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RESEARCH PROJECT COMPLETION REPORT  
OWRR PROJECT NO. A-017-OKLA.

OPERATIONAL STABILITY OF THE EXTENDED AERATION PROCESS

Submitted to

The Oklahoma Water Resources Research Institute  
Oklahoma State University  
Stillwater, Oklahoma

Prepared by

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## COMPLETION REPORT

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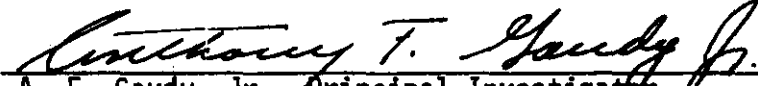
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#### PREFACE

In accord with the joint goals of the Oklahoma Water Resources Research Institute, the Center for Water Research in Engineering, and the Bioengineering and Water Resources Division of the School of Civil Engineering of Oklahoma State University, this research project was designed with the dual aim of accomplishing useful research and training graduate student researchers in important areas pertinent to the development and use of the nation's water resources. In addition to the independent value of the research findings of this project, the work has allowed the principal investigator to gain needed information on the extended aeration process, as well as on the metabolism of various cellular components, both of which have applications and useful ramifications to other work under way in the bioengineering laboratories. The project has also permitted a former student, Dr. M. Ramanathan, to participate (while he was a faculty member) in research and training in this important area. Three graduate students have received training through participation in research pertinent to this project, and have made significant inputs into the conduct of the work. Mr. W. Raghaidee conducted his Master's thesis research in this area. This thesis does not form one of the appendices to this report, since most of the work which is included in his thesis is covered in Appendix 2, which is a publication relative to his thesis findings. Mr. T. V. DeGeare worked for a time on research pertinent to this project, but did not do a thesis in this area. Mr. Alan W. Obayashi (working on the doctorate) is currently working in an area allied to the interests of the project. Mr. P. Y. Yang (MS thesis, Appendix 4; currently working on the doctorate) has participated in this project since its initiation. He also worked on the preliminary research pertinent to the extended aeration process before initiation of the current project. He has been employed as a research assistant on this project throughout its duration, and he is co-author of the present completion report.

The accompanying document represents a detailed report on the completion of the project, and embodies pertinent information suggested in OWRR "Reporting Guidelines." It consists of the summary report, which includes an introduction, statement of experimental approach, review of project findings, discussion summary, and conclusions. In the summary report, the four appendices are referenced as sources of detailed information.

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## COMPLETION REPORT

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A. F. Gaudy, Jr., and P. Y. Yang

#### ABSTRACT

This project was initiated to determine if an activated sludge process could operate with total cell recycle over an extended period of time. Previous researchers had concluded that such a process (the extended aeration or total oxidation activated sludge process) could not function and that cell or sludge wastage would be necessary. Some had concluded on the basis of short term studies that the process was theoretically impossible and that, because of the biological production of an inert organic fraction of cell mass, the sludge concentration would continually build up. The inert fraction would thus become an ever-increasing fraction of the activated sludge and the system would eventually undergo metabolic failure, i.e., lose the ability to purify waste waters. It was felt that these conclusions had never been subjected to complete testing; also, that if the system were to fail, there was need for engineering data to estimate how long such a system could be successfully operated before failure. Accordingly, a laboratory scale pilot plant activated sludge process was operated in which positive control of sludge retention in the system was obtained by passing all contents through a centrifuge and returning the separated biomass to the aeration chamber. Thus no sludge was wasted (purposely or inadvertently). This system was operated for three years and there was no evidence of metabolic failure; excellent purification was obtained. The sludge concentration did not continually build up, but underwent periods of accumulation and de-accumulation. The periods of de-accumulation corresponded to cycles of metabolism wherein autodigestion of the biomass exceeded new growth due to metabolism of the inflowing waste. At times the sludge was observed to build up to rather high concentrations, and it was reasoned that the autodigestion might be helped or controlled by an engineering modification of the process in which a portion of the sludge could be withdrawn and subjected to chemical hydrolysis. This concept (the "hydrolytic assist") was tested, and it was found that the hydrolyzed sludge was a readily available substrate for the remaining (unhydrolyzed) sludge. These findings (including a flow diagram for the process) were published. Also, some laboratory pilot plant research has been accomplished which indicates that the new process holds considerable promise.

#### KEY WORDS

total oxidation - extended aeration - aerobic digestion - autodigestion  
- chemical hydrolysis - microbial ecology

## INTRODUCTION

The current accelerated interest in water pollution control emphasizes the importance of secondary treatment, i.e., the removal of biologically oxygen demanding organic matter from waste water effluents. Various biological processes are available, and among these the activated sludge process is widely used. There are various modifications of the activated sludge process, and the extended aeration or total oxidation process is one which has been used increasingly over the past twenty years. This process, which was developed in the early 1950's, has found wide application for the treatment of wastes in rural communities, hotels, schools, and some industrial installations. Among original claims for the process were its simplicity of operation, its low cost, and its stability to environmental changes. The process differs from other modifications of activated sludge in that in its original form, no provision was made for disposal of excess biological sludge; all sludge was returned to the aeration chamber where it was thought that the process of endogenous respiration of the organic matter produced from the organic carbon in a waste would balance new growth of cells and an equilibrium condition would eventually evolve, thus providing for the "total oxidation" to  $\text{CO}_2$  and water of all of the organic matter in the waste. Soon after the process was conceived, many researchers conducted investigations relative to the operation of activated sludge systems without sludge wasting, and the general conclusion which was drawn was that the process was biologically unsound; i.e., it was theoretically impossible. The general consensus was that there would be a gradual buildup of biological solids in an activated sludge system operated without sludge wasting. Some ten to fifteen per cent of the

organic matter, it was felt, would be channeled into permanently inert materials. In time the system would thus build up a larger and larger inactive fraction which could neither be used as substrate by other organisms, nor could function as a viable biomass for the removal of organic substrates. Regardless of these objections by research engineers, the process has been used to an increasing extent and, were it not for the published conclusions warning against the use of the process on the basis of its so-called theoretical unsoundness, it might enjoy much wider application today.

In proposing the present study, it was felt that there were not sufficient data to warrant some of the conclusions (which had been made) concerning theoretical unsoundness of the process. Also, it was felt that if biologically inert materials did build up, it would be wise from an engineering standpoint to determine how long it would take for such a buildup to occur. It was reasoned that if two or three years were required for the system to undergo a functional biochemical failure, the process could still be more widely utilized, especially for higher strength industrial wastes. Thus, from the standpoint of theoretical concepts of biological engineering and from the standpoint of practical engineering utility of the extended aeration or total oxidation process, further work seemed warranted.

#### EXPERIMENTAL RATIONALE

The present work was conducted along four main lines of approach. The most important line of investigation consisted of the operation of an extended aeration laboratory pilot plant over a long enough period of time to determine when and if the system would break down biochemically.

It was essential to operate the system with complete and positive retention of biological solids. Another aim was to test, as the system aged, its responses to quantitative shock loadings. The third and fourth lines of investigations developed as the work proceeded. The results indicated that it was desirable to initiate experiments to determine the ability of various microorganisms in the activated sludge to metabolize various fractions of other microorganisms, i.e., studies of the natural causation for the auto-oxidation or autodigestion of the heterogeneous microbial population comprising the biomass were made. Also, it became advisable to investigate various ways and means by which biochemical engineering control over the biological solids concentration in the system might be effected. It was essential from the standpoint of drawing valid conclusions concerning the theoretical soundness or unsoundness of the process that close experimental control be maintained. For this reason, laboratory investigations using a defined synthetic waste, rather than field investigations using natural wastes, were employed.

#### I. Long-term Operational Stability of the Extended Aeration Process

Since it was expected that at various times during the pilot plant operation, just as one would expect in field operation, biological solids might experience settling difficulty, thus causing inadvertent loss of solids from the system over the clarifier effluent weir, positive means for retaining these biologically synthesized cells in the system was necessary. To accomplish this requirement, all effluent from the clarifier was routinely passed through a Sharples centrifuge, and any solids which would have exited the system were harvested from the

centrifuge and returned to the aeration chamber. During long-term operation of the pilot plant, various analyses for substrate removal and for biological solids concentration and composition were routinely performed. Microscopic observations were frequently made to assess any gross changes in the ecological makeup of the sludge.

Also routinely the substrate removal capability of the sludge was examined by performing batch experiments at high and low initial biological solids concentration to determine the pattern of substrate removal under batch as well as under continuous flow conditions. Also, the endogenous oxygen uptake of the sludge was determined periodically since it was felt that endogenous  $O_2$  uptake might provide a useful operational parameter for comparison with the results of the substrate removal studies.

## II. Responses to Quantitative Shock Loads

Shock loading studies were conducted under two general conditions. Under one condition, the extended aeration process was operated as a batch system. The base line performance under this condition of operation was established and, following this, the system was subjected to a series of shock loadings and the behavior of the system was compared with the base line behavioral pattern. Under the second condition, the extended aeration process was operated under continuous flow conditions and a base line behavioral pattern was established. This was followed by a series of shock loadings, and the response of the system was compared to the continuous flow operational base line. Throughout the study, auxiliary experiments were performed to determine substrate removal rates and endogenous oxygen uptake rates.



### III. Studies on the Autodigestion of Biological Solids in the Extended Aeration Process

Studies were undertaken to gain insight into the ease and/or difficulty with which various fractions or components of microbial cells may serve as carbon source for other cells in the system. Three major sources of possible organic substrate for "cannibalizing" cells are: (1) the soluble material contained inside a cell, (2) insoluble cell wall material, and (3) the slime layer or capsule surrounding the cell wall.

### IV. Possible Biochemical Engineering Control of the Biological Solids Concentration

It will be recalled that the centrifugation of the effluent was performed to maintain positive control over the loss of biological solids. However, in practice such an expedient would be impractical from an economical standpoint and it was reasoned that at times, biological solids concentration in the system might build up to such a point that settling in the clarification chamber would be greatly hampered because of the high solids concentration. It was hypothesized that at such times the system might be given a "chemical assist" by withdrawing some sludge from the bottom of the clarifier and rendering it soluble by chemical hydrolysis so that it could be returned to the aerator in soluble rather than insoluble form as substrate for the intact sludge.

## REVIEW OF RESULTS

The results of these experimentations are reported in detail in three published scientific articles (Appendices 1, 2, 3) and in a

Master's thesis (Appendix 4). The review of results which follows is intended to summarize the research findings and to cross-reference the material contained in the appendices. In the review which follows, frequent references are made to figures and tables in the four appendices. In all cases the reference is identified by figure or table number, and appendix number.

## I. Long-term Operational Stability of the Extended Aeration Process

### a. Performance Characteristics of the Extended Aeration Pilot Plant

The bench scale extended aeration pilot plant employed in this study is shown in Figure 1, Appendix 2, and a flow diagram is shown in Figure 2, Appendix 1. The total volume of the reactor was 9.4 liters (i.e., 6.2 liters aeration chamber, 3.2 liters settling chamber). During continuous flow operation, the feed rate was set to provide an overall detention time of 24 hours (approximately 16 hours aeration and 8 hours settling). During periods of batch operation, a 23-hour aeration period and a one-hour settling period were employed. Biological solids contained in the effluent were collected, centrifuged, and returned to the aeration chamber, i.e., there was no wastage of sludge except for a very small amount which was taken from the unit for various chemical analyses. The composition of the synthetic waste which was fed to the unit is shown in Table I of Appendix 1. A detailed procedure for initiation, sampling, and operation of the unit is described in Appendix 1. The general characteristics of the biological solids and the biochemical purification efficiency throughout the operation period (1000 days) are plotted in Figures 3 through 6 in Appendix 1, and Figures 2 through 5 in Appendix 3.

It is significant to point out that from day 285 to day 307 (see Figure 3, Appendix 1), the biological solids concentration decreased from approximately 8500 to 2400 mg/l. The filtrate COD which had been averaging approximately 40 mg/l (i.e., approximately 95 per cent COD removal efficiency), rose to 100 mg/l during the early period of the decrease in solids concentration, but soon returned to its previous level. This decrease in solids concentration was a more severe depression in solids than any of the other fluctuations which were observed. Again it is emphasized that the solids were not lost or wasted; also there was no operational change (pH, temperature, substrate concentration, etc.) which could have caused the decrease. This decreasing cycle in solids concentration appeared to be a natural one brought about by the biological system itself. If this decrease in solids were caused solely by lysis of cells with the lysed material not being used as substrate, the organic products released upon cell disruption or dissolution would have been manifested as a rise in filtrate COD. However, from the results shown in the figure, the COD removal efficiency was hardly interrupted during this phase, and it is apparent that one or more species of microorganisms in the system metabolized the released organic products.

Biological solids concentration remained at approximately 3000 mg/l for about one hundred days, then began a gradual rise until a concentration of 14,000 mg/l was attained at day 534 (see Figure 5, Appendix 1). During this period of operation, the biochemical purification efficiency was approximately 95 per cent. During the next five hundred days of operation (see Figures 5 and 6, Appendix 1, and Figures 2, 3, 4, and 5, Appendix 3), the biological solids concentration showed cyclic increases and

decreases. It fluctuated between 14,000 and 18,000 mg/l. During this period, the biochemical purification efficiency remained between 85 and approximately 100 per cent. Between days 630 and 1000 (see Figures 2, 3, 4, and 5, Appendix 3), the biological solids were sometimes carried out of the settling chamber and would have escaped in the effluent had they not been subjected to centrifugation. During the period when large amounts of solids were found in the settling chamber effluent, the pH in the system was experiencing a downward trend. Corrective measures were taken; larger amounts of phosphate buffer were added to the system. During this period, the biochemical efficiency was not interrupted.

It should also be noted that excess nitrogen source was present in the synthetic feed to the pilot plant ( $\text{COD/N} = 530/53 = 10/1$ ; see Table II in Appendix 1). Analyses run for ammonia, nitrate and nitrite nitrogen indicated that at times the sum of these forms of nitrogen in the effluent was equal, or nearly so, to the nitrogen concentration in the feed. Thus there was some indication that the nitrogen source in the system was at times totally reusable.

#### b. Substrate Removal Capability

In order to gain insight into the ability of the extended aeration activated sludge to remove substrate, as it was chronologically aged, two types of experiments were performed. In one, the continuous flow of waste to the pilot plant was stopped for a day and the unit was fed as a batch system. The unit was then sampled frequently to determine the rate of substrate removal. Also, a very small amount of cells was removed from the unit and used to study the growth characteristics and substrate removal ability of these seeding organisms when they were present at very

low initial concentration. The detailed procedures for these experiments are given in Appendix 4. In all experiments, biological solids concentration and analyses for chemical oxygen demand (COD) were made, and in some experiments, additional analyses were made using a specific determination for carbohydrate (the anthrone test). The calculation of specific substrate removal rates (mg COD/gr sludge/hr) is given in Appendix 4. Figure 18 in Appendix 4 shows a graph (upper portion) of specific substrate utilization rate vs. days of operation of the pilot plant for the first 720 days. It is seen that the specific substrate removal rate for both types of experiments (i.e., high and low initial solids concentrations) tended to follow similar patterns. The course of substrate removal and biological solids accumulation for these batch experiments from which the specific substrate removal rate was calculated are shown in Figures 19 through 53, Appendix 4. It is highly significant to point out here that the results of the batch experiments using high initial solids concentration indicated extremely rapid removal of the organic substrate which was fed, even though the unit was subjected to continual aging throughout the study. Purification was usually effected in 30 to 60 minutes, although the detention time in the aerator was sixteen hours during continuous flow and twenty-three hours during batch operation. Thus this extended aeration activated sludge, operated with total cell recycle, not only maintained excellent substrate removal efficiency over a prolonged period of operation and sludge aging, but indeed possessed and continued to possess throughout the experimentation a considerable metabolic reserve capacity. The results of the experiments at low initial solids concentration indicated that aging of the

sludge, as it is done in the extended aeration process, does lead to promulgation of cells with rather low specific growth rates. It was also found during the study that, during the course of substrate removal, accumulation of metabolic intermediates and/or endproducts was not appreciable. This observation is based upon comparison of the COD analyses and the anthrone analyses for substrate removal.

c. Endogenous Oxygen Uptake Rate of the Extended Aeration Sludge

Periodically, a very small amount of sludge was taken from the extended aeration system and its endogenous oxygen uptake was measured using the Warburg apparatus. In Figure 6, Appendix 3, values during the 1000-day period of operation are shown. The endogenous respiration rate is expressed as  $\text{mg O}_2/\text{hr}/\text{gr}$  sludge. The average of twenty-one determinations prior to day 250 was approximately 3.5. Just following day 250, the values began an increasing trend which corresponded to the period of decreasing cell concentration (see Figure 3, Appendix 1). Some time thereafter, the endogenous  $\text{O}_2$  uptake rate followed a decreasing trend as the biological solids concentration in the unit gradually increased. The decreasing trend appeared to become asymptotic to a low limit between 1 and 1.5  $\text{mg O}_2/\text{hr}/\text{gram}$ . It was determined by experimentation that this value for endogenous  $\text{O}_2$  uptake rate was between 5 and 10 per cent of that for young cells grown from a small inoculum of cells taken from the aeration chamber. Thus it can be concluded that the mature population which develops in the extended aeration process is less biologically active (at least by this parameter) than is a freshly grown "young" microbial population.

## II. Responses to Quantitative Shock Loads

The response of extended aeration activated sludge to quantitative shock loads is described in detail in Appendix 2. Figure 2, Appendix 2, shows the performance of the extended aeration activated sludge under both batch and continuous flow conditions of operation prior to administering shock loadings. It can be seen in Figure 2, Appendix 2, that both the concentration of biological solids and the filtrate COD attained a somewhat higher level under batch operation than under continuous flow operation. The high residual filtrate COD values observed during batch operation are attributable to the fact that only a small amount of supernatant was discarded under this type of operation. Thus the buildup of residual COD may be due at least in part to soluble organic metabolic products which are biologically non-degradable. During continuous flow this would normally go out with the effluent, i.e., in the 5-10 per cent COD in the effluent; it may also have been due in part to a buildup of organic salts from the synthetic waste.

The response of the extended aeration activated sludge under both batch and continuous flow conditions to quantitative shock loadings is shown in Figure 3, Appendix 2, and as adjudged by the filtrate COD it can be seen that increasing the feed COD from 500 to 2500 mg/l, carbon source did not adversely affect the removal efficiency. Thus the system must be judged capable of accommodating rather severe shock loading conditions.

## III. Studies on the Auto-digestion of Biological Solids in the Extended Aeration Process

The results of experimentation involving long-term operation of the extended aeration process without sludge wasting provided evidence that

the concept of the total oxidation process was not inconsistent with sound microbiological principles. The results indicated that biological solids concentration would not build up continuously; there were periods of increases followed by periods of decreasing solids concentration. The only interpretation which could be placed upon the periods of decreasing biological solids concentration was that at these times, portions of the sludge which had already been synthesized were serving as food material (carbon source) for other cells in the system. Since there was no evidence for a buildup of an inactive fraction, it could only be concluded that the proteins, carbohydrates, fats, nucleic acids, etc. of various species were metabolizable by other species in the system. It became important, therefore, to gain some insight into the relative metabolic availability of certain cell fractions.

Some experiments of this type are described in detail in Appendix 3. A number of experiments were run in which the soluble cell components (i.e., those released upon cell breakage by ultrasonic vibration) were used as carbon source for cells harvested from the extended aeration unit or for cells developed freshly from municipal sewage. In some cases there was a noticeable lag period followed by rapid growth, whereas in others the cell sonicate was removed without an acclimation period but at a somewhat slower rate. The results of one experiment on cell sonicate is shown in Figure 8, Appendix 3. It can be seen that approximately 50 per cent of the initial COD was removed very rapidly, i.e., within thirty minutes. Most of the remaining COD was removed within ten hours. Approximately 90 per cent of the cell sonicate was subject to biological purification. Studies on the biological availability of other cell components or fractions are currently under way.



