatment is demanded. This function is served regardless of whether the system is operated as a closed or an open system. If operated as a closed system, the effect is obvious; there is no effluent and hence no discharge of nutrient to receiving waters. If operated as

an open system, the effluent will have been subjected not only to conventional treatment but also algal culture, and hence the greater part of the AGP will have been removed.

TYPES OF APPLICABLE PHOTOSYNTHETIC SYSTEMS

Although the potential combinations of systems are many, four types of photosynthetic systems stand out. They are: 1) systems design primarily for waste treatment; 2) regenerative agro-industrial complexes designed as protein factories; 3) operation designed to convert solar energy to the chemical energy of methane; and 4) small-scale regenerative systems for family farms or small villages.

The first system is one in which wastes from a community or industry are converted into algae in a line process, designed to take only those wastes which require disposal. All algae produced in a plant would be sold on the open market as a protein supplement, the water recycled, and the values received would be used to defray a part or perhaps all of the cost of waste disposal.

In the second system--agro-industrial complex designed as a protein factory--all animal wastes would be recycled in the system, and exports consisting of animal products, rather than of algae, would be the output of system. A hypothetical system of this type was described in 1962 (24), and the present report described a pilot plant application of the type.

The third system, namely a process for converting light (solar) energy to the chemical energy of methane seemed rather exotic to the authors in 1959, the time they conceived and developed the process (25, 26); but today, in view of the increasing concern over the conservation of energy, its approach to everyday practicability has been

advanced considerably. The process involves an approach to solar power that goes back to nature's sequence, namely solar energy to plant life, to fuel by way of growing algae on wastes and digesting the algae to produce methane. The algae fix the visible light spectrum of solar energy into algal protoplasm. The algae are killed and subjected to conventional anaerobic digestion. Through the digestion stage, the chemical energy of cellular protoplasm is transformed into the chemical energy of methane.

Although the fourth type of system appears to be feasible today, it has not been demonstrated as a miniature regenerative system of subsistence size designed to be incorporated in family farms or small villages. Only excess animal products would be exported from the system, and essential food, feed, and water would be imported. Such a system has a substantial potential for decentralized waste disposal and recycle in tropical countries having rural or semirural populations currently existing at a lower than minimum standard of living. Geographically, the system would find its most immediate and practical application in countries characterized by the possession of a tropical climate. Certain key components of these regenerative systems were demonstrated as being technically feasible on an experimental basis in the studies reported herein. There is a high probability that they would perform much better in tropical areas. This is because temperature, more frequently than too little light, is the limiting factor to algal growth and bacterial decomposition in the temperate zone. Temperature would always be near optimum in many areas of India, and light would be more than adequate except perhaps during the Monsoon season. Thus Bombay, Nagpur, or Kanpur should be excellent locations for trying the system.

A detailed description of the design for an application of the fourth system is given in Appendix A.

## V. RECOMMENDATIONS

Inasmuch as the advantages accompanying the application of photosynthesis to agricultural waste treatment, as was demonstrated by the work reported herein, it is imperative that the additional research and development needed to ensure its successful utilization in general practice be given a high priority. The research remaining to be done was outlined in the introductory remarks to Part III of this report. For the sake of emphasis, they are repeated here: 1) Determine the rate and extent of solids (sludge and salts) build-up when the hydraulic portion of the system is maintained as a "closed" system. ("Closed" is meant in the sense that no water other than overflow from the animals' drinking water supply is added to the system.) 2) Determine the required rate of repletion of desired elements and removal of inhibitory substances. 3) Determine the minimum pond area requirements per bird or animal. 4) Assess the utility of dewatered settled solids residue as a feedstuff for ruminants, especially with the protein content enhanced by adding harvested algae to the material. 5) Try the system with animals other than poultry--for example, dairy cattle or steers. 6) Determine the effects of "scale-up" in questions 1 through 5 by conducting a demonstration-sized operation. 7) Determine the ability of animal wastes to increase the treatability of vegetable wastes.