#### IV. Possible Biochemical Engineering Control of the Biological Solids Concentration

Experimentation during the first two years of the project indicated that while the biological solids concentration did not build up continually, and that while there were downward or decreasing cycles in cell concentration, the amount of sludge held in the system at any one time might reach such high values as to militate against settleability and compaction in the settling chamber. In field practice, one could expect that at such times sludge might be lost in the effluent. The experimental results, coupled with continual observation of the behavioral patterns in the unit (both macroscopic and microscopic), indicated the important role of microbial ecology. It could be expected that biological solids might build up until a balance of species was reached which would permit some species to become substrate for others. One could not predict the periodicity of the increasing and decreasing cycles in sludge concentration, but one would expect that one of the most difficult operations to accomplish biochemically was the solubilization of cell walls and capsular or slime materials. It was reasoned that one might be able to enhance the autodigestive process by performing the solubilization chemically rather than biochemically. Thus it was reasoned that during periods of high biological solids concentration one might withdraw cells from the underflow of the clarifier, hydrolyze the sludge, and return the solubilized biological mass to the aeration chamber as a source of soluble rather than insoluble carbon source. Various conditions of hydrolysis and hydrolyzing chemicals were tried, and the results of preliminary experimentations indicated that acid hydrolysis under rather

mild conditions (120°C, 15 psi, pH 1.0) did indeed render biological sludge soluble. When the hydrolyzed sludge was fed to the extended aeration sludge, it was found that the system experienced no difficulty in metabolizing the chemically prepared substrate. These experiments are described in Appendix 3 (see Figure 9). Approximately 50 to 60 per cent of the COD of the hydrolyzed sludge consisted of proteinaceous material and 8 to 15 per cent carbohydrates.

The results of these experimentations indicated to us that the proposed extended aeration activated sludge process incorporating chemical hydrolysis for controlling solids concentration which is shown in Figure 10, Appendix 3, provides a very useful engineering modification to the extended aeration process.

#### SUMMARY, DISCUSSION, AND CONCLUSIONS

Previous researchers had concluded that the extended aeration or total oxidation activated sludge process was, from a biological standpoint, theoretically unsound, because the microbial cells developed in response to feeding waste waters to the system could not, in their entirety, be used as carbon sources for other microbial cells. Thus, as time went on and the system became chronologically older, the inert organic fraction (inert in the sense that it could not be used as substrate and inert in the sense that it consisted of dead organic matter incapable of removing substrate) would continually build up and become a sufficiently large percentage of the biological solids in the system so as to cause eventual biochemical failure. Thus, since the early days of its conception, the process has carried with it the onus of theoretical

unsoundness. Regardless of this, the process has enjoyed increasing use and has thus represented somewhat of a paradox in the area of pollution control engineering experience. One of our aims at the outset of the present research was to determine how long it might take before the system would eventually fail. After three years of continual operation of a pilot plant under highly controlled conditions with positive control over sludge retention in the system, we have determined through our observations and analysis of the data that the system is not theoretically unsound from a biological standpoint, and there is no evidence for the continual buildup of a biologically inert fraction of the activated sludge due to metabolically inaccessible cell components. The system does not operate at solids equilibrium, as was suggested by the originators of the process, but undergoes periodic cycles of sludge accumulation and of de-accumulation. The system undergoes periodic changes in species predominance and, at times, species which can metabolize other species predominate in the system. At times, the species capable of being cannibalized represent a major source of food material, and at these times the digestion of solids exceeds new growth on the substrate in the incoming wastes and the biological solids concentration decreases. The extent of the accumulation and de-accumulation cycles in biological solids and the periodicity of the cycles is not predictable, and a way and means of exerting engineering control over the concentration of biological solids in the system was sought. Our investigations indicated that the soluble portions of the cells in activated sludge were amenable to biological attack. It was reasoned that the most difficult metabolic activity for one cell to perform upon another was solubilization of the

insoluble fractions of the cell, e.g., cell walls and cell capsules of slime material. Therefore, whole cells (or portions of the activated sludge) were hydrolyzed chemically under conditions which solubilized most or all of the cell. It was found that, after neutralization of the solubilized cells, the material could be fed to intact cells in the system, and this material served as an excellent source of organic carbon. Thus an engineering process modification to the extended aeration process was envisioned, and preliminary tests indicated that the type of flow sheet shown below is highly recommendable as a means of exerting engineering control over the operation of an extended aeration process.

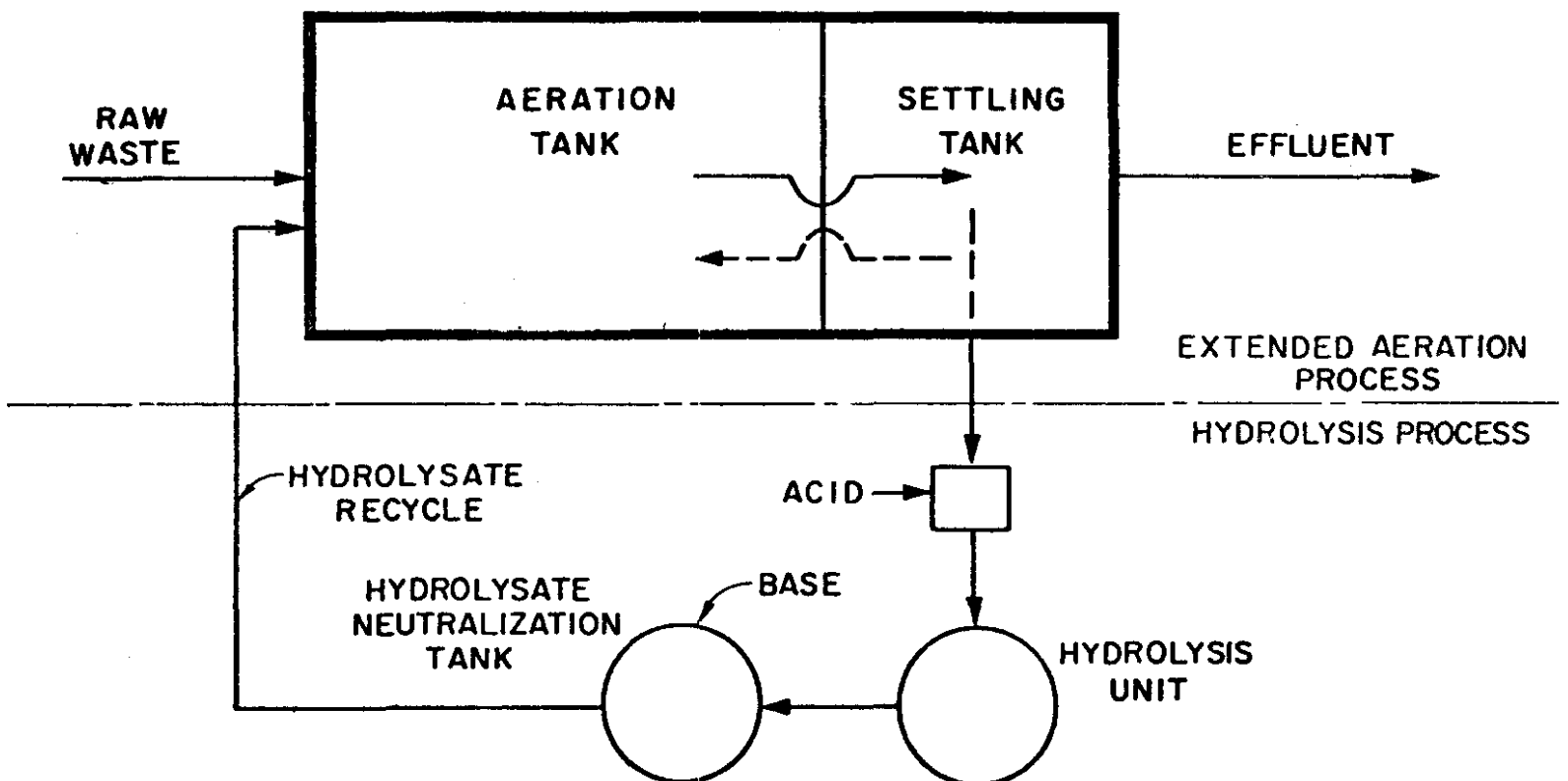


Figure 10, Appendix 3

This process, which embodies a "chemical assist" to the biological process, accomplishes chemically a function which is biologically difficult and accomplishes biologically a function which is difficult and costly to do chemically. The hydrolysis portion of the modified extended aeration treatment plant can be operated intermittently, i.e., when biological solids concentration is deemed too high or is so high as to interfere with settling in the settling chamber. The system we have recommended appears to have many advantages. The potential of the system is discussed more fully in Appendix 3.

In general, it is felt that the conclusions which it has been possible to make through the conduct of this research project have fulfilled the primary objective of the proposed research. Had we accomplished short-term experiments and not been interested initially in the determination of how long it might take before an extended aeration process exhibited a biochemical or functional failure, our findings concerning the theoretical soundness or unsoundness of the process in all probability would have been very similar to those of other workers who have studied the process previously. However, our long term studies under highly controlled conditions have shown that biological solids will not continually build up in the system, and that there is no evidence for a gradual buildup of an inert fraction of the biological solids. These findings provide a more sound theoretical basis for the extended aeration process or total oxidation process in general and, when coupled with our recommended modification, i.e., the chemical hydrolysis of a portion of the activated sludge, offer considerable promise not only for enhancement of the secondary biological treatment process, i.e., removal of organic

matter from waste waters, but also for disposal of biological sludge developed during the treatment process. This work has been extremely useful and has also pointed the way to further studies on the engineering process we have recommended, and to engineering science research on metabolism of activated sludge.

**APPENDIX 1**

**Gaudy, A. F. Jr., Ramanathan, M., Yang, P. Y., and DeGeare, T. V.,  
"Studies on the Operational Stability of the Extended Aera-  
tion Process." Journal Water Pollution Control Federation,  
42, 165-179 (1970).**

## STUDIES ON THE OPERATIONAL STABILITY OF THE EXTENDED AERATION PROCESS

By

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In a previous report the responses of a laboratory scale "extended aeration" plant to various levels of quantitative shock loading were presented (1). It was shown that an extended aeration activated sludge could accommodate a five-fold increase in concentration of inflowing organic loading without retardation of purification efficiency measured as chemical oxygen demand (COD) of the filtered reaction liquor. The previous work was undertaken in order to gain an insight into the operational stability of this process when subjected to an environmental change (quantitative shock loading) commonly encountered in field operation. Although the response of the extended aeration process to shock loadings is an important concern, there is another of even greater significance relative to the use of this process. The suggestion by Porges, et al. (2) that an activated sludge plant could be operated with return of all biological solids to the aerator and that a point would be reached wherein net autoxidation would balance net synthesis of new biological sludge, permitting the system to operate at or near some

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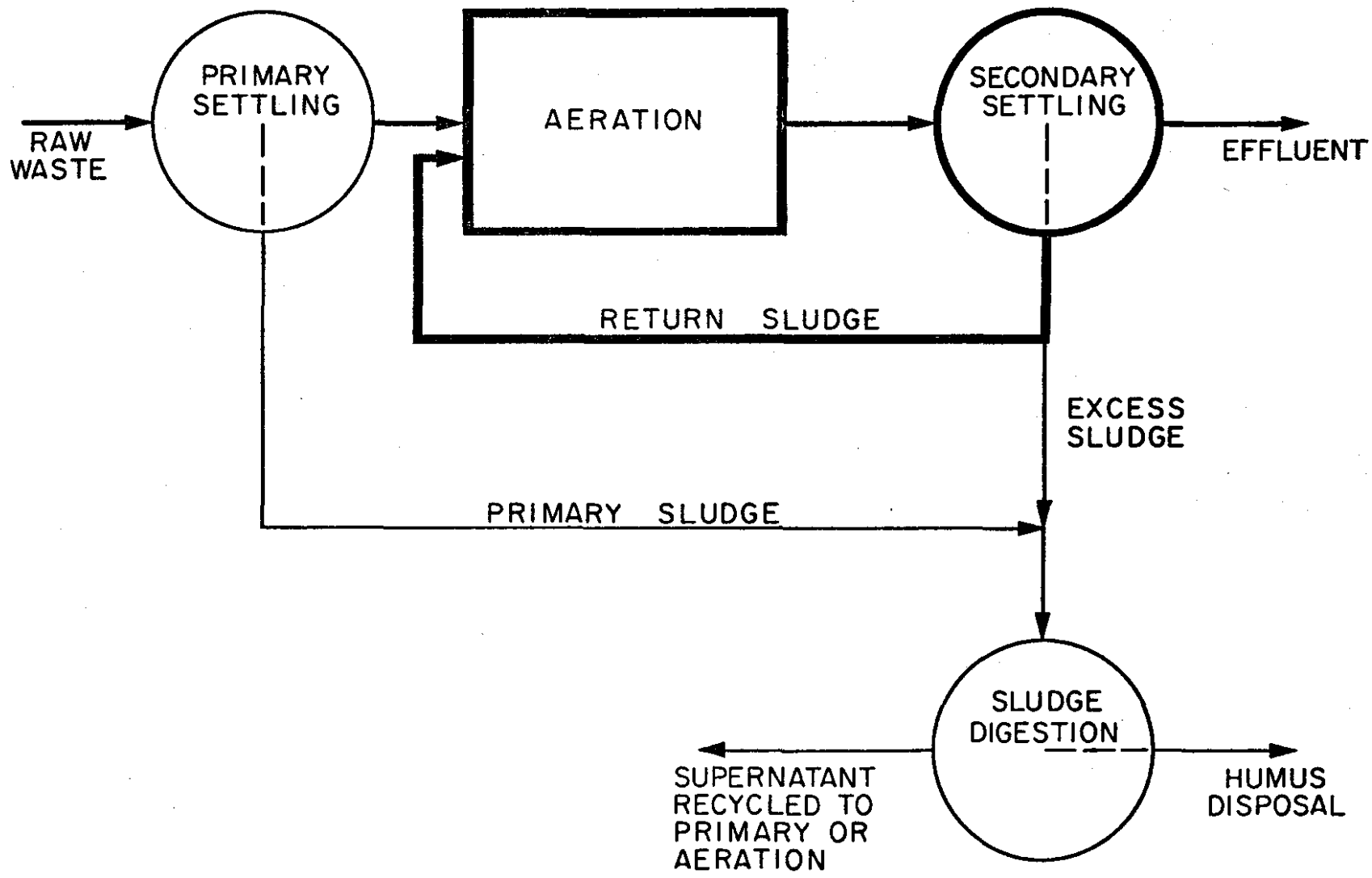
Presented at the 20th Oklahoma Industrial Wastes and Pollution Control Conference, Stillwater, Oklahoma, March 31-April 1, 1969.



equilibrium biological solids concentration, stimulated a considerable amount of research on the process. This research was indeed warranted because, if such a process could be successfully operated, it would have considerable advantages over other modes of operation of activated sludge plants. These advantages are immediately apparent when the simplified flow diagram for standard activated sludge shown in Figure 1 is considered. For the extended aeration process, only the portion of the diagram shown in heavy lines is required. Facilities for the treatment (aerobic or anaerobic) and disposal of excess sludge comprise a major portion of the initial and operational costs of the treatment plant. Elimination of these facilities has apparent advantages. The cost entailed in providing the somewhat longer detention time in the aeration tank needed for the concomitant metabolism of incoming substrate and biologically synthesized material in the reactor is less than that for the usual facility for separate sludge treatment and disposal. Some of the research which has been accomplished on this process was reviewed in a previous paper; the following citations bring into focus the prevalent opinion of workers in the field concerning the process, delineate its present status, and indicate the reasons for initiating the present experimental research effort.

It is generally accepted that in theory "total oxidation" is not possible, since a portion of the soluble organic carbon in the waste which was channelled into synthesis of microbial cells is permanently resistant to biological attack (3)(4)(5)(6). Thus, if no sludge is wasted, the inactive, non-usable organic fraction in the sludge gradually increases and the system must ultimately fail. Also, because of the existence of this aerobically undigestible fraction, the concentration





of sludge in the system cannot come into a true steady state equilibrium; i.e., organic solids will continue to accumulate in the system until failure occurs or until solids are inadvertently or purposely wasted. It has been generally concluded that some sludge must be periodically wasted in order for the extended aeration process to succeed. Indeed, in the course of some of the research which has been conducted on the process, it has been noted that at times biological solids were lost in the effluent. The general impression which is gained from the literature is that in cases where sludge has not been intentionally wasted, successful purification of the waste has been made possible due to unintentional solids losses in the effluent. As pointed out by Pfeffer (7), the problem of accumulation of sludge and the nondegradable fraction thereof is complicated when municipal wastes are considered, since they contain varying quantities of particulate organic and inorganic materials which may be incorporated into the sludge. However, most of the basic engineering research which has been accomplished to investigate or determine the validity of the total oxidation hypothesis has used soluble organic matter, and the crux of the problem regarding the theoretical validity of the concept of total oxidation resides in the reported nondegradability of a portion of the biological solids which have been synthesized during the purification process.

Regardless of apparently well-documented theoretical reasons for expecting the process to fail, it has been used to an increasing extent. However, were it not for theoretical objections, this method of treatment might be more widely employed, especially for soluble industrial wastes. In assessing the problem it was our conclusion that the main question yet to be answered concerning extended aeration processes was:

How long could such a system, in which all biological solids were returned to the aerator, be operated before the "inactive" fraction built up to such an extent that the biochemical efficiency of removal of a reasonable organic loading deteriorated, causing failure of the system? In posing this question, the type of failure envisioned was a biochemical failure, that is, loss of substrate removal efficiency, not loss of biological solids in the effluent. The latter type of failure is indeed a serious one, but it is one which is subject to possible correction through application of various engineering expedients, whereas biochemical failure is a mechanistic one which is far more critical.

In making the studies herein reported, it was essential to provide for more positive retention of biological solids than could be provided by plain sedimentation, since it was expected that sludge settleability (as the sludge concentration built up) would become as much a problem in our laboratory studies as it is known to be in the field or as it has been proven to be in other laboratory investigations. In preliminary experimentation, a "cellophane bag" type reactor in which a plastic membrane, permeable to the soluble substrates but not to the cells, was employed. Such a reactor would be ideal for studies involving total cell retention; however, operational problems were encountered and, rather than expending investigational effort in the solution of these particular problems, a decision was made to employ a more conventional reactor and settling chamber, but to pass the effluent through a centrifuge and return all cells carried over in the settling tank effluent to the aerator. At the initiation of the investigation, the primary purpose was to determine how long biological solids would continue to accumulate in the system and when biochemical failure would occur, and

to determine from various subsidiary experiments and analyses on the extended aeration system itself whether the onset of biochemical failure could be detected prior to actual deterioration of substrate removal efficiency.