An appropriately sized demonstration operation for conduct at the University's Richmond Field Station would be one geared to a pond having an area of two-thirds acre; inasmuch as such a pond already is in

existence at the Station, a pond area of that size would warrant a facility having a capacity for 4000 hens.

The design specifications for such a facility are given in Table 7. Information in the table covers all of the data pertinent to arriving at the dimensions named therein, and is self-explanatory.

TABLE 7

Design Specifications for a 4000-Bird Demonstration Facility

A. Equipment Available

1. Two-thirds acre high-rate algae pond

2. Algae removal equipment (continuous centrifuge; batch centrifuge; drum dryer)

3. One-quarter acre anaerobic-facultative pond; bottom needs to be cleaned of sulfur deposits and/or lined with plastic (visqueen).

4. Complete chemical laboratory, maintenance shop, etc.

B. Chicken Feed and Water

0.25 lbs/bird-day (av feed consumption). 1000 lbs/day = 30,000 lbs/mo = 15 tons/mo.

Assume feed costs $4.50/100 lb or $90/ton. ∴ 15 tons/mo x $90/ton = $1350/mo Feed cost per month.

1. Feed Storage

Assume feed to weigh 50 lb/ft$^3$. ∴ one-months supply required min. of 600 ft$^3$ Net months feed storage requirements.

Farm agent and/or literature should be consulted for proper feed storage construction.

2. Water Supply

Water troughs or pressure release valves. Farm agent or poultry growers should be consulted for the amount of maintenance required for each. Adequate quantity of water available at SERL.

C. Egg Production

Assume a maximum egg production of 75% of the original number. ∴ 4000 x 0.75 = 3000 eggs/day = 250 dozen/day.

For convenience in handling and marketing the eggs, could be sent to a processor for cleaning, candleing, etc., except for eggs used in analyses. The eggs should be counted and weighed.

Return on eggs may defray some operational costs of the project.

D. Manure Production

Assume wet manure production is 0.4 lbs/day-bird. 4000 x 0.4 = 1600 lbs/day.

TABLE 7 (cont.)

Design Specifications for a 4000-Bird Demonstration Facility

E. Expected Fatality Rate

Assume 1½% month. 4000 x 0.015 = 60 birds/month.

F. Chickens

Assume 20 week old pullets - cost $1.70/ea. 4000 x $1.70 = $6,800.

G. Building

Assume bird density @ 0.5 $ft^2$/bird.
Assume aisle required @ 0.5 $ft^2$/bird.
∴ 1 tier - 1 bird/$ft^2$ of floor area
2 tiers - 2 birds/$ft^2$ of floor area
3 tiers - 3 birds/$ft^2$ of floor area.

Assume the use of flushing manure troughs at a slope of 6 in per 10 ft.

For convenience, assume 2 tiers.

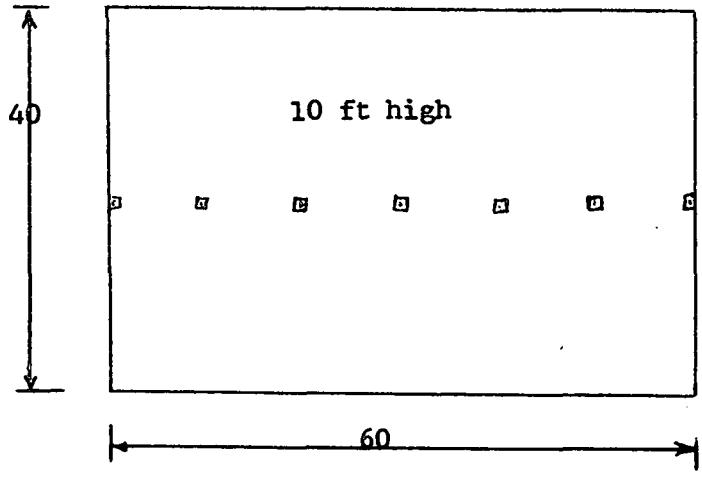
∴ 4000 birds ÷ 2 birds/$ft^2$ = 2000 $ft^2$ building.