#### MATERIALS AND METHODS

The bench scale extended aeration pilot plant employed in these studies was essentially the same as the one used in the shock loading studies reported previously (1). A flow diagram is shown in Figure 2; a more detailed drawing of the movable wall separating the aeration and settling chambers is presented elsewhere (1). For the present study, the position of the wall provided for an aeration chamber volume of approximately 6.2 liters, and settling chamber volume of approximately 3.2 liters. During continuous flow operation the feed rate was set to provide an overall detention time of 24 hours (approximately 16 hours aeration and 8 hours settling). During periods of batch operation, the separating wall was removed and the unit was fed once daily. During such periods of operation a 23-hour aeration period was employed. After 23 hours of aeration a sample was taken, the mixed liquor was permitted to settle for one hour, one liter of supernatant was removed and centrifuged, and any biological solids in the supernatant were returned to the unit in one liter of feed solution, bringing the system back to the operating level (9.4 liters). The composition of the synthetic waste shown in Table I is that which was fed during continuous flow operation. During periods of batch operation the concentration of each constituent was increased 9.4-fold. The organic loading employed was approximately 50 lb COD/1000 cu ft aeration capacity (32 lb BOD<sub>5</sub>/1000 cu ft) during continuous flow operations, and 32 lb COD/1000 cu ft (22 lb BOD<sub>5</sub>/1000



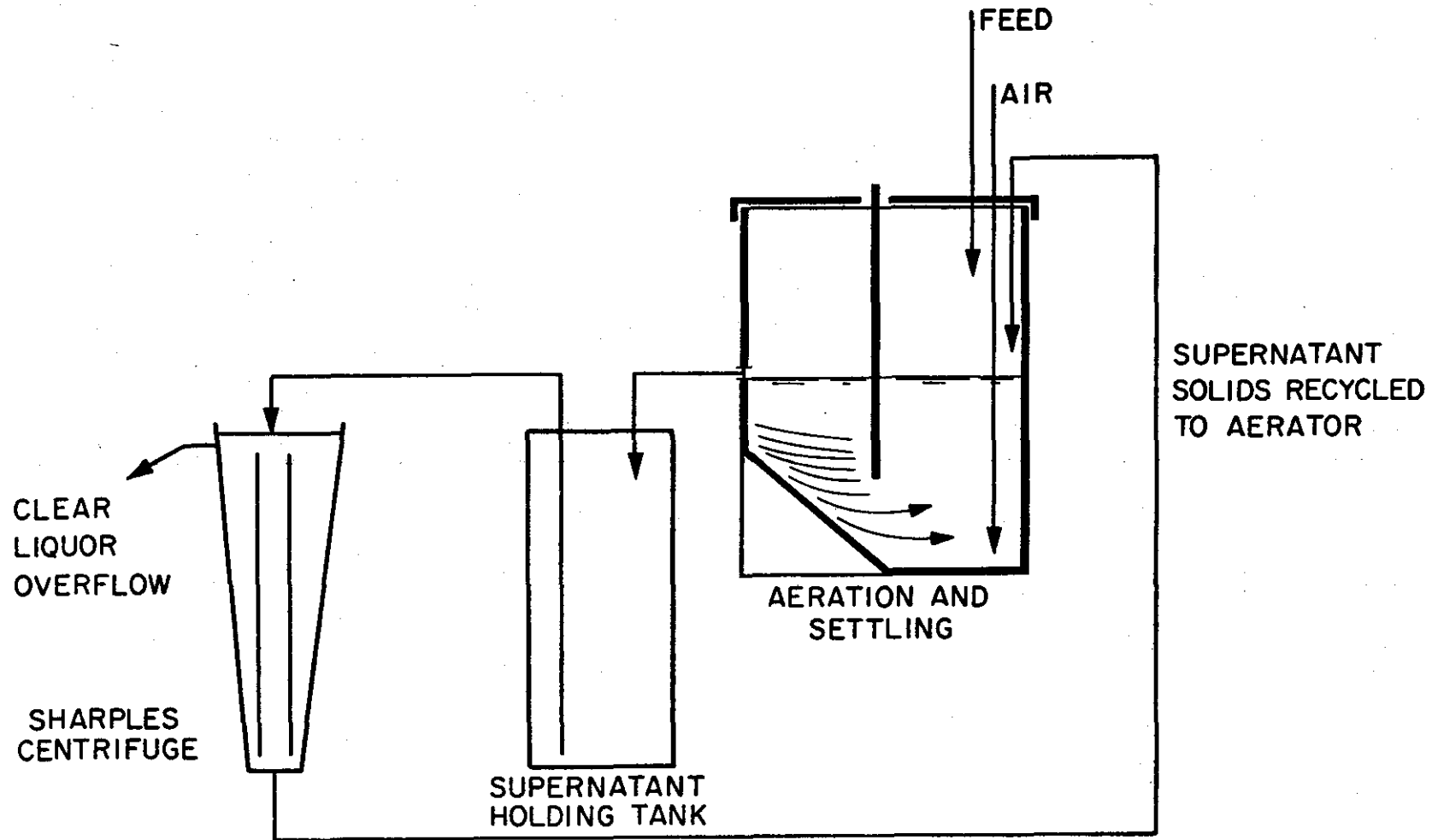




TABLE I  
COMPOSITION OF FEED FOR 500 mg/l GLUCOSE  
AS SUBSTRATE

Glucose	500	mg/l
$(\text{NH}_4)_2\text{SO}_4$	250	mg/l
$\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$	50	mg/l
$\text{FeCl}_3$	0.25	mg/l
$\text{CaCl}_2$	3.75	mg/l
$\text{MnSO}_4 \cdot \text{H}_2\text{O}$	5	mg/l
Phosphate Buffer, 1.0 M ( $\text{KH}_2\text{PO}_4 + \text{K}_2\text{HPO}_4$ ), pH 7.0	5	ml/l
Tap Water	50	ml/l

cu ft) on the basis of total volume of the system (or aeration volume) during batch operation. During continuous flow operation, sterile feed solution was channelled to the aerator through a dual unit positive displacement pump arrangement (Minipump - Model MM2-B-96R). One unit was either being cleaned (sterilized) or on standby while the other was in use. The feed reservoir was changed and sterilized daily. There were never signs of contamination; thus the nature and concentration of feed remained constant. The effluent from the settling chamber was collected in the holding tank (see Figure 2). Periodically (once or twice daily) the contents of the holding tank were passed through the centrifuge and any solids which had been carried over from the settling chamber were scraped from the bowl and returned to the aeration chamber. Thus, throughout the study no biological solids were either inadvertently

or intentionally lost from the system. The only solids "lost" from the system were those taken for samples to assess operational behavior.

Daily (or nearly so), 15 ml samples of mixed liquor were removed for analysis. At less frequent intervals, small samples were removed for measurement of endogenous  $O_2$  uptake and periodic examination of substrate removal rate. The removal of 15 ml of mixed liquor daily represents a solids removal of 0.16 per cent  $\left(\frac{15}{9400} \times 100\right)$ . Allowing for removals for the other purposes listed above, the total amount of solids removed from the system is estimated to be no more than 0.2 per cent. It should be noted that the biological solids concentrations which were measured represent the concentrations prevailing in the total system (aeration and settling chamber), not those in the aeration chamber alone. A few samplings from both chambers showed that at times the biological solids concentrations in both compartments were approximately the same, and at times they were rather widely divergent. The procedure which was adopted for sampling was as follows: Just prior to removing a sample, the settling chamber outlet was stoppered and the feed was momentarily shut off. The dividing wall was then lifted and the contents of the settling and aeration chambers were allowed to mix. The 15 ml sample was then pipetted from the tank and the dividing wall was again inserted into place. The settling chamber effluent port was then reopened and the feed restarted. Any solids which overflowed to the holding tank before subsidence in the settling chamber took place were then centrifuged and returned to the aerator. This procedure allowed a direct assessment of the course of daily solids accumulation in the system. Biological solids concentration was determined using the membrane filter technique (8). Chemical oxygen demand (COD) was also run on the

filtrate (8). At periodic intervals, COD's were also run on the effluent from the settling chamber. Protein and carbohydrate contents of the sludge were determined using, respectively, the biuret and anthrone (9) tests. Endogenous  $O_2$  uptake of the sludge was determined with a cell suspension obtained from a 15 ml sample of reactor mixed liquor, and washed twice in 0.1 M phosphate buffer solution. Periodically, substrate removal rate of the sludge was determined in the bench scale pilot plant. During periods of batch operation this was accomplished by sampling at frequent intervals after feeding the unit. During periods of continuous flow operation the feed flow was cut off, the unit was fed as a batch system, and the course of substrate removal was assessed. Continuous feeding was then resumed. Periodically throughout the experimentation, dissolved oxygen in the aeration chamber mixed liquor and in the settling chamber effluent was measured electrometrically. Also frequent checks on the pH were made. The airflow to the system was maintained at 2000 cc/min/liter. Temperature was maintained at  $23 \pm 2^{\circ}$ .

## RESULTS

Development of the extended aeration activated sludge used in the present experimentation began on March 31, 1967, at which time the bench scale pilot plant was put into operation, using seed from the primary clarifier effluent from the Stillwater municipal treatment plant. Since at the time of preparing this report the unit has been in operation for nearly two years and massive amounts of data have been collected, it seems of little value to show all of the daily results for this period. Also, as described in the previous report on the shock load response of extended aeration activated sludge (1), the initial

system was divided in order to obtain sludge for the shock loading studies. However, in the interest of completeness, a brief summary of the history of the sludge from its initiation until a portion of it was taken for separate shock load studies is warranted.

At the initiation of experimentation, the feed consisted of 1000 mg/l glucose, and the salts concentrations shown in Table I were appropriately increased. During the first thirty days of operation the biological solids concentration gradually rose to slightly over 6000 mg/l, and then gradually decreased to 4700 mg/l by the thirty-ninth day of operation. It is emphasized that the decrease in solids was not due to loss of solids in the effluent, since all effluent was passed through the Sharples centrifuge and cells not settled in the settling tank of the extended aeration unit were returned to the aeration chamber. Also, there was no disruption in COD removal efficiency; it remained close to 95 per cent throughout this period. From day 40 to 70, biological solids concentration rose steadily to approximately 12,000 mg/l. Throughout this period the purification efficiency, based on either filtrate COD or settling chamber supernatant COD, was approximately 95 per cent.

Between days 70 and 80, biological solids concentration decreased to approximately two-thirds of its previous value, i.e., to 8500 mg/l. Unlike the previous instance of decreasing solids concentration, there was a rise in the solids concentration in the effluent, and a rise in filtrate COD. During this period, COD removal efficiency dropped to between 80 and 85 per cent. Again, it is emphasized that this drop in solids concentration was not due to wasting of solids (either purposely or accidentally). During the period of decreasing solids, a small

amount of froth (approximately one inch in depth) was noted on the surface of the mixed liquor in the aeration chamber, and it appeared that a portion of the population had undergone a gradual lysis. Solids concentration levelled off at approximately 8500 mg/l, and within five days (by day 85) the COD removal efficiency had returned to 95 per cent. During the next few months of operation the solids concentration continued to rise gradually, and COD removal efficiency varied between 90 and 95 per cent. At times, settleability problems were encountered but, as always, the biological solids were centrifuged and returned to the aerator.

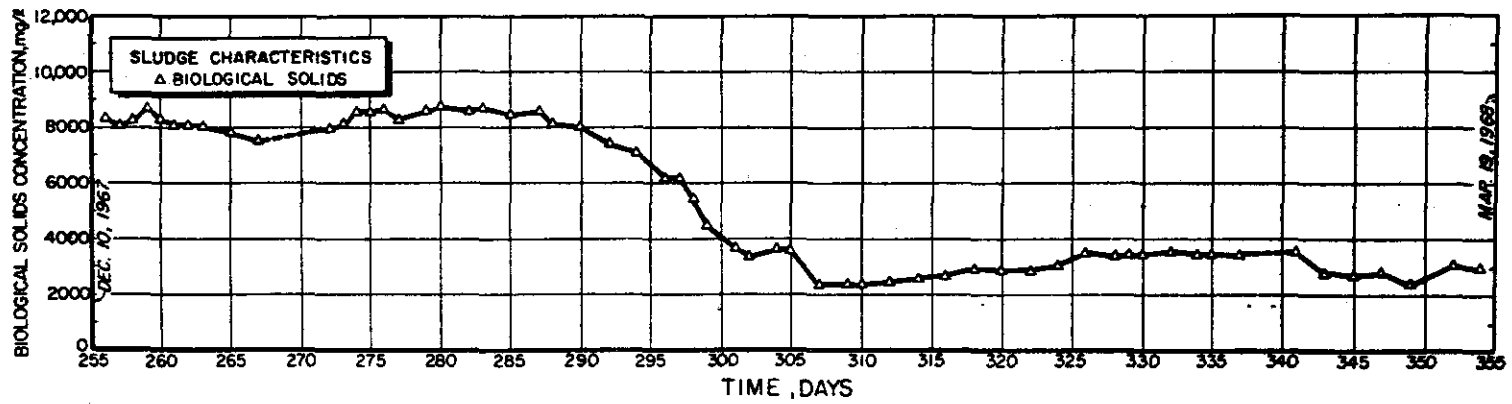
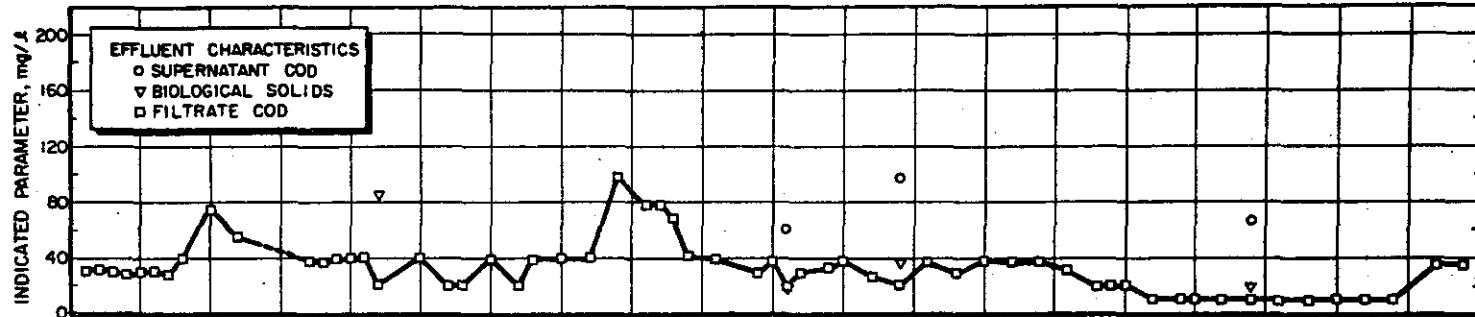
Beginning on September 5, 1967 (day 159), the mode of operation was changed from continuous flow to batch. This was done partially as a convenience and partially because it seemed desirable to observe the behavioral patterns under once-daily rather than continuous feeding. The total daily organic loading was maintained constant. By the 192nd day of operation (October 8, 1967), the biological solids concentration had built up to nearly 30,000 mg/l. At this concentration the sludge did not settle, but the biochemical efficiency of the unit was not impaired. At this time plans were made to divide the sludge in order to run another total oxidation unit so that the shock load responses of such a sludge could be studied. The results of those studies have been reported previously (1). On day 193 there was a slight decrease in biological solids concentration in the unit, and again a small degree of frothing was noted on the surface of the aeration liquor. By day 196 the solids concentration had decreased to 17,600 mg/l. Thus in a period of four days the system experienced a one-third decrease in biological solids concentration--a change equal in magnitude to that

which had previously occurred; substrate removal efficiency remained high. The population (or a portion thereof) appeared to be undergoing lysis, and the lysed products were metabolized by the remaining cells.

On October 12, 1967 (day 196), the sludge was divided and approximately half of it was used to initiate the unit employed for the shock loading studies previously reported. The original unit was operated on a batch feeding cycle until December 4, 1967 (day 249), at which time continuous flow was resumed. In order to maintain a relatively constant organic loading (on the basis of COD per unit weight of sludge in the original unit), the feed concentration was halved; i.e., 500 mg/l rather than 1000 mg/l was fed. From this time forward the performance data are shown in Figures 3, 4, 5, and 6.

In mid-January, 1968 (see Figure 3), the system began another cycle of decreasing solids concentration. From day 285 to 307, the biological solids concentration decreased from approximately 8500 to 2400 mg/l. The filtrate COD which had been averaging approximately 40 mg/l (i.e., approximately 95 per cent COD removal efficiency) rose to 100 mg/l during the early period of the decrease in solids concentration, but in less than a week returned to its previous level. This cyclic decrease in solids concentration was a more severe depression in solids than the ones previously observed, but it was more gradual, requiring over twenty days. Again, it is emphasized that the solids were not lost or wasted; also there was no operational change (pH, temperature, etc) which could have caused the decrease. The cycle appeared to be a natural one brought about by the biological system itself. As before, there was during this period a slight froth on the surface of the mixed liquor. The general appearance was one observed when microbial cells undergo





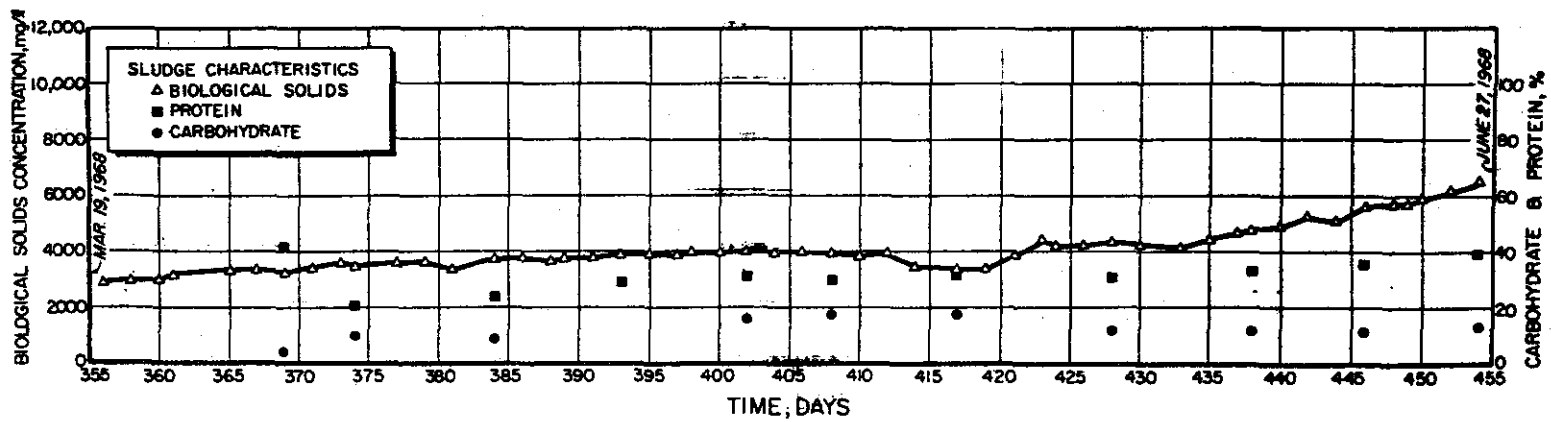
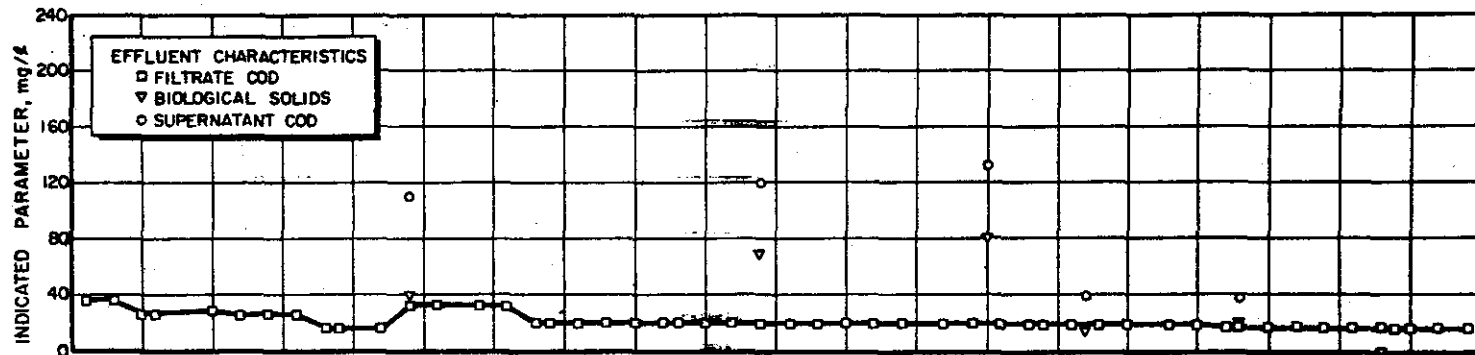


lysis. It is apparent that if the lysis did occur, the organic products released upon cell disruption or dissolution were metabolized by the intact cells, since the COD removal efficiency was hardly interrupted during this phase.

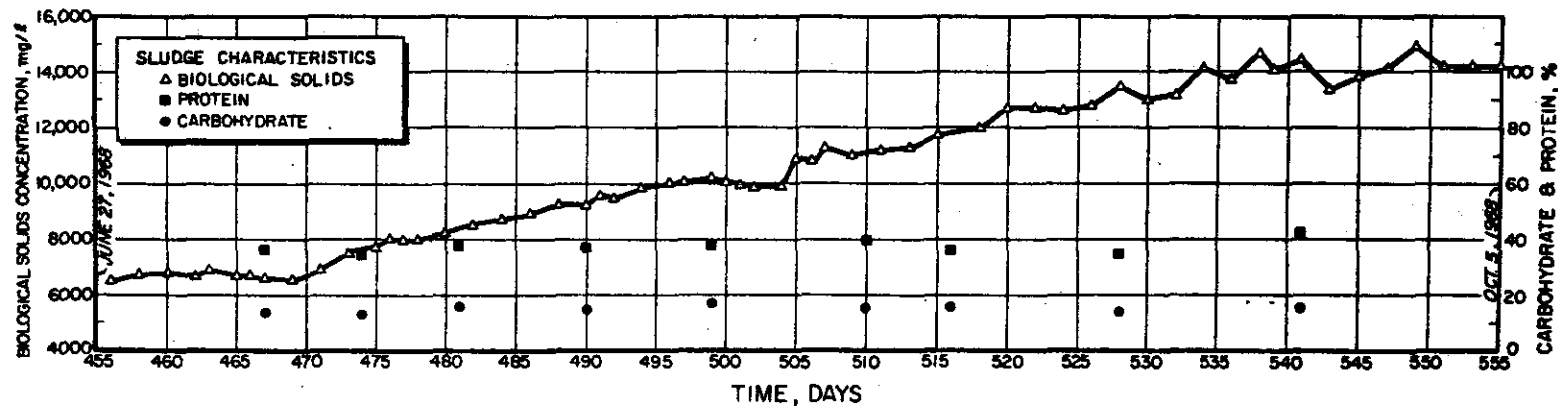
The biological solids concentration remained at approximately 3000 mg/l until late March, and then (see Figure 4) began a gradual rise during the next one hundred days of operation. During this time it can be seen that the filtrate COD remained very low (approximately 95 per cent removal efficiency). Spot checks on the biological solids concentration and COD concentration in the supernatant showed that at times the overall efficiency was approximately 80 per cent or below (see days 379, 404, and 420), and at times the overall efficiency was close to or as good as biochemical efficiency (see days 427, 438, and 446). The protein content of the sludge was approximately 30-35 per cent, and carbohydrate content ranged between 10 and 20 per cent.

Figure 5 shows that during the next one hundred days of operation (day 455 to day 555) the biological solids concentration continued to rise and a value of 14,000 mg/l was attained on October 5, 1968 (day 555). Both the biochemical and total purification efficiency over most of this period was excellent. Up to day 540 there was essentially no solids carryover in the settling chamber effluent. Sludge composition with respect to protein and carbohydrate content remained essentially the same as for the previous 100-day period. The characteristic behavior of the process from October 5, 1968, to January 13, 1969 (day 555 to day 655) is shown in Figure 6. The biological solids concentration followed a generally increasing trend through the fall of 1968, levelled off at approximately 17,000 mg/l, and early in 1969 began a slight

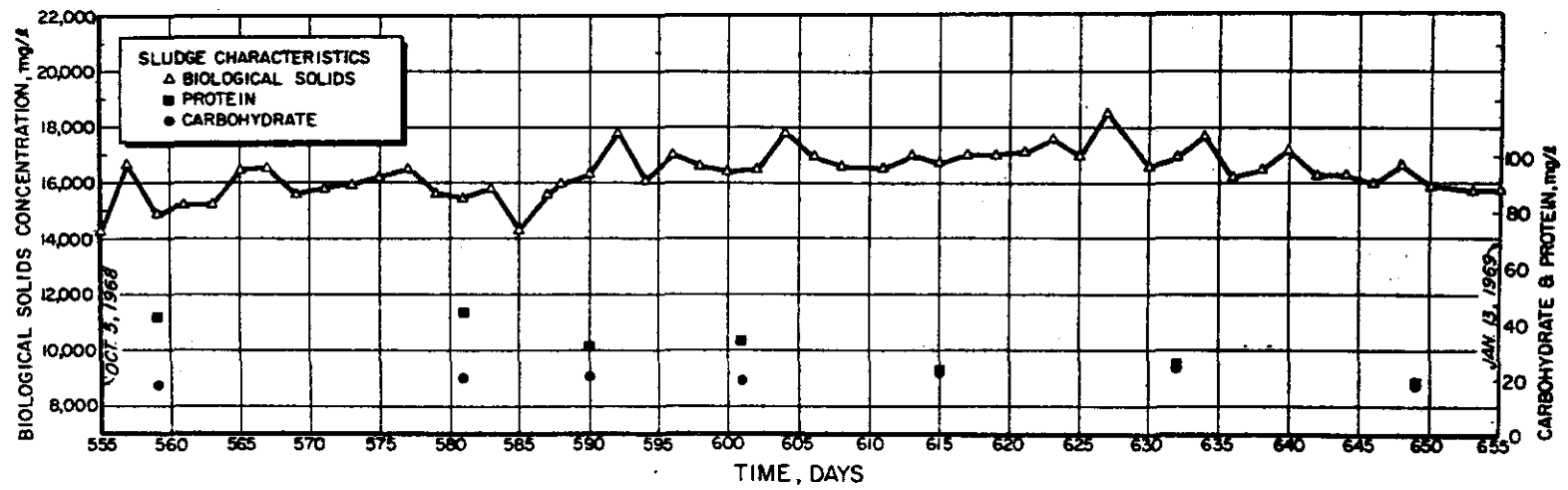
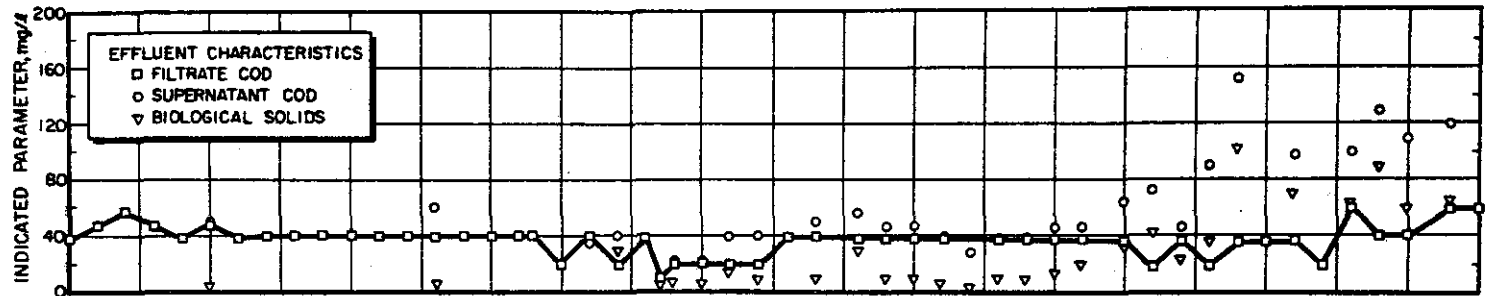












decline. Through the fall and winter months the biochemical efficiency remained at approximately 90 per cent, and sludge settleability and COD removal efficiency were excellent, as may be seen by the low supernatant COD and the biological solids concentration in the effluent. Late in 1968 the solids carried over in the settling tank supernatant rose; again, it is emphasized that the solids were not lost but were harvested by centrifugation and returned to the aeration chamber. It is interesting to note that during the late part of 1968 and on into 1969 the protein content of the sludge decreased to between 20 and 25 per cent, whereas the carbohydrate content approached 20 per cent. At the time of preparing this manuscript, January 27, 1969, biological solids concentration in the unit was 14,600 mg/l.

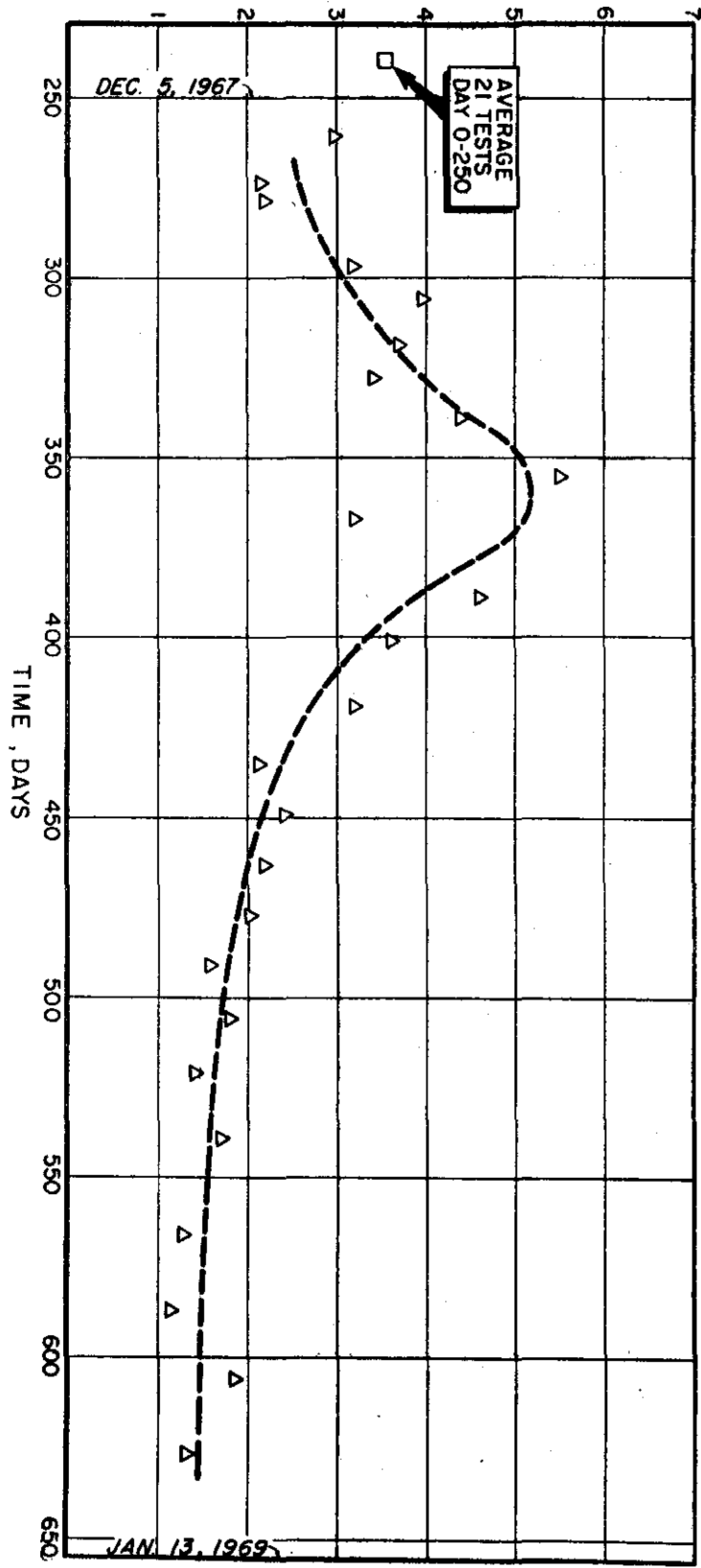
At various times throughout the experimentation a small sample of sludge was taken for measurement of its endogenous respiration rate (expressed as mg O<sub>2</sub>/hr/gm sludge). The values obtained are plotted in Figure 7. The average of twenty-one determinations prior to day 250 was approximately 3.5 mg O<sub>2</sub>/hr/gm sludge. Just prior to the beginning of the downward cycle in solids concentration, the values varied between 2 and 3, and there was a rising trend during the period of decreasing solids concentration (Figure 3), and for some time thereafter. The endogenous O<sub>2</sub> uptake rate followed a decreasing trend as biological solids concentration gradually increased and appeared to become asymptotic to a lower level between one and 1.5 mg O<sub>2</sub>/hr/gm.

After attainment of this low endogenous rate it was of interest to determine the endogenous rate of new or young cells grown up from a small inoculum of cells taken from the unit. Endogenous rates between 18 and 10 mg O<sub>2</sub>/hr/gm of sludge were observed. Thus, the endogenous



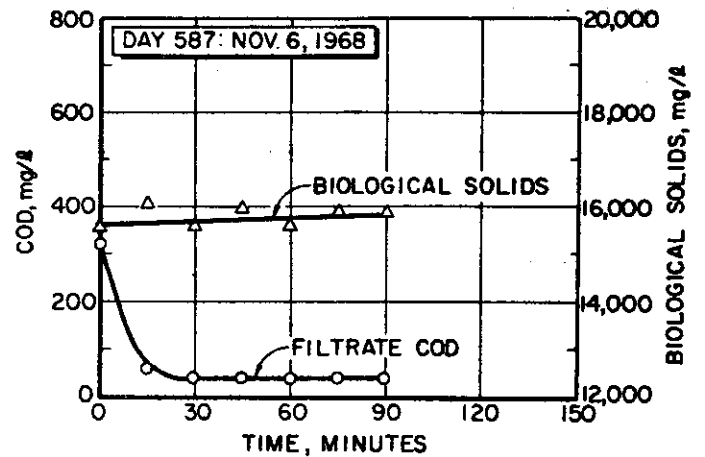
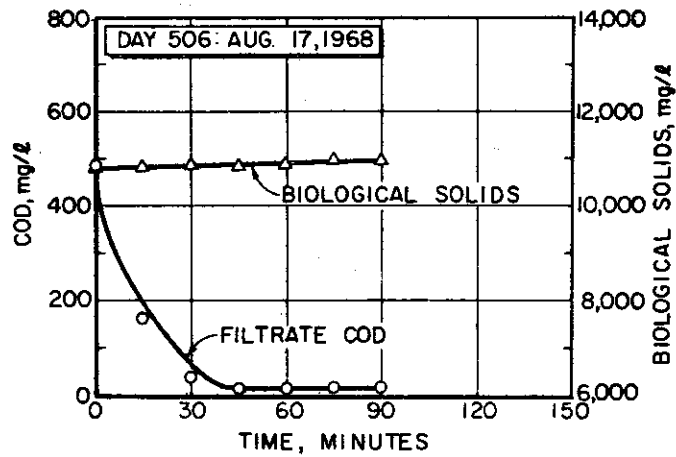
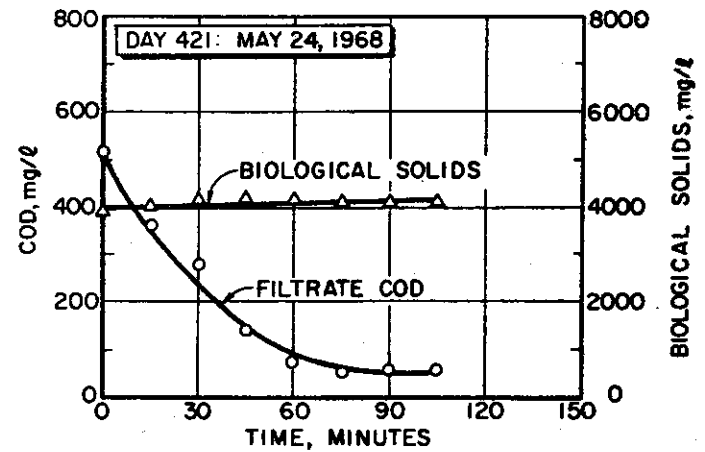
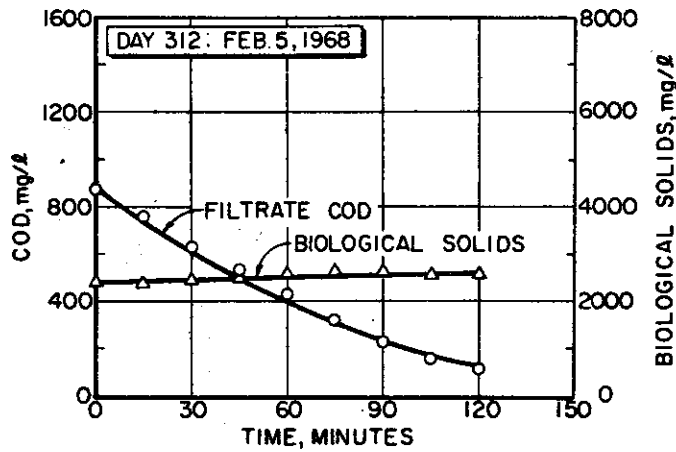


Mg O<sub>2</sub> PER HOUR PER Gm SLUDGE



activity of the extended aeration sludge is approximately 5 to 10 per cent of that for young cells of the same origin. At approximately two to three-week intervals the feed pump was stopped and the feed was added on a batch basis in order to assess the COD removal capability of the sludge. A considerable number of such assessments was made, and it was found that the COD was always removed very rapidly. Four such runs are shown in Figure 8. These were selected in order to show the COD removal patterns shortly after sludge concentration had attained the low value at the end of the decreasing cycle (day 312), and during the succeeding period of solids accumulation (days 421, 506, and 587). For the run of day 312, the unit was fed with a greater concentration than 500 mg/l because it was felt that the higher feed value would permit more sampling points to be obtained before the COD was assimilated. This practice was later changed because it became apparent that sufficient points to assess COD removal capability could be obtained at the 500 mg/l feed level. In all cases it can be seen that the substrate was removed very rapidly. It is particularly interesting to note that on days 506 and 587 (after the endogenous uptake of the sludge had dropped below 2 mg O<sub>2</sub>/hr/gm sludge) the rate was particularly rapid. It is evident that as the biological solids accumulated, any loss in unit capability was amply made up by an increased total substrate consuming capability of the biomass. On day 587 the COD was removed so rapidly by the high biological solids concentration that a considerable amount of COD (approximately 170 mg/l) was removed in the short (3-5 minute) period which elapsed between the time of substrate addition and filtration of the sample (see initial point).





## DISCUSSION

At the outset of this report it was emphasized that much research has been previously accomplished on the extended aeration process, and that it is generally concluded that biological solids concentration will continue to build up in such systems because there is a biologically inert fraction of the synthesized cells which cannot be metabolized by the aerobic population in the aeration tank. In most of the studies previously reported, some solids were wasted either purposely or because they escaped in the effluent or wasted supernatant. The investigational question which was posed in designing the present study might be stated as follows: If a way could be found to retain all of the synthesized solids in the system, would not the inactive fraction become a larger and larger portion of the sludge, eventually becoming so large that it would cause a biochemical failure, i.e., cause loss of ability to metabolize substrate? The results after nearly two years' operation of an extended aeration system with no sludge wasting indicate that the system has not yet lost its biochemical efficiency. It is still providing approximately 90 per cent COD removal efficiency for a feed COD of approximately 530 mg/l and an aeration chamber detention time of sixteen hours. The effluent quality compares favorably with that of the more conventional (sludge wasting) activated sludge plants. The results also indicate that sludge does not accumulate "ad infinitum." There have been periodic cycles of decreasing solids concentration followed by succeeding periods of solids accumulation. Furthermore, during the periods of decreasing solids concentration there was no gross leakage of COD in the effluent. These decreases in biological solids concentration cannot be attributed to escape of biological solids in the effluent, and the

causation for the decreasing solids concentration cannot be attributed to external causes (changes in pH, temperature, etc.).