Minimum feed storage 600 $ft^3$. Assume feed storage @ 4 ft.
600 $ft^3$ ÷ 4 ft = 150 $ft^2$.

Total area 2,000 $ft^2$ + 150 $ft^2$ = 2,150 $ft^2$.

Assume 40 ft width. 2150 $ft^2$ ÷ 40 ft = 53.75 ft.

For building convenience, design structure 40 ft x 60 ft x 10 ft high.




Metal roof, fiberglass sides

Suggest construction similar to pilot plant

TABLE 7 (cont.)

Design Specifications for a 4000-Bird Demonstration Facility

H. Sedimentation Tank

Two 6 ft x 6 ft x 10 ft high conc. pits or two circular 8 ft dia x 8 ft concrete cyclinders. Two turbine pumps similar to the present pilot plant one should be adequate. ~ $600-$800 ea.

I. Misc. Equipment (depending on final design)

Pumps
Gauges
Metering devices
Centrifuges (manure solids separation).
Overhaul existing units as required.

# APPENDIX A

## DESIGN OF AN ALGAL REGENERATIVE SYSTEM FOR SINGLE FAMILY FARMS AND VILLAGES

A schematic diagram of a typical small-scale algal regenerative system is shown in Figure A-1. In addition to its living occupants, the system would involve an anaerobic digester, a series of algal growth chambers, a sedimentation chamber, sand beds, a solar still, and a gas exchanger. Inasmuch as it is combined with a residence needing gas for cooking, the anaerobic digester should be covered so that combustible gas would accumulate under the cover at a pressure sufficiently above atmospheric, to permit its conveyance. Excess gas, which would be rich in methane, (i.e. 55 to 65%) would be conveyed from the gas dome through conduits into the residence, where it could be used for household purposes. Digested solids would be periodically drawn from the digester to be used for soil conditioner or fertilizer in the growth of vegetables on a soil plot nearby.

The hypothetical minimum sized system described above would provide waste disposal and nutrient recycle for 4 humans, 1 cow, and 50 chickens. Such a system was arbitrarily selected as the most elementary that could be operated. Because of its small size it would be most economical for research studies. Based upon design data collected for a single family system, larger units could be made for larger populations.

The manner in which the size of the components of this family-sized system was estimated, was as follows: The algae production unit