At the start of these experiments it was anticipated that the biological solids concentration would in all probability steadily increase, and that the system would eventually undergo biochemical failure. The main purpose of the experiments was to determine how long it would take before failure occurred. Had this not been the aim of the study, and had the duration of the experiments been shorter, or had some sludge wasting been practiced, the findings and the conclusions based upon them would in all probability have been very similar to those of other workers who have studied this process. However, the results which were obtained indicate that it is possible for the accumulation of biological solids concentration to be periodically self-relieved, and this internal regulation must be ascribed to natural biological causation.

There are a number of natural explanations or theoretical possibilities for periodic acceleration of autodigestion of a biomass. The fact that cells undergo a decrease in mass during endogenous respiration is well known, but the endogenous respiration of any particular microbial species cannot normally be expected to lead to total oxidation of the biomass. However, it is known that some species undergo complete or nearly complete autolysis after attaining a maximum growth level (10)(11). Thus the released materials become food for other species. In cases where autolysis leads to the dissolution of the cell (i.e., solubilization of structural components) as well as simple disruption or tearing of the cell wall and subsequent release of internal components, the entire biomass of the cell could become food for another cell; thus, the autolyzed cell could be totally oxidized. Furthermore, it is known

that various microorganisms produce enzymes which cause lysis or complete dissolution of other cells (12)(13); thus, the autolytic process is not the only one which could lead to breakdown of cellular macromolecules to readily available sources of energy leading to total oxidation of a particular cell or species of cells in a mixed population. In addition, cells may be disrupted by bacteriophage.

In the water pollution control field the presence of lytic agents in the cell-free filtrate obtained from a natural microbial population undergoing endogenous respiration has been demonstrated (14). Also, it can be reasoned that without doubt an activated sludge represents an extremely complicated ecosystem, and some cells in the biomass enter the food chain of the higher organisms, such as protozoa and the slime molds. Thus, predacity is not solely expressed by interplay between bacterial species, but involves the entire population in the ecosystem. Thus, the possibilities for autolysis, lysis by enzymes produced by other bacteria, cell disruption by bacteriophage, predacity by higher organisms, all of which can and do undoubtedly go on in the very complicated ecosystem represented by an activated sludge, contribute possible explanations for the continued successful functioning of an activated sludge process in which all biological solids are returned to the aerator.

It cannot be expected that an equilibrium or steady state solids concentration can be attained, since such a state would require a precisely balanced ecological regime in which one or all of the natural lytic or predatory agents for various species were present in the proper ratio at all times. Since an activated sludge system is a dynamic one with respect to the biological population it contains, i.e., one in which many changes in species predominance occur, such a precise



balance could seldom occur, and the solids concentration in the system will at times exhibit an increasing trend, and at other times exhibit a decreasing trend. It seems also to be apparent, based upon the results of the present experimentation, that it is not axiomatic that an inactive fraction which cannot be metabolized will continue to build up under conditions of total sludge recycle. It seems obvious that a given microbial cell cannot totally oxidize itself, but it is not impossible or improbable that all of the organic matter from any particular microbe can serve as food for other species in the ecosystem. Concerning capsular material (largely polysaccharide in nature), it is well known that some organisms utilize the capsular material which they have produced as a source of energy, and some do not (15). However, even in cases where the capsule is not utilized by the particular species which produced it, it is certainly possible that this material (or any other cellular material) can be used as a source of carbon by another species. If an organism can produce the hydrolytic enzymes to split polymers, thus solubilizing the material, it can be expected that various species will induce enzymes for the terminal metabolism of these carbon sources.

The time required to complete a cycle of net accumulation and net decrease of sludge cannot be predicted, nor can the periodicity of such cycles, but the results of the experimentation to date attest to the fact that such cycles do exist.

Also, the data of Washington, et al. (16) suggest that cyclic reduction in biological solids accumulation can occur in extended aeration processes. In their studies, batch systems were employed, and there was apparently no positive control of solids loss in the effluent. However, during a period of operation of approximately one year and

four months (July, 1962-November, 1963) they observed a gradual rise in solids concentration followed by a gradual decline and subsequent rise. Their data were such that the decline could not be attributed to loss of solids in the effluent. They felt that the decline in solids was not the result of lysis of a portion of the population, but that a portion of the biologically inert volatile solids fraction was metabolized by an organism (or organisms) which adapted to this material. Their basis for ruling out lysis was that the decrease in solids concentration was not accompanied by an increase in effluent COD. However, it is entirely expectable that lysed materials would be metabolizable by the remaining intact cells. Regardless of the various interpretations which can be placed upon their results, the fact remains that the results indicate there was a period of accelerated autodigestion which relieved solids accumulation. They concluded that it was uncertain that there was any long-term cyclic reduction in the accumulated volatile solids. It is unfortunate that they terminated the experiments, since their data indicate that the system may have been poised for a second cycle of decreasing solids concentration. The results of the present study, as well as those of Washington, et al. (16), indicate that periodic relief of net solids accumulation via metabolic processes can be expected. Available information in the basic literature reinforces the expectation that periodic net autodigestive cycles should occur in complicated natural microbial ecosystems. It is known from the results of recent experiments with heterogeneous populations that the soluble portion of the cells released after breakage of cell walls provides an excellent substrate for microbial growth (17). In other recent work employing heterogeneous populations, systems have been observed wherein essentially

total oxidation of sludge synthesized in the log and declining growth phase has occurred in a subsequent prolonged endogenous phase (18)(19).

A considerable amount of time may be required for total oxidation to occur, but it must be remembered that "in theory" since all sludge is returned to the system, it can be retained until the ecological situation is one which fosters an accelerated autodigestive cycle. Thus, if the sludge can be retained in the system, the amount of time which may be required does not offer a particular problem.

There is, of course, a very real and practical engineering problem when sludge accumulates for a sufficient time to allow the concentration to exceed that which can be effectively separated by gravity sedimentation in a conventional clarifier operation. At such times, suspended solids appear in the effluent of total oxidation plants. This is indeed a significant problem, but it seems fortunate that it is the major problem concerning the process, since from the results of the present study it does not appear that the "theory" of total oxidation is an erroneous one. While there is much work yet to be done, the results to date do much to remove the stigma of theoretical or mechanistic "unsoundness" of the process, and the findings have caused the authors to re-evaluate its useful possibilities for treatment of soluble industrial wastes. There are various engineering expedients which might be employed to alleviate the sludge settleability problem during periods of extremely high solids concentration. Improvements in various types of proprietary sedimentation equipment are constantly being made. Also, it seems possible that the thick mixed liquor from the aeration chamber could be split into parallel settling tanks and diluted with effluent or water from the receiving stream to decrease

the sludge concentration to an optimum settling range. Also, research on various additives to enhance settling is presently being accelerated by various chemical manufacturing firms. Thus there is reason to believe that the settling problem is not an insurmountable one and can and can eventually be solved.

#### SUMMARY AND CONCLUSIONS

In summary, the experimental results herein presented warrant the conclusion that an extended aeration activated sludge system without sludge wasting can be operated (with reasonably good biochemical efficiency) without continual solids accumulation. It is indeed doubtful that such would be the case in a simple ecosystem. In order for a portion of the population to be used as food, other organisms must successfully prey upon these living sources of carbon. To allow the cycle to continue, there must be diversity of the population, shifts in predominance of bacterial species and in the predator population. In short, the ecosystem must be a very complicated, ever-shifting one such as may be expected to exist in waste treatment plants. It is somewhat paradoxical that the simplest biological treatment process depends for its theoretical *raison d'être* upon the extreme complexity of the ecosystem.

The treatment and disposal of sludge from activated sludge plants represents one of the major problems of the pollution control field. The extended aeration process offers a means of obviating the need for sludge treatment and disposal facilities, and results of the present work indicate that a total oxidation process is not inconsistent with sound microbiological theory. While much more research into the biological mechanisms responsible for successful net autodigestive periods

(and possibly ways of controlling them) is needed, present results warrant some re-evaluation and possible extension of the scope of application of extended aeration processes for soluble organic industrial wastes.

#### ACKNOWLEDGEMENT

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## REFERENCES

1. Ramanathan, M., Gaudy, A. F., Jr., and Ragthaidee, W., "Responses of Extended Aeration Activated Sludge to Quantitative Shock Loads." Proc. 19th Oklahoma Industrial Wastes and Pollution Control Conference, Stillwater, Oklahoma (1968). (Submitted for publication in Journal Water Pollution Control Federation.)
2. Porges, N., Jasewicz, L., and Hoover, S. R., "Aerobic Treatment of Dairy Wastes." Jour. Applied Microbiology, 1, 262-270 (1953).
3. Symons, J. M., and McKinney, R. E., "The Biochemistry of Nitrogen in the Synthesis of Activated Sludge." Sewage and Industrial Wastes, 30, 874-890 (1958).
4. Washington, D. R., and Symons, J. M., "Volatile Sludge Accumulation in Activated Sludge Systems." Journal Water Pollution Control Federation, 34, 767-789 (1962).
5. Busch, A. W., and Myrick, N., "Food Population Equilibria in Bench-Scale Biooxidation Units." Sewage and Industrial Wastes, 32, 949-959 (1960).
6. Kountz, R. R., and Fourney, C., Jr., "Metabolic Energy Balances in a Total Oxidation Activated Sludge System." Sewage and Industrial Wastes, 31, 819-826 (1959).
7. Pfeffer, J. T., "Extended Aeration." Water and Sewage Works, 113, 207-214 (1966).
8. "Standard Methods for the Examination of Water and Waste Water." 12th Edition, American Public Health Association, New York (1965).
9. Gaudy, A. F., Jr., "Colorimetric Determination of Protein and Carbohydrate." Industrial Water and Wastes, 7, 17-22 (1962).
10. Moustafa, H. H., and Collins, E. B., "Molar Growth Yields of Certain Lactic Acid Bacteria as Influenced by Autolysis." Jour. Bacteriol., 96, 117-125 (1968).
11. Stolp, H., "Bacteriolysis." Annual Review of Microbiology, 19, 79-104 (1965).
12. Ensign, J. C., and Wolfe, R. S., "Lysis of Bacterial Cell Walls by an Enzyme Isolated From a Myxobacter." Jour. Bacteriol., 90, 395-402 (1965).
13. Burke, M. E., and Pattee, P. A., "Purification and Characterization of a Staphylolytic Enzyme From Pseudomonas aeruginosa." Jour. Bacteriol., 93, 860-865 (1967).
14. Simpson, J. R., "The Biological Oxidation and Synthesis of Organic Matter." J. Inst. Sew. Purif., (2), 171-180 (1965).

15. Oginsky, E. L., and Umbreit, W. W., "An Introduction to Bacterial Physiology." W. H. Freeman & Company, San Francisco (1954).
16. Washington, D. R., Hetling, L. J., and Rao, S. S., "Long-term Adaptation of Activated Sludge Organisms to Accumulated Sludge Mass." Proceedings 19th Industrial Waste Conference, Purdue University, Ext. Ser. 117, 655-666 (1965).
17. Obayashi, A. W., and Gaudy, A. F., Jr., Unpublished Data (1969).
18. Thabaraj, G. J., and Gaudy, A. F., Jr., "Effect of Initial Biological Solids Concentration and Nitrogen Supply on Metabolic Patterns During Substrate Removal and Endogenous Metabolism." (In Preparation, 1969).
19. Thabaraj, G. J., and Gaudy, A. F., Jr., Unpublished Data (1969).

Ramanathan, M., Gaudy, A. F., Jr., and Raghuidee, M., "Responses of Extended Aeration Activated Sludge to Quantitative Shock Loads." Proceedings, 19th Oklahoma Industrial Water Conference, Oklahoma State University, Stillwater, Oklahoma (1968).

APPENDIX 2



# RESPONSES OF EXTENDED AERATION ACTIVATED SLUDGE TO QUANTITATIVE SHOCK LOADS

By

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## INTRODUCTION

If they could be successfully operated, extended aeration systems could achieve the ultimate objective of aerobic biological waste treatment, i.e., the complete mineralization of organic wastes. The principle upon which such systems are designed is the basic surmise that, with provision of sufficient contact time, the stabilization of organic materials can be achieved without any net accumulation of sludge. The ultimate BOD of the waste is exerted in the aeration tank; the aeration time is much longer than for conventional activated sludge processes (usually 24 hours rather than 4-8 hours), and sludge disposal and primary treatment facilities are not employed in extended aeration systems. This process has been claimed to possess many advantages, e.g., low initial cost and maintenance requirements, and ability to withstand severe shock loads.

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If the advantages mentioned above are in fact true, wider application of this process would seem warranted; however, the process has been used only for rather small scale installations. Wider application of the process has not come about since, based upon theoretical rationale and experimental data, it has been doubted that "total oxidation" is possible. It is the purpose of this paper to present results obtained with an extended aeration system operated over a fairly long period without sludge wastage, under varying degrees of quantitative shock loadings. It is believed that the results presented provide insight toward a better understanding of the process.

#### REVIEW OF LITERATURE

The total oxidation or extended aeration process for treatment of waste waters has received considerable attention during the past decade. However, as is the case with most new processes, it has been the subject of much controversy. The accumulation of inert metabolic products and inactive biological solids are major subjects for discussion.

Extended aeration processes are designed to utilize fully the endogenous respiration of sludge. Porges, et al. (1) in studies employing skim milk as substrate, reported that "true" endogenous phase oxidation occurs with one per cent loss in sludge weight per hour. The "true" endogenous phase refers to oxidation which occurs after the assimilation of exogenous substrate and the oxidation of storage products within the cell. They also concluded that with

extended periods of aeration the net accumulation of sludge could approach zero. Thus the system could be operated at solids equilibrium, thereby negating the need for sludge wastage and disposal. Kountz (2) supported this concept, however, he reported that the suspended solids concentration should not exceed 3000 mg/l in order to facilitate clarification. In a later study, Forney and Kountz (3) confirmed the attainment of equilibrium with skim milk waste in a continuous flow reactor. The equilibrium value of biological solids concentration was twelve times the substrate concentration in the feed.

While the preceding work had indicated that extended aeration systems could be operated without sludge wasting, Symons and McKinney (4) reported that operation without sludge wasting was not possible. Their conclusions were based upon studies with sodium acetate as substrate in batch-fed units. Extracellular polysaccharides, which they felt were resistant to biodegradation, accumulated in the system during the 35-day experimental period.

In a later study with skim milk waste, Kountz and Forney (5) reported accumulation of sludge at the rate of 0.122 lb/day. They were able to operate the system at equilibrium by wasting sludge at the same rate as it was accumulating. The equilibrium weight of sludge was fourteen times the weight of substrate fed per day. Busch and Myrick (6) studied both batch and continuous flow total oxidation systems in the laboratory, and concluded that establishment

of an equilibrium solids level was unattainable even after 103 days of operation. Their results are in support of the concept that sludge wasting is required to operate total oxidation systems at equilibrium; however, they pointed out that for certain types of industrial wastes for which the ratio of respiration to synthesis is high, total oxidation systems might be plausible. Washington and Symons (7) reported volatile solids accumulation in the amount of 10 to 15 per cent of the ultimate BOD removed and referred to this accumulation as inert and/or inactive organic cells.

In a batch study using glucose as substrate, McWhorter and Heukelekian (8) observed the typical rise in sludge concentration wherein the peak concentration coincides with the time of substrate removal. However, they continued to aerate the sludge for a prolonged period, and observed a decrease in biological solids concentration. Eventually the rate of decrease approached zero, and the terminal amount of sludge produced was 12 per cent of the amount of substrate added. This figure agrees well with the value of 10 to 15 per cent reported by Washington and Symons. McWhorter and Heukelekian noted this agreement, and referred to the portion of sludge not subject to autooxidation as an inactive cell mass.

Based upon work cited above, there has been general agreement that a gradual buildup of suspended solids concentration can be expected in systems operated without sludge wasting. Also, it has been generally concluded that an

activated sludge cannot totally oxidize itself. Some 10-15 per cent of the organic feed is channeled into "permanent synthesis" of biological materials. While the residual fraction may be termed inert or inactive because it has lost its facility for self destruction (auto-oxidation), there would appear to be some room for question concerning the inertness or inactivity of this residual sludge as an agent for substrate removal; i.e., what proportion of this sludge (if any) contributes to the so-called "active fraction" of the activated sludge mass. The impression gained from the literature is that it is assumed that none of it does. However, there are few or no data upon which to base an unequivocal conclusion in this regard. There is little doubt that the "active fraction" (with respect to substrate removal) of the residual biomass after prolonged endogenous respiration is smaller than the active fraction of the biomass from a more conventional (sludge wasting) activated sludge process, but there is doubt that there is no "active fraction" after prolonged endogenous respiration. In addition, it seems appropriate to point out that studies of the type reported by McWhorter and Heukelekian are of more immediate applicability to considerations of aerobic digestion as a separate unit process than to the total oxidation process in which exogenous substrates are continuously added.

From the foregoing review it can be seen that the results of some research workers indicate that there is a gradual accumulation of biologically inert organic solids

which should ultimately cause functional failure of "total oxidation" plants. However, no one seems to have studied the system over a sufficiently long period to determine the time required to produce such failure. While there may be some theoretical basis for expecting an accumulation of inert and nonoxidizable organic solids, it would seem ideal from an engineering standpoint to determine the extent of the period of useful operation. The concept of combining secondary treatment and autodigestion of the sludge is indeed an intriguing one which has obvious engineering advantages, and if the process could be operated successfully for a year or so before disposal of a portion of the sludge became necessary, it would still offer an attractive alternative to the conventional practice. In view of the above considerations and the still controversial concern over the need for eventual sludge wasting, a long-term laboratory study was designed in which an extended aeration plant was operated with all solids returned to the aeration chamber except for a small, known amount used for analyses to assess system performance. Also, since a buildup of inert biological solids would be expected to have an adverse effect upon the ability of the system to take shock loads, such studies were also included. The present report deals primarily with the shock load aspects of the work.

#### MATERIALS AND METHODS

Before describing the experimental procedures for the shock load studies herein reported, it is appropriate to

provide a brief history of the development of the extended aeration sludge employed. The sludge was developed from an initial sewage seed obtained from the effluent of the primary clarifier at the municipal sewage treatment plant at Stillwater, Oklahoma. The extended aeration unit (total volume of aeration and settling chamber = 9.4 liters) was put into operation on March 31, 1967, and operated under continuous flow conditions with 1000 mg/l glucose as carbon source (detention time, 24 hours). All biological solids, except a small portion taken for analysis, were returned to the aeration chamber. The solids level gradually built up and on October 12, 1967 (196 days after starting the unit), approximately half of the system was transferred to a second unit of the same type and both portions were diluted to 9.4 liters with tap water. Since the biological solids concentration was now approximately halved, the feed concentration was reduced (to 500 mg/l glucose). One system was used for the shock load studies reported herein, and the other was retained for continuing studies on the long-term behavior of the extended aeration process. The composition of the standard daily feed is given in Table I. In specific shock load experiments, higher glucose levels were employed, and the inorganic nutrients were increased proportionately.