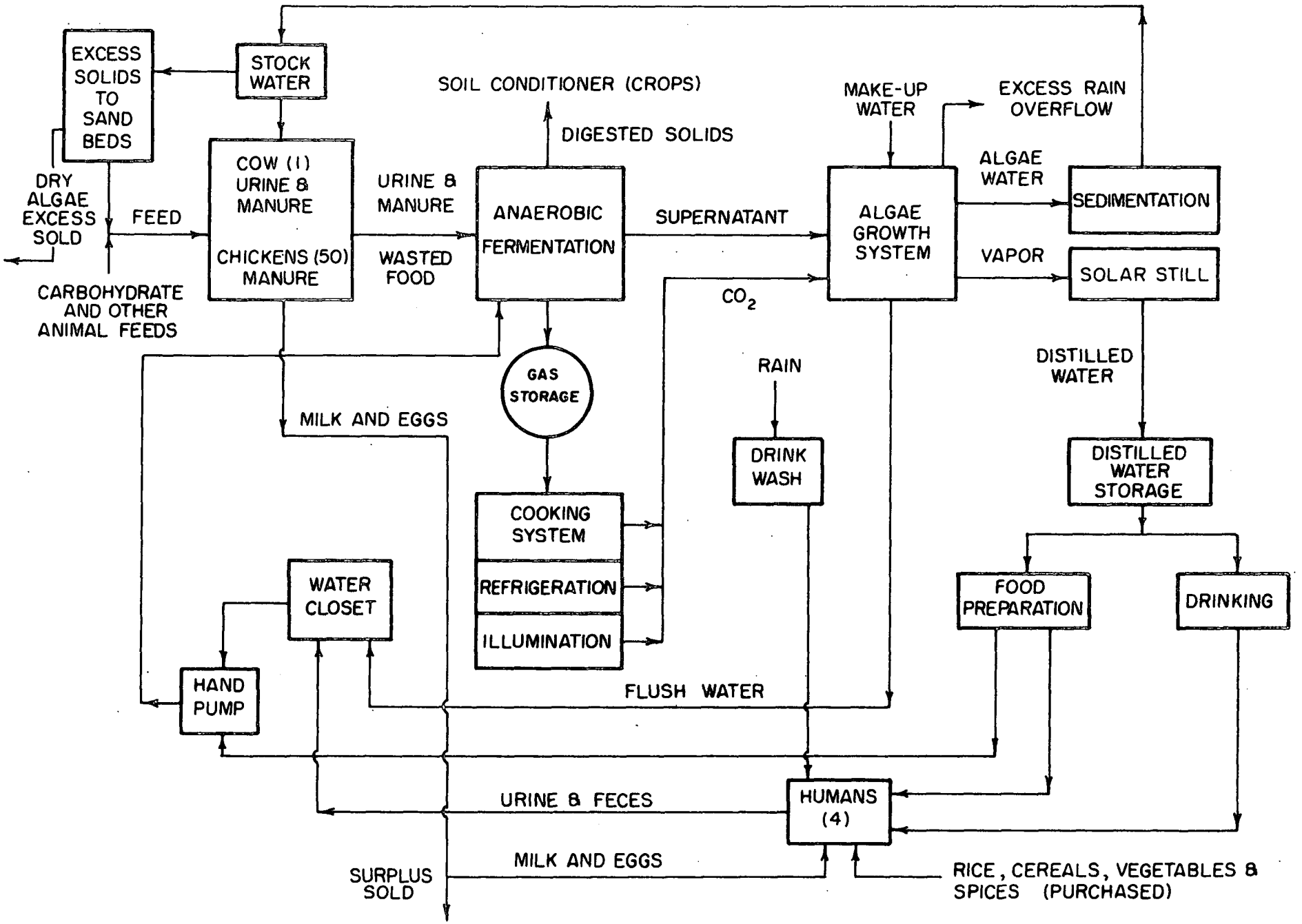



FIGURE 9. SCHEMATIC DIAGRAM OF SINGLE-FAMILY MICROBIOLOGICAL ORGANIC WASTE RECYCLE SYSTEM

which is the most crucial part of the system was sized on the basis of waste nitrogen from the contributing population. For example, an efficient milk cow may process 120 grams of nitrogen daily, excreting 20 grams in milk and 100 grams in feces and urine. An efficient laying hen may process 3 grams of nitrogen daily, excreting 0.6 grams in eggs and 2.4 grams in feces. With one cow and 50 chickens, the animal contribution in waste would be 2,220 grams (4.9 lbs) of nitrogen per day. Each human excretes about 12 grams of nitrogen daily; therefore four persons would waste about 48 grams of nitrogen per day. The aggregate amount of waste nitrogen would then be 268 grams daily. Past experience indicates that in such a system, nitrogen, phosphorous and other elements would always be in excess with respect to carbon. It also indicates that under such conditions the composition of the algae may be assumed to be 10% nitrogen, as compared with 6 to 8% nitrogen found in less nitrogen-rich waters. Thus, if all waste nitrogen were to be recycled, the amount of algae grown each day would be 2,680 grams (5.9 lbs). If the production rate is assumed to be 0.33 grams per gram of algae per day, the standing biomass of algae required would be 8,130 grams (18 lbs). Assuming a concentration of 500 mg per liter, the culture volume required would be 16,250 liters (4294 gal).