TABLE I

COMPOSITION OF FEED FOR 500 mg/l GLUCOSE  
AS SUBSTRATE

Glucose	500	mg/l
$(\text{NH}_4)_2\text{SO}_4$	250	mg/l
$\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$	50	mg/l
$\text{FeCl}_3$	0.25	mg/l
$\text{CaCl}_2$	3.75	mg/l
$\text{MnSO}_4 \cdot \text{H}_2\text{O}$	5	mg/l
Phosphate Buffer, 1.0 M ( $\text{KH}_2\text{PO}_4 + \text{K}_2\text{HPO}_4$ )	10	ml/l
Tap Water	100	ml/l

Figure 1 shows the cross section of the laboratory scale total oxidation units. For batch operations, the adjustable baffles were removed from the reactor, since no clarifier was required. For batch operation, the reactor volume was 9.4 liters. For continuous flow operations, the baffles were inserted and the feeding was accomplished using a Milton Roy positive displacement pump. The baffles divided the reactor into an aeration chamber and a clarifier. The aeration chamber volume was 5.7 liters, and the clarifier volume, 3.7 liters. Feeding rate was adjusted to 24 hours overall detention time, i.e., 17 hours detention time in the aeration tank, and 7 hours in the clarifier. The adjustable baffles and clarifier walls proved to be useful in providing a "stilling" action in the settling chamber and in directing return sludge to the aeration



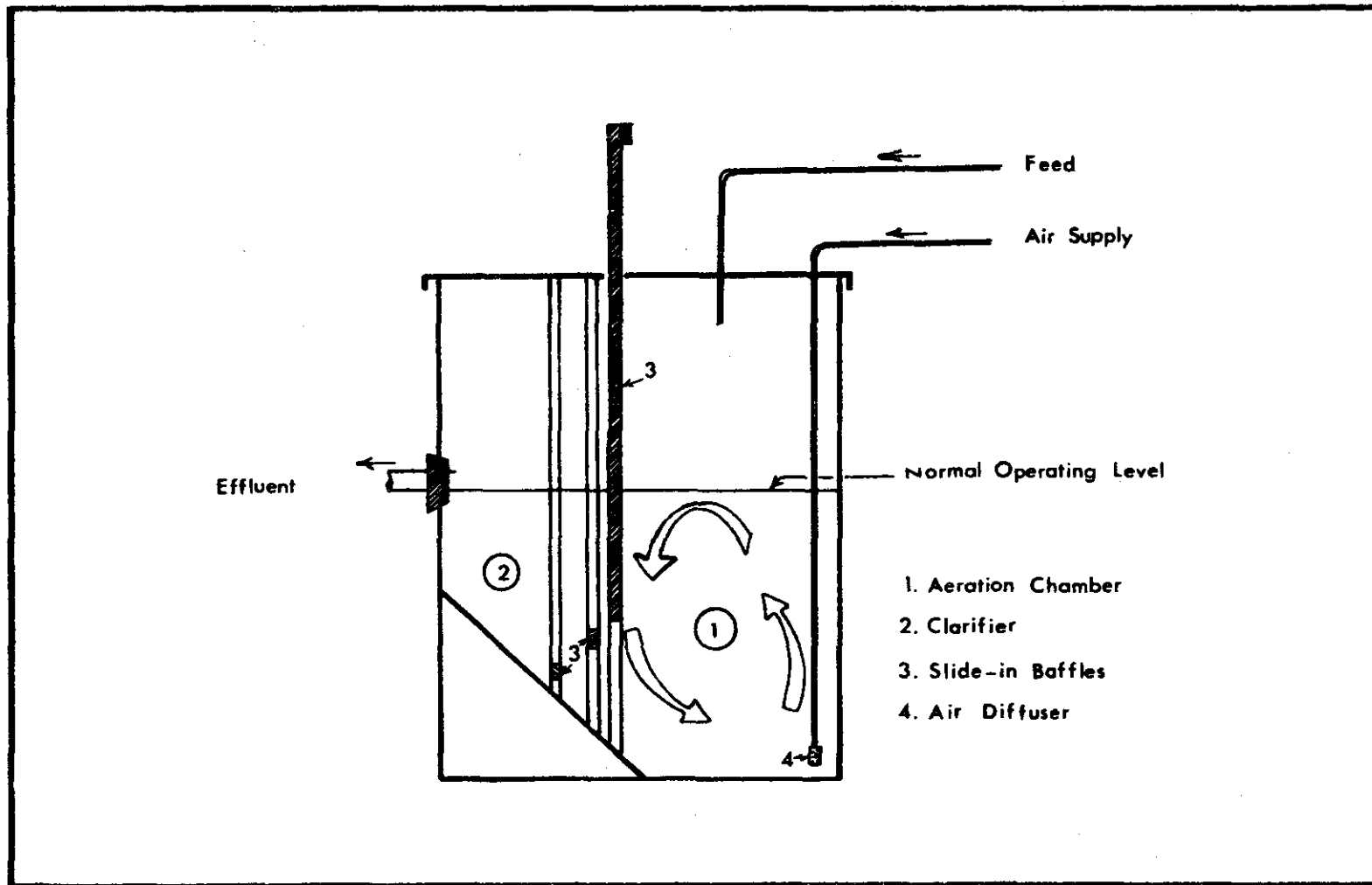


Figure 1. Laboratory Scale Continuous Flow Extended Aeration System

chamber. Mixing in the aeration chamber was accomplished by air diffusion at the rate of 20 liters per minute. The rate of air supply was the same in both batch and continuous systems. At times when the continuous flow effluent exhibited a degree of turbidity (poor settling), the suspended solids carried over in the effluent were recovered by centrifugation (using a Sharples super speed centrifuge) and returned to the reactor, thus enabling positive control of sludge wastage regardless of the settling characteristics of the solids.

#### Experimental Protocol

The experiments for this study were conducted in two phases:

1. The development of "equilibrium" under batch operation and administration of shock loads of desired quantity, and
2. The development of "equilibrium" under continuous flow operation and administration of shock loads of the same magnitude as studied under batch conditions.

Beginning on October 20, 1967, the unit employed for the shock load studies was operated under batch conditions and studies under Item 1 were conducted. On March 1, 1968, continuous flow operation was again initiated and studies under Item 2 were undertaken.

### 1. Batch Operation, Single Daily Feeding

Twenty-three hours of aeration and one hour of settling were allowed between successive feedings. After settling, one liter of supernatant was removed from the reactor and centrifuged, and the separated solids were returned to the aerator. The unit was then brought to aeration volume with one liter containing 9.4 times the concentration of each nutrient shown in Table I. Daily samples were taken to determine the concentrations of suspended solids and the chemical oxygen demand (COD) of the mixed liquor. When the system approached an approximate equilibrium with respect to solids and COD concentration, the shock load experiments were conducted.

The shock load experiments with the extended aeration sludge were conducted at both high and low biological solids concentrations. The experiments with high initial solids concentrations were conducted in the extended aeration unit. For the low initial solids concentrations, a small amount of mixed liquor was removed from the extended aeration reactor (usually 10-15 ml) and mixed with feed solution to make one liter of reaction mixture. The same organic shock load was applied to both high and low solids systems. Shock loads of 1000, 1500, 2000, and 2500 mg/l glucose were applied. Samples were taken for analysis at frequent intervals (usually every fifteen minutes for high initial solids systems and every forty-five minutes for low initial solids systems) to determine the rate of substrate removal and solids growth.

## 2. Continuous Flow Operation

When the experiments with the batch-fed system were completed, the unit was operated with continuous addition of growth medium (500 mg/l glucose as carbon source). The system was allowed to attain "equilibrium" before shock loads were applied. During shock loads, the inflow medium was changed to the desired level of glucose. Feeding at this level was continued until a new "equilibrium" was established. Experiments with low initial solids concentration were also conducted simultaneously, as explained for batch-fed systems. After sufficient data were collected at one shock load level (usually 48 hours), the feed was changed to the original level (500 mg/l glucose) and operated at this level until the next shock load was applied. Then the next higher level of shock load was applied. This procedure was repeated until all of the shock load experiments (1000, 1500, 2000, and 2500 mg/l glucose media) were completed.

Before changing the feed concentration, i.e., administering a shock load, the biological solids concentration in the aeration chamber was measured, then the waste flow was momentarily turned off and the baffle which formed the settling chamber wall was removed, allowing mixing of the solids in the aeration and settling chambers. Another sample for biological solids was taken, and the settling chamber wall replaced. Pumping of the feed was resumed (at the shock load concentration), and sampling was begun to assess the transient behavior of the system.

In addition to the above studies, oxygen uptake measurements (Warburg respirometer) on washed cell suspensions were made frequently to determine the "unit activity" of sludge; a reaction liquor volume of 40 ml was used. The term "unit activity" is herein defined as the milligrams of oxygen consumed per gram of washed cells per hour. The cells from the total oxidation unit were washed twice with 0.1 M phosphate buffer, and resuspended in the buffer solution before oxygen uptake measurements were made. In experiments conducted with low initial solids concentration, the carbohydrate content of the filtrate was also measured.

#### Analytical Procedures

Suspended solids determinations were made using the membrane filter technique, and chemical oxygen demand was measured in accordance with Standard Methods (9). The carbohydrate content of the filtrate was measured by the anthrone method (10). Periodically, dissolved oxygen in the aeration chamber was measured electrometrically.

### RESULTS AND DISCUSSION

The results obtained during this investigation are presented in the following manner. First, the daily performance of the system under batch and continuous flow operation (before initiation of the shock load experiments) is presented. Second, the responses to shock loads of systems under batch and continuous flow operation are shown for both high and low initial solids concentrations. Third, the results of a study on the unit activity of the sludge are presented.

Figure 2 shows the performance under batch and continuous flow operation before shock loads were applied. The system was operated under batch conditions for 51 days before shock loading, and under continuous flow conditions for 27 days before applying shock loads. No sludge was wasted throughout the experimental period; however, a small amount was taken for analysis. Only one sample was taken each day (0.16 per cent of the mixed liquor volume). In the batch system, the sample was taken at the end of the 23-hour aeration period. It can be seen from the figure that both the concentrations of biological solids and filtrate COD reached higher values in the batch system than in the continuous flow system. Also, both systems appeared to approach an equilibrium condition with respect to solids and COD. Even though only a limited amount of information was collected during this part of the investigation, it can be said that the continuous flow extended aeration process performed more satisfactorily than did the batch-fed system. The mean biological solids concentrations were 8000 and 7800 mg/l for batch and continuous systems, respectively. The mean effluent COD concentrations differed by a considerable amount; they were, respectively, 396 and 47 mg/l for batch and continuous operations. The average organic loadings employed were 0.62 and 0.64 lbs COD/lb SS/day for batch and continuous systems, respectively.

Figure 3 shows the overall performance data for both batch and continuous systems during the period when shock

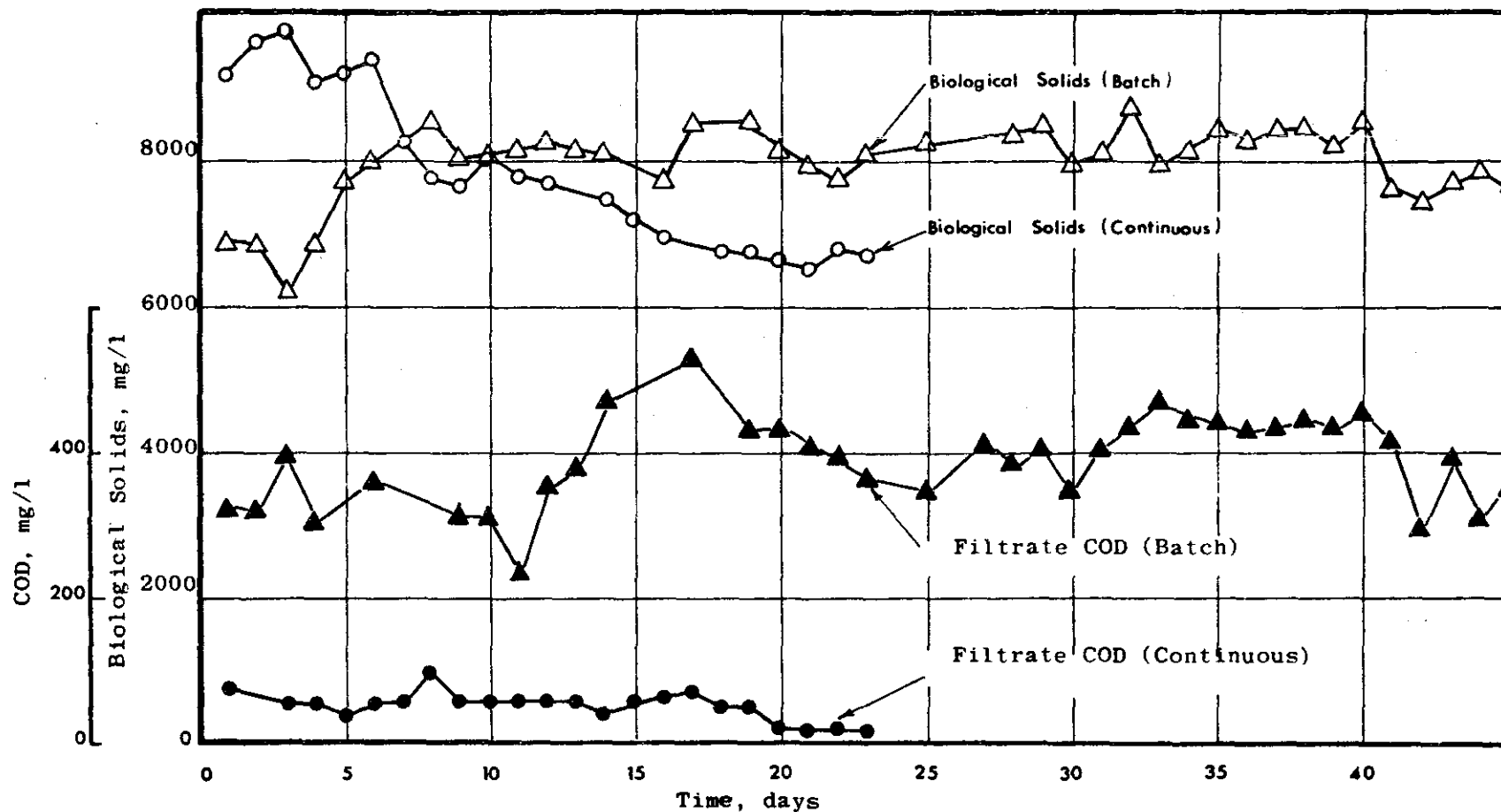


Figure 2. Biological Solids and Effluent Concentration in an Extended Aeration Process under Batch and Continuous Flow Operation. Normal Feed = 500 mg/l Glucose; No Shock Loading Applied.

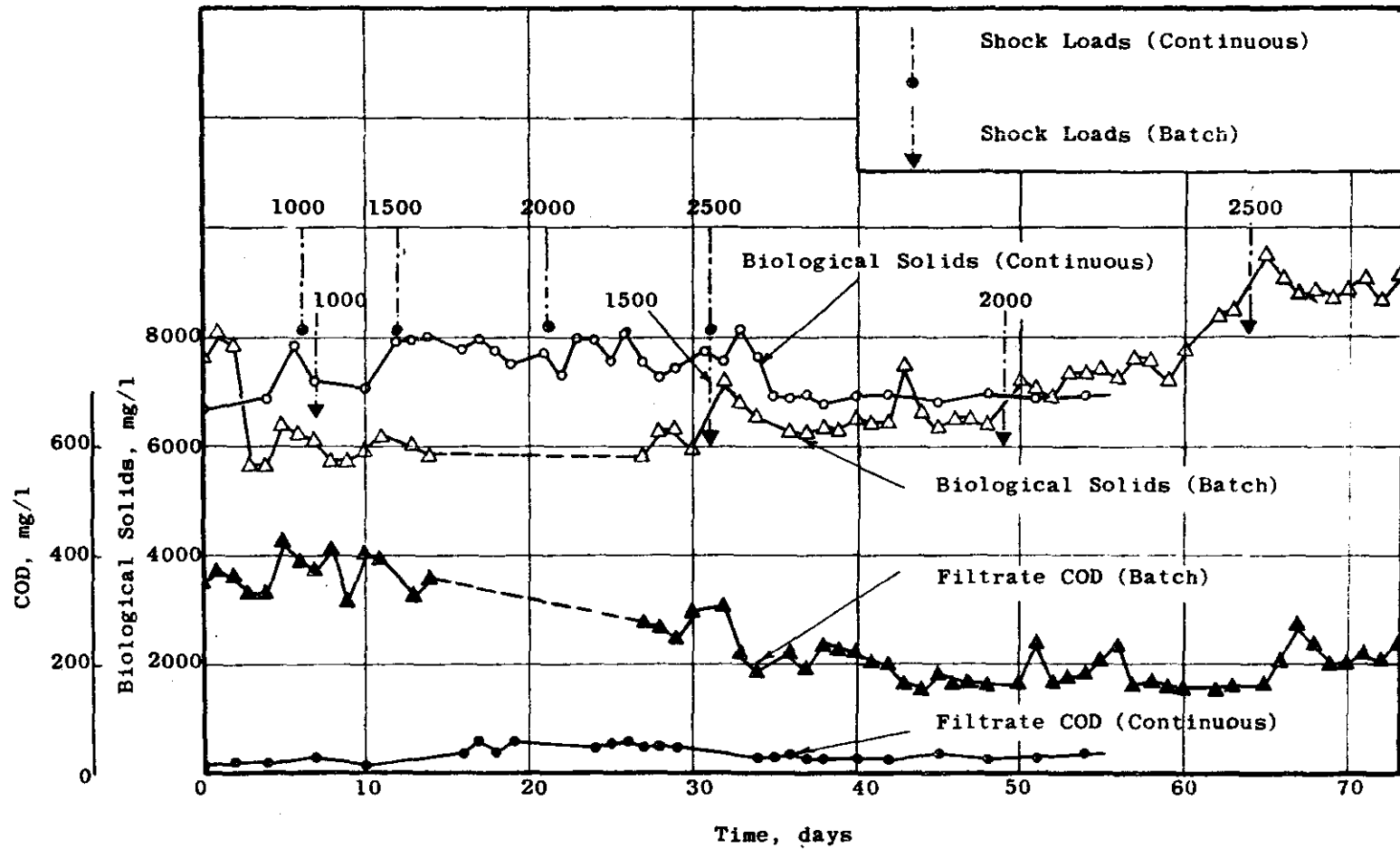


Figure 3. Biological Solids and Effluent COD Concentration in an Extended Aeration Process under Batch and Continuous Flow Operation. Normal Feed = 500 mg/l Glucose; Shock Loadings Applied as Indicated.



load experiments were being carried out. The days on which shock loads were applied and the magnitudes of the shock loads are indicated by arrows in Figure 3. Details of responses to shock loads are shown in subsequent figures. "Day zero" on the time scale corresponds to the last day plotted for each system in Figure 2. Thus, the system was operated as a batch system for a total period of 118 days, and as a continuously-fed system for 77 days during the experimental period; i.e., the period of batch operation was 118 days, followed by reestablishment of continuous flow operation. However, it is emphasized that the sludge was almost one year old before these experiments were initiated, having been developed under continuous flow conditions without cell wastage. It can be seen from the figure that increasing the feed up to five times (from 500 mg/l glucose to 2500 mg/l glucose) did not adversely affect the performance of the system. The average daily performance before and after the shock loads was relatively constant; i.e., within the normal fluctuations in the system parameters (compare Figures 2 and 3). As mentioned in the preceding paragraph, the continuously-fed system performed better than the batch-fed system with respect to effluent quality. The COD of the effluent (filtrate) in the continuous system never exceeded 60 mg/l. COD's were also run on unfiltered effluent; generally they were 30 to 50 mg/l higher than the filtered effluent COD, attesting to the generally good settling characteristics of the sludge. For

clarity of presentation of the results, these values were not plotted. At times the unfiltered effluent COD was considerably in excess of this range; indeed, at one time during the experimental period, the biological solids concentration in the effluent rose to 200 mg/l. It is important to re-emphasize that the solids carried over with the effluent were recovered and returned to the aeration tank. In general, sludge settleability was as good or better than experienced with conventional activated sludge processes.

Figures 4 through 11 show the transient responses of the extended aeration activated sludge to the various shock loads. The results obtained by direct sampling in the unit and for a separate system with low initial solids are shown for each experiment. Figure 4A shows the response of the batch-fed sludge to 1000 mg/l glucose. There was a rapid decrease in COD concentration, and in 75 minutes it attained a terminal value of approximately 400 mg/l, which is consistent with the rather high residual COD concentration seen in Figures 2 and 3. The decrease in COD was accompanied by a corresponding increase in biological solids concentration (from 5900 mg/l to 6500 mg/l).