In determining the design depth for a culture, experience has shown that algal cultures which are adequately mixed will attain a concentration such that light penetrates to 1/3 the culture depth. Light of daylight intensity will penetrate about 12 cm into a culture having a concentration of 500 mg per liter, therefore the culture depth should be 36 cm. A 36-cm deep culture occupies 28 $cm^2$/liter. Therefore to contain about 16,250 liters one would need an area of 58 $meters^2$

(623.5 sq ft). This would be provided by a circular pond about 8.6 meters (28.2 ft) in diameter, but for structural uniformity, the diameter selected should be about 9 meters. Thus a total area of about 64 sq meters (688 sq ft) would be provided by adding a small factor of safety to the computed 58 sq meters. Thus a circular culture 64 meters in area, 9 meters in diameter, and 36 cm deep is required. Practically speaking, a culture of this volume and dimensions in India should produce between 2 and 5 pounds dry weight of 50% protein algae per day, depending on temperature and light.

The size of the digestion element of the system was based on an established microbiological minimum per capita criterion of 1 $ft^3$ (0.028 $m^3$) for humans, 10 $ft^3$ for cattle, and ¼ $ft^3$ for chickens. The minimum aggregate volumetric digester requirement for a population of 4 humans, 50 laying hens, and one milk cow is 26 $ft^3$ (0.736 $m^3$). The minimum manageable digester, however, should be 1 meter in diameter and 2.5 meters deep. This would provide an active volume of 55 $ft^3$ (1.56 $m^3$), which is almost twice the computed minimum requirement. Such an oversize digester should provide superior fermentation and excellent gas conversion.

The digester, as noted previously, should be provided with an inverted dome-type cover for gas pressurization. It should also be equipped with a charging chute through which the manures and night soil and food and feed waste could be introduced to the liquid digestion phase without permitting gas to escape. This may be accomplished by constructing the recharge chutes with a side entrance below the water line. For most efficient use, the digester should be at the center of the ponding system and living area so that overflow supernatant would

move directly into the algal pond system, so that heat may be conserved.

Although the algae culture would only be about 0.36 meters deep, the algae pond walls should be 0.5 meters high to provide freeboard. The culture floor should slope slightly outward from the center so that any settled algae would be moved away from the digester during mixing. The pond system itself should actually be 3 concentric ponds in series, offering protection against transfer to or survival of pathogens in the final pond. Three or more ponds in series insures an excellent degree of treatment.

In the outer final section of the pond system, a collector hopper ½ meter wide and 1/3 meter deep would be provided as a sump in which settleable algae could be collected. The algae would be drawn periodically through a valve into a watering system for the cow or onto sand beds for drying. A total of 8 sq meters of sand beds involving 4 beds of 2 sq meters each would be required. The sand beds should be underdrained so that water could be collected and returned to the system if needs be.

The human occupants of the system will require fresh potable, low solids water. This would be provided by a 20 $ft^2$ (1.86 $m^2$) solar distilling apparatus mounted so that it captures vapor from the algal culture. A 20 $ft^2$ solar still would yield about 10 gallons (37.85 liters) of distilled water per day, which should be adequate for drinking and cooking for 4 humans. During the monsoon, the solar still would be used as a catchment area for rain water to be used in lieu of distilled water. During the dry season of the year, evaporation would be extensive from the algal culture surface, and on the hottest, windiest days

could be as much as 757 liters (200 gallons) per day from the 64 sq meters of surface. A source of makeup water will therefore be required. Ground or surface water may be used, but it will have to be piped or carried manually into the system each day. This water would be used for bathing, for drinking water for the chickens (chickens are not tolerant to salt), and for cleansing purposes. The quality of this water should be carefully controlled lest excess spillage lead to unsanitary conditions. If the system were covered with transparent materials to admit light but prevent evaporation, excess heating of the culture probably would occur. However, the possibility of utilizing high-temperature strains of algae in a system with transparent cover should be investigated, since in a covered system the need to import large amounts of water would be reduced. During heavy rains, the overflow from the final pond of an open system would be suitable for disposal to surface or ground waters.