Figure 4B shows the response of the same sludge at a lower concentration. It can be seen from the figure that almost 36 hours were required to assimilate 1000 mg/l glucose COD. The carbohydrate curve indicates that a maximum of 300 mg/l metabolic intermediates and/or end products were produced during the metabolism of glucose, and a large

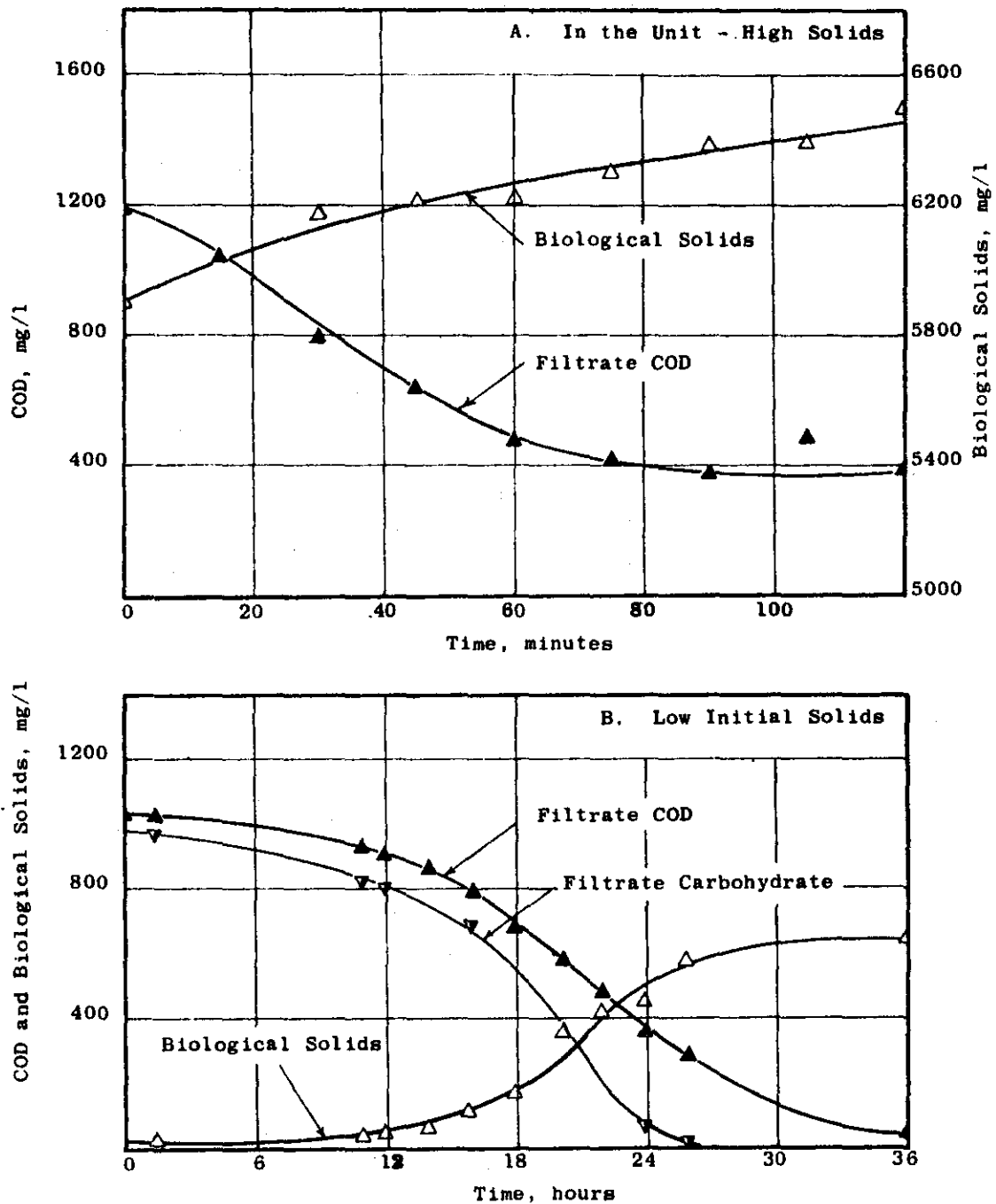


Figure 4. Response of Batch-operated Extended Aeration Activated Sludge to Quantitative Shock Loading with 1000 mg/l Glucose.

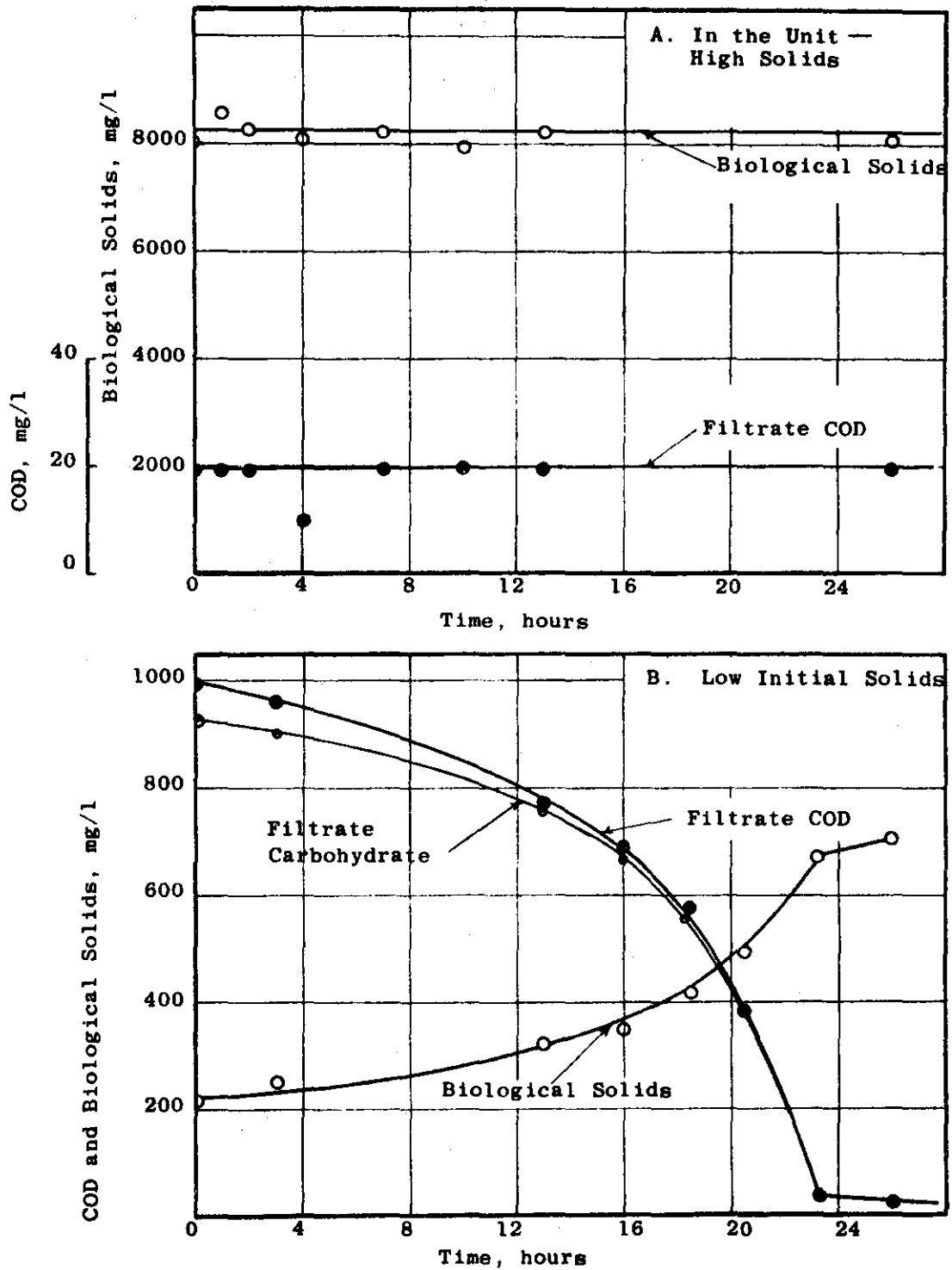


Figure 5. Response of Continuous Flow Extended Aeration Activated Sludge to Quantitative Shock Loading Applied as an Increase in Feed Concentration from 500 to 1000 mg/l Glucose.

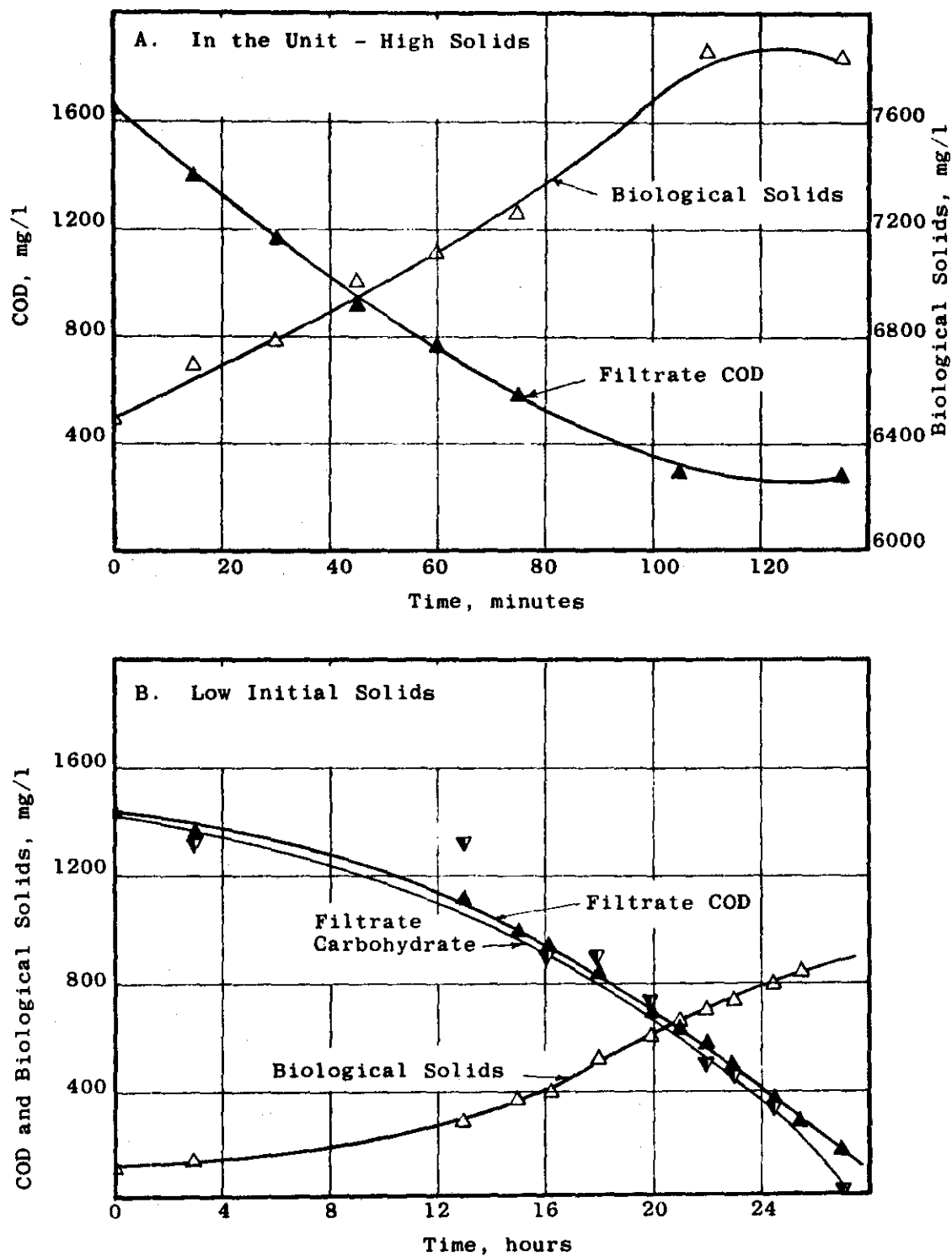


Figure 6. Response of Batch-operated Extended Aeration Activated Sludge to Quantitative Shock Loading with 1500 mg/l Glucose.

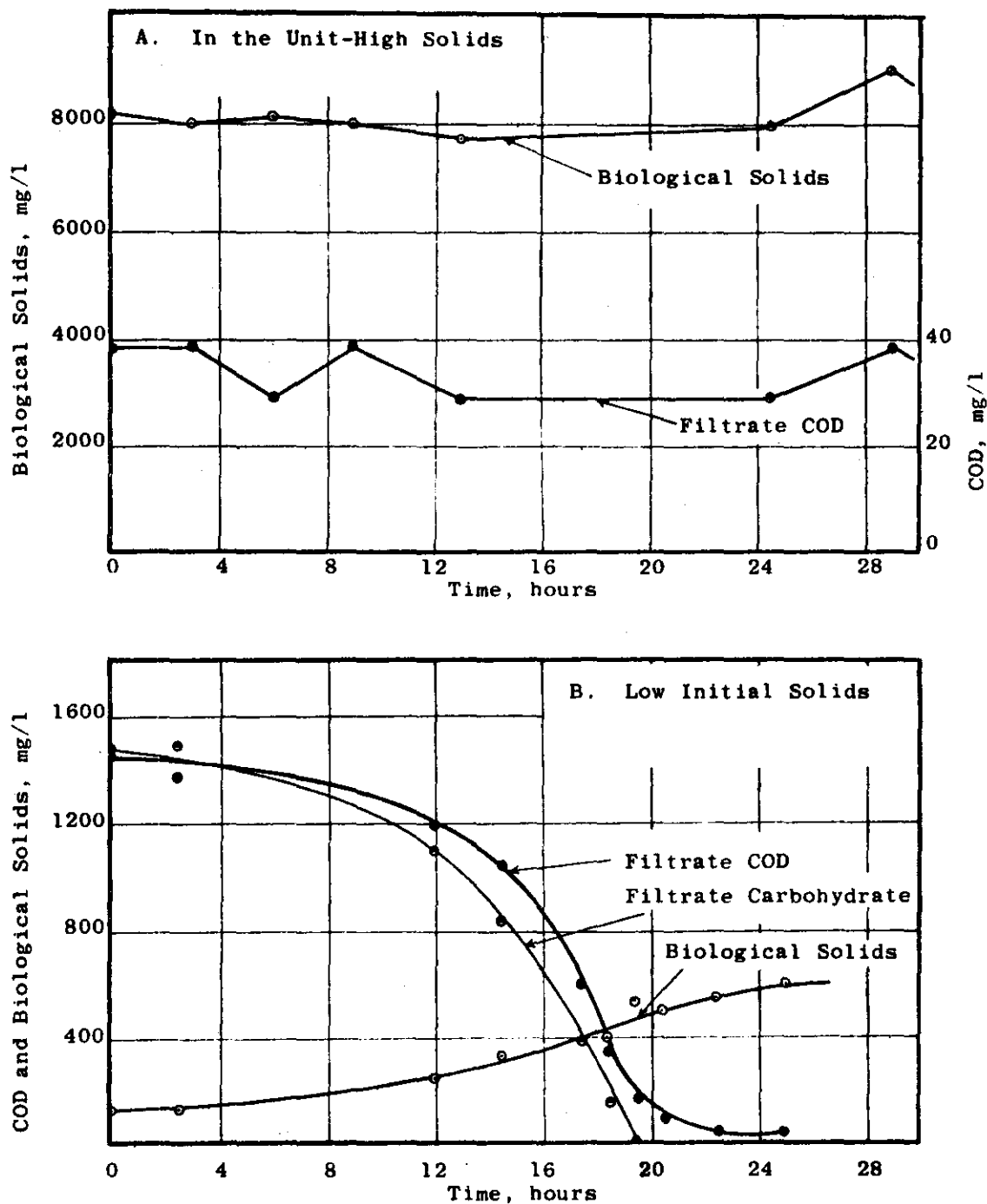


Figure 7. Response of Continuous Flow Extended Aeration Activated Sludge to Quantitative Shock Loading Applied as an Increase in Feed Concentration from 500 to 1500 mg/l Glucose.

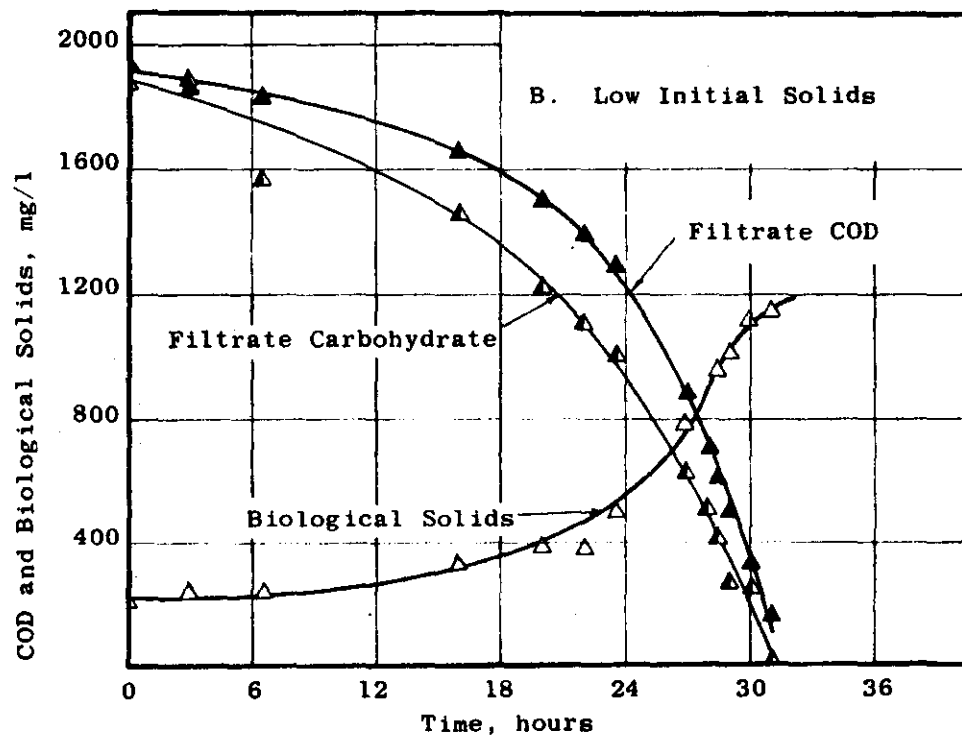
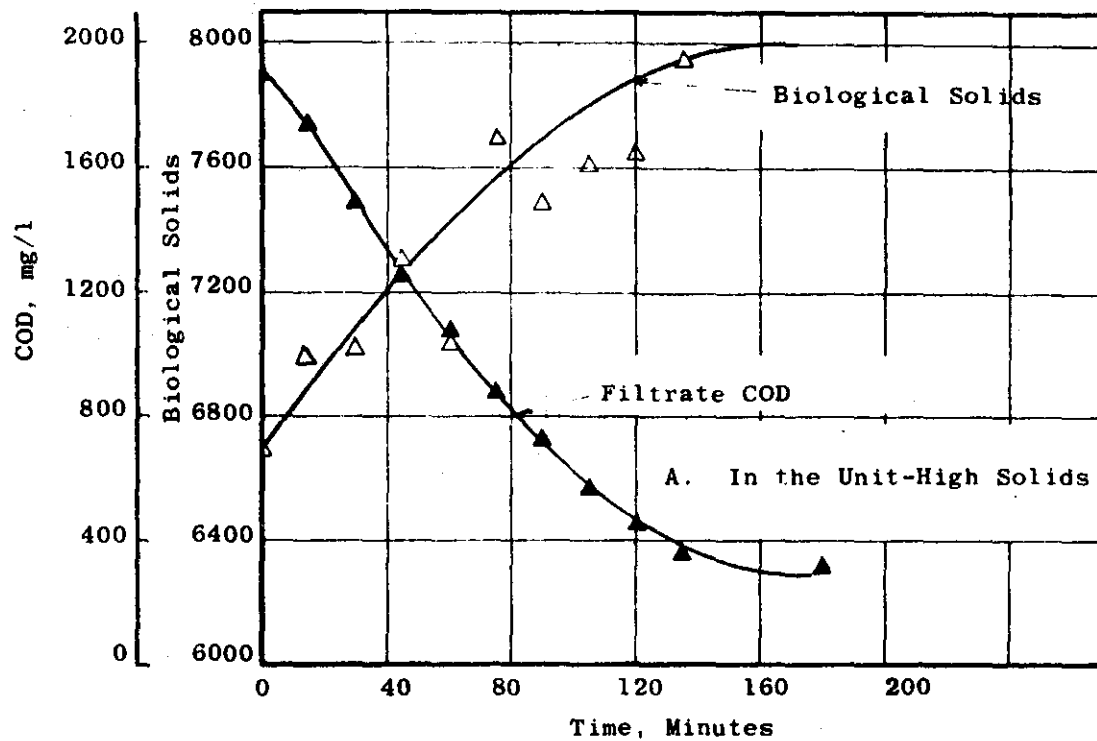


Figure 8. Response of Batch-operated Extended Aeration Activated Sludge to Quantitative Shock Loading with 2000 mg/l Glucose.