Mixing of the algal system is essential and may be accomplished by brooms or paddles. The floor of the pond should be constructed of sufficient strength that individuals in boots could wade freely about the bottom during the mixing process.

By placing the algal culture above the house, the entire digestion and algal growth system could be located on a plot of land 10 meters by 10 meters; and should it be feasible to support the culture above the ground, it would provide shelter and living space beneath it for the animals and for the humans as well. While from a management standpoint there would be large economic advantages to having larger than single-family units, individual units would have the advantage that each family could take personal responsibility for its own system, and those most

diligent would benefit greatly from their own efforts.

A typical family unit of the size designed above and incorporated in the shelter complex is shown in Figure A-2. In operation of the system as shown, all manure, urine, wasted food, night soil, and clean-up water would be charged into the digester shortly after it is produced, or at least once daily. In the digester, fermentation once established would occur steadily and gas would be produced. Operationally, particular care would have to be exercized to avoid the unnecessary loss of any useful component; hence, providion should be made for all solids, liquids, and gases to be recycled or consumed. In the digester, complex substances would be decomposed in the form of organic acids, ammonia, $CO_2$, and methane. The methane would be burned. With the addition of the nutrients to the digester, soluble substances would be displaced into the algae culture, where they would provide substrate for algal growth. The methane, under slight pressure, would be available for gas cooking; and the gases from the gas-fired burners would be vented by convection to the algae culture, where a part of $CO_2$ content of the gases would be absorbed by the growing algae. Algal slurry would be provided to the cow as its sole source of drinking water, thus forcing it to consume algal protein in the wet form. Excess algae in the trough would be drawn onto sand beds and dried for use as cow or chicken feed, or for sale.

If living space were provided below the algal culture, it would provide shelter from the elements for both the humans and animals. The algal culture and digester would provide a buffer against rapid changes in temperature for the occupants, and the occupants would in turn to some extent warm the algae culture and digester during cool periods.

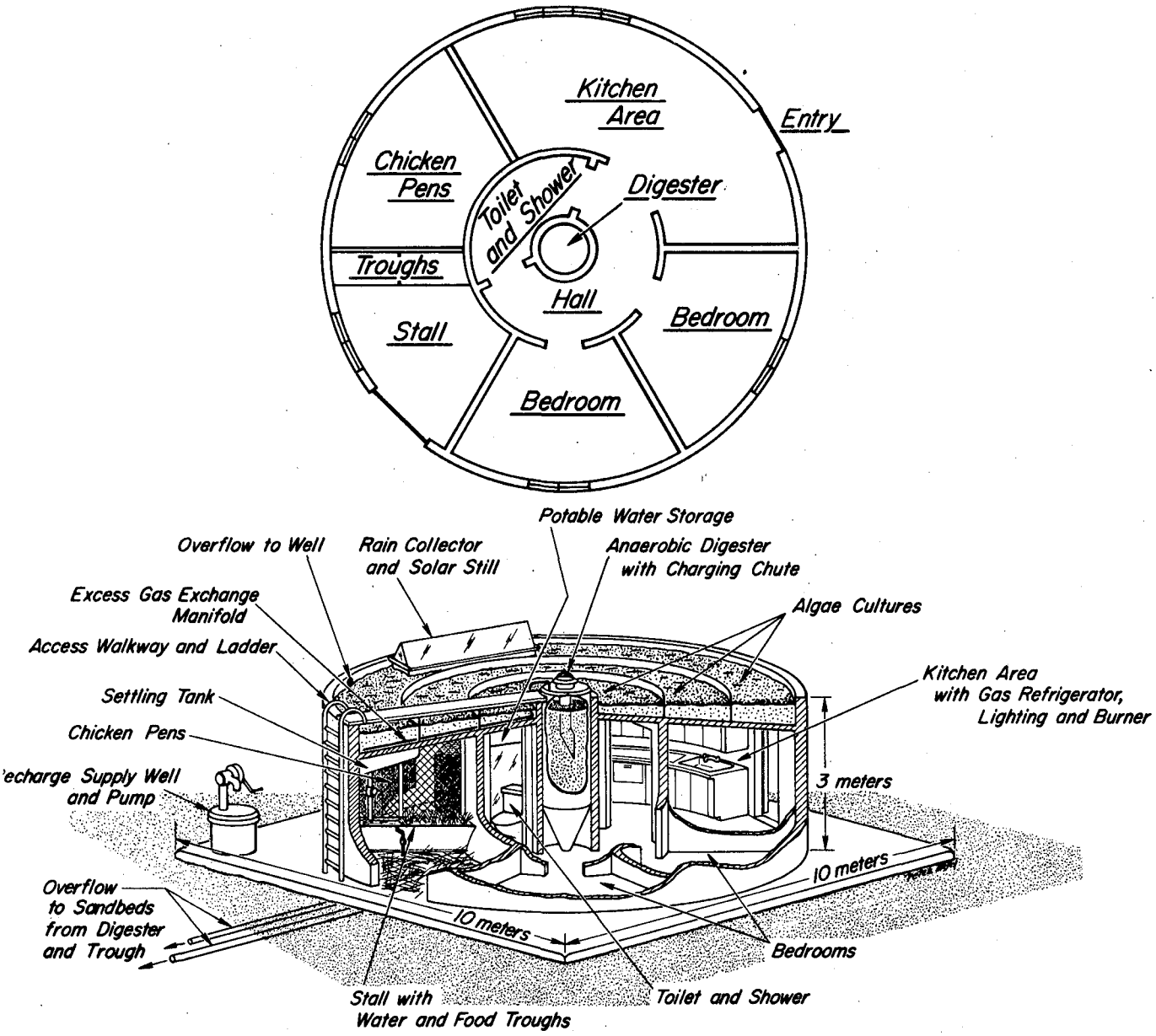



FIGURE 10. SCHEMATIC DIAGRAM OF A DWELLING UNIT FOR A FAMILY OF 4 AND THEIR LIVESTOCK WHICH INCORPORATES A MICROBIOLOGICAL RECYCLE SYSTEM FOR WATER, NUTRIENTS AND ENERGY IN A CONVENIENT AND HYGIENIC ENVIRONMENT

The system schematically shown in Figure A-2 is an advanced form of unit, which theoretically could provide an ample and hygienic environment for a family and its essential livestock. Such a unit could be bult of local materials or perhaps prefabricated for import. Because it is largely powered by sunlight energy, the feasibility of such a system should be greatest in tropical regions of the world, but it would be of use in other areas during the summer period. The advantages of such a system would be provision of a highly livable system for its occupants, provision of efficient decentralization of algal culture, efficient and hygienic waste management, and the recovery of valuable nutrients and water from wastes. Its major disadvantage would be the need for a substantial capital investment, a factor lacking for most impoverished families. However, through experimental studies with prototype units, functions and materials can be experimentally tested and, if required, modified in such a way that costs can be minimized, and operations perfected to a point at which the systems would be economically self-supporting. If this were possible, an economic incentive would exist for investors or for governmental agencies to provide such units on a long-term loan basis. The economic base for repayments of loans would come from maximizing production of milk, eggs, vegetables, and surplus algae, and minimizing the cost of essential inputs to the system such as supplementary feeds. Surplus eggs, milk, vegetables, and algae would be sold or bartered, and a portion of the money used to repay the capital investment for the system. A preliminary economic analysis indicates that a gross income of between $250 and $1,000 per year could be realized with such systems. Operationsl costs would be from $50 to $100 per year. If the higher income level could be attained, such units would probably

become economically attractive. On the other hand, if only the lower levels were reached, the use of such systems probably would be economically infeasible or would require substantial subsidies.

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