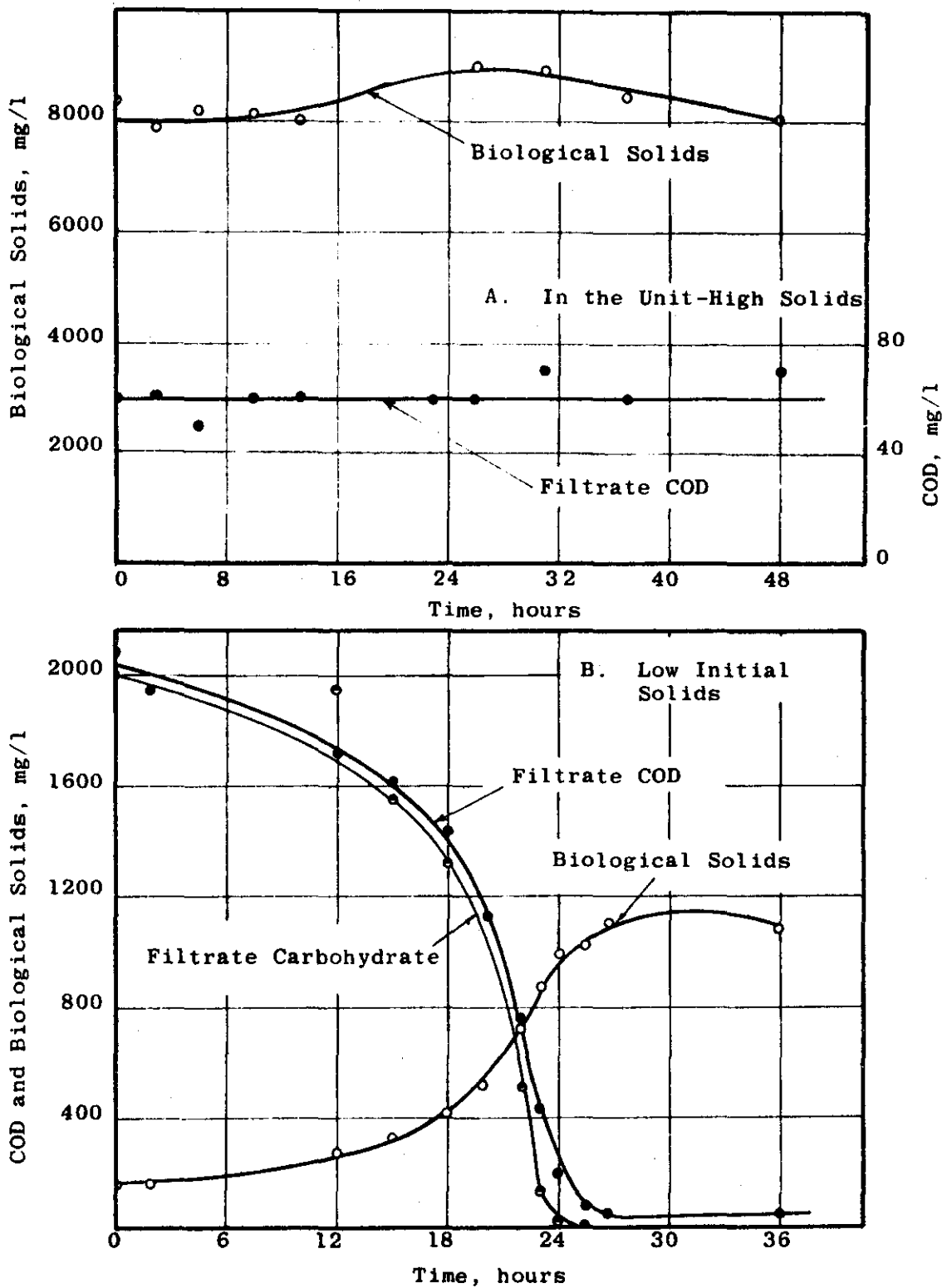


Figure 9. Response of Continuous Flow Extended Aeration Activated Sludge to Quantitative Shock Loading Applied as an Increase in Feed Concentration from 500 to 2000 mg/l Glucose.



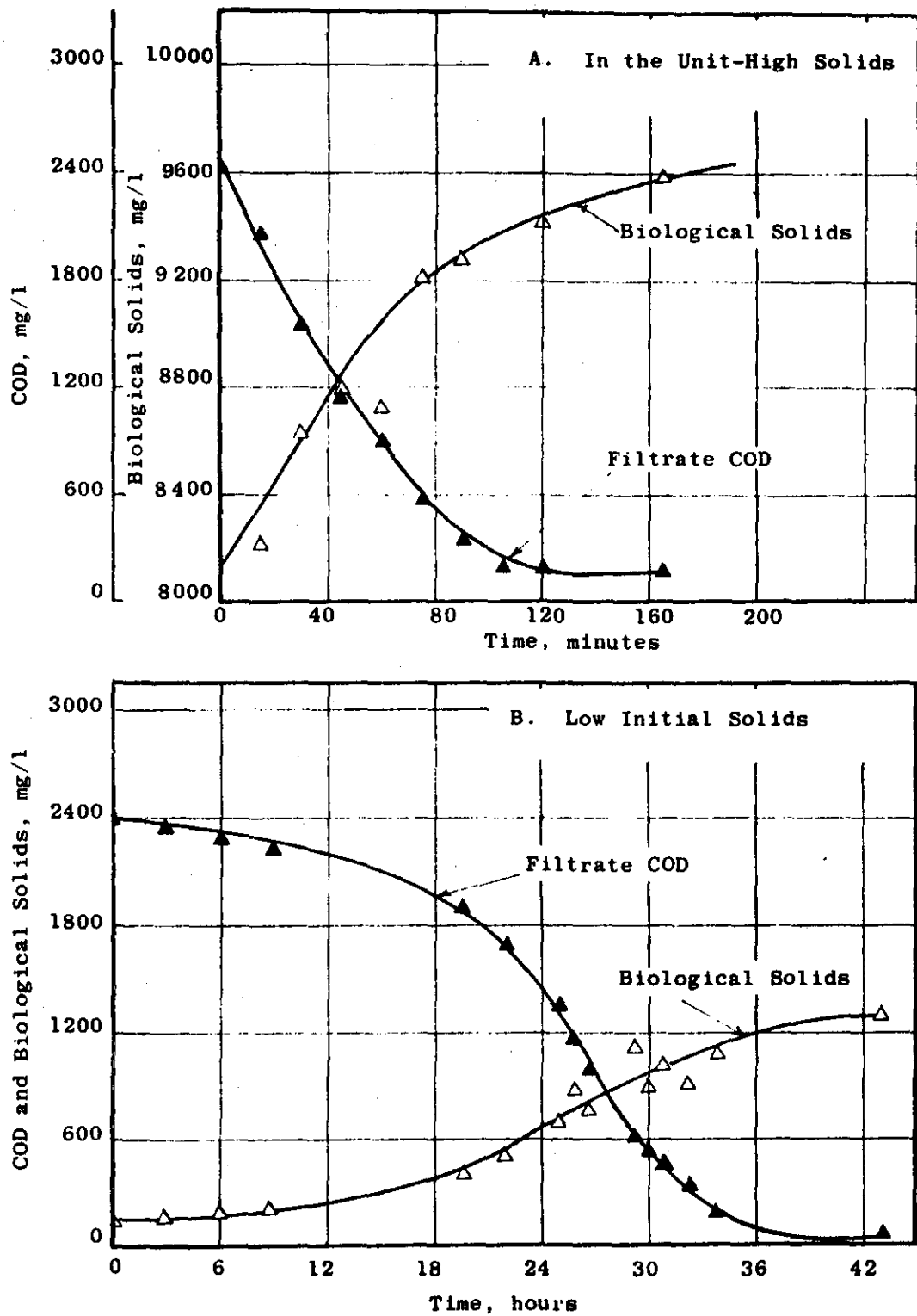


Figure 10. Response of Batch-operated Extended Aeration Activated Sludge to Quantitative Shock Loading with 2500 mg/l Glucose.

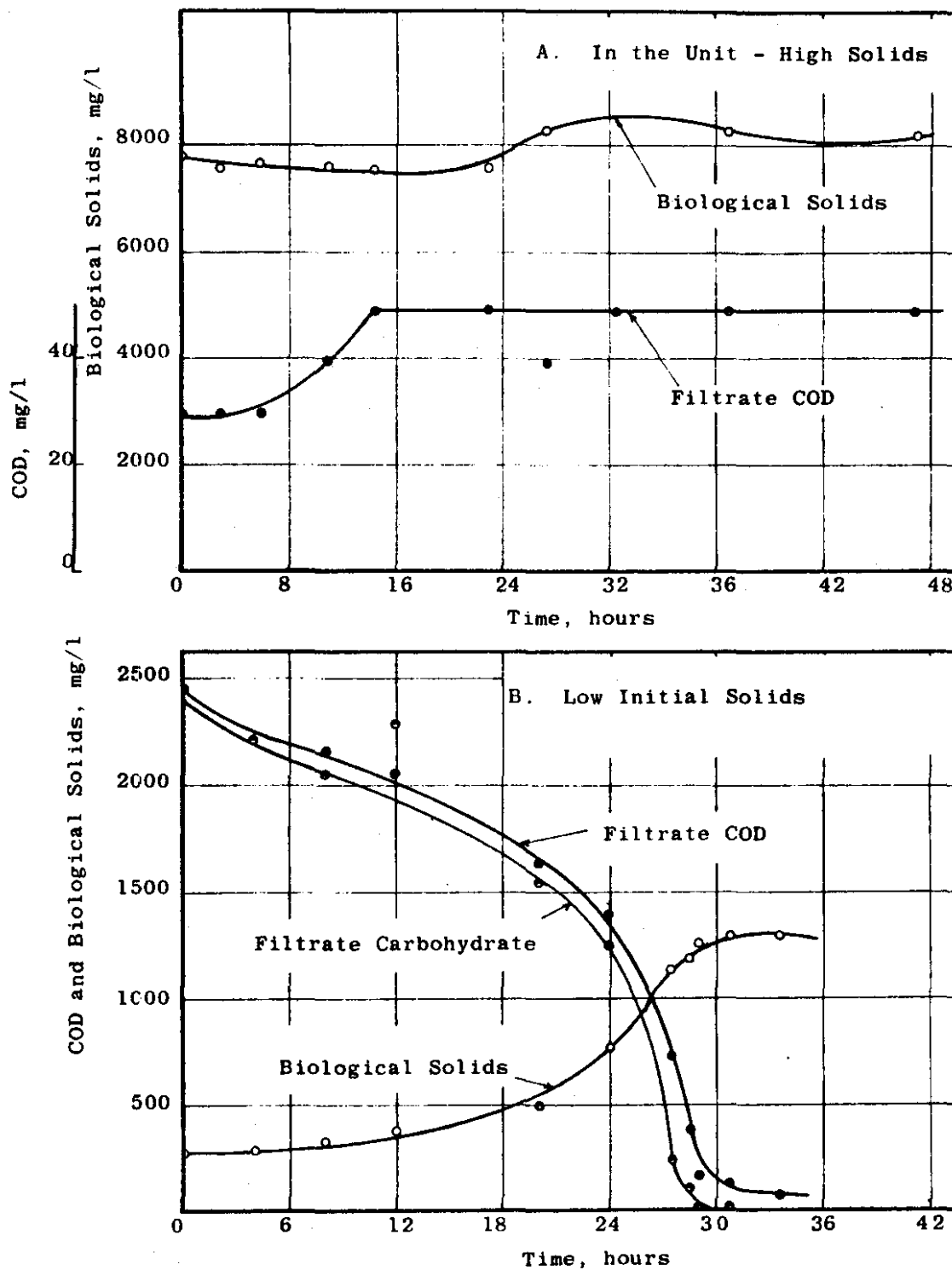


Figure 11. Response of Continuous Flow Extended Aeration Activated Sludge to Quantitative Shock Loading Applied as an Increase in Feed Concentration from 500 to 2500 mg/l Glucose.

portion of these products was subsequently utilized. Comparison of Figures 4A and 4B indicates that the high residual COD observed in the unit (see Figures 2 and 3) was probably due to very gradual accumulation of biologically "hard" end products of metabolism rather than to diminishing ability of the sludge to remove carbon source.

Figure 5A shows the response of the continuous flow extended aeration sludge when the glucose concentration in the feed was increased from 500 mg/l to 1000 mg/l. This shock load had essentially no effect on the performance of the system. There was no disruption of COD removal efficiency and little or no change in biological solids concentration. Figure 5B shows the results of a batch experiment using the same sludge at a lower concentration. The sludge was fed with 1000 mg/l glucose. The COD was removed in approximately 23 hours, and no significant quantity of metabolic products accumulated in the medium.

Figures 6 through 11 show results for similar experiments in which the sludge was subjected to shock loads of 1500, 2000, and 2500 mg/l glucose. At the 1500 mg/l shock loading level the carbon source was rapidly removed under the batch operation conditions (Figure 6); under continuous flow operation (Figure 7) there was no effect on either biological solids concentration or COD removal. Only when the shock loading was increased to 2000 mg/l glucose in the continuous system was an increase in the biological solids concentration observed (from approximately 8000 mg/l

to 9000 mg/l, Figure 9A). However, the COD of the mixed liquor remained low (60 mg/l) during the entire period of shock loading. Within 48 hours the biological solids concentration returned to its pre-shock load level. When the glucose concentration was increased to 2500 mg/l (Figure 11A), both the biological solids and filtrate COD concentration increased in the continuous flow system. However, the increase in filtrate COD was rather small. Biological solids increased from 7400 to approximately 8500 mg/l and the filtrate COD increased from 30 to about 50 mg/l. The above results clearly indicate that with respect to their biological efficiency of substrate utilization, "total oxidation" processes can withstand rather severe shock loads. For all experiments the lowest dissolved oxygen concentration recorded in the aeration chamber was 4 mg/l. Dissolved oxygen in the settling chamber under continuous flow operation averaged slightly above one mg/l.

It should be noted that during the experiments conducted under continuous flow conditions, the settling chamber wall was in place at all times except for a brief period immediately before the shock was administered. The zero time sample was taken during this period. Samples for biological solids concentration were obtained directly from the aeration chamber. Therefore the fact that there was little change in biological solids concentration during the step increase in influent feed concentration should not be interpreted unequivocally as an indication that the carbon source was not partially channelled into synthesis of

sludge at the 1000 and 1500 mg/l glucose shock levels. The upflow settling chamber contained a considerable amount of sludge during the period of increasing feed, and there was exchange of biological solids in the settling and aeration chambers, i.e., sludge growth in response to these shocks could have been swept into and accumulated in the settling chamber. At the high shock levels (2000 and 2500 mg/l glucose) an increase in biological solids concentration in the aeration chamber was noted during the shock period.

Table II shows values of specific substrate utilization rate calculated for each shock load experiment. The specific substrate utilization rate values for batch experiments were obtained by dividing the amount of COD removed by the product of the average value of biological solids concentration and the time required to remove the COD.

The specific substrate utilization rate for the continuous unit was calculated as follows:

$$\text{Specific substrate utilization rate} = \frac{\text{COD}_f - \text{COD}_e}{\bar{t} \cdot \bar{S}}$$

where

$\text{COD}_f$  = COD in the feed, mg/l

$\text{COD}_e$  = COD in the effluent, mg/l

$\bar{S}$  = mean value of biological solids concentration in g/l during the shock load

and  $\bar{t}$  = mean residence time in the unit in hours

TABLE II  
SPECIFIC SUBSTRATE UTILIZATION RATE

Figure Number	Date	Age of Sludge* Days	In the Unit (High Solids) mg COD/gram SS/hour	Separate Batch (Low Solids) mg COD/gram SS/hour
<u>Batch Operation</u>				
4	12- 4-67	249	99.6	90.0
6	1- 6-68	282	97.2	83.3
8	1-24-68	300	98.6	82.0
10	2- 9-68	316	126.8	94.0
<u>Continuous Operation</u>				
5	3-28-68	364	4.9	92.2
7	4- 4-68	371	7.6	153.0
9	4-12-68	379	9.8	141.0
11	4-25-68	392	13.6	99.8

\*Time in days since startup of the extended aeration unit

From Table II it can be seen that the specific substrate utilization rate did not deteriorate during the period of this experimentation. The low values of specific substrate utilization rate under continuous flow operation are due to the fact that the entire detention period (24 hours) in the unit was used in the calculation, and the shock load was applied gradually, i.e., a step increase in organic loading. Comparison of these low rates with the higher values obtained for the accompanying low solids batch studies helps explain why the sludge under continuous flow operation could successfully handle the shock loads; its substrate removal

capability was much higher than was needed to accommodate the shocks at the detention time employed. Even at the most severe shock, the expressed specific substrate removal rate which successfully accommodated the shock was approximately one-seventh of the specific removal rate of which the sludge was capable had it received the shock as a slug dose. The low solids batch experiments also indicate that the activity of the sludge, during either batch or continuous flow operation, varied from 92.2 to 153 during the experimental period, but did not decrease as the sludge aged. In general, the specific substrate removal rate was somewhat higher for sludge tested during the period of continuous flow operation. This result would seem reasonable, since under batch operation the sludge was subjected to a feed-rest-feed cycle and, as seen previously, the feed was metabolized rapidly (e.g., see Figure 4A) and there was a very long resting (or endogenous) period wherein it would be expected that cell function and capability might be retarded.

The oxygen uptake values measured on washed cell suspensions are shown in Table III. The activity of the sludge, as measured by unit oxygen uptake, exhibited a somewhat decreasing trend; however, the specific substrate removal rates (Table II) did not exhibit a corresponding decrease. Therefore, it would appear that unit oxygen uptake cannot be used as a sole parameter for assessing (or predicting) the substrate removal capability of the sludge. The system has received no shock loads since April 25, 1968.

and unit activity of the sludge has not been measured since June 7, 1968; however, the bench scale plant is still providing excellent COD removal efficiency, measured on filtrate or supernatant, after 510 days of operation (as of August 21, 1968). The unit which was not subjected to any shock loads is providing similar results.

TABLE III  
OXYGEN UPTAKE VALUES OF WASHED EXTENDED  
AERATION ACTIVATED SLUDGE

Date	Age of Sludge* Days	Unit Activity mg O <sub>2</sub> /gram SS/hour
11-30-67	245	3.7
12-14-67	259	2.6
1- 6-68	282	3.7
1-24-68	300	3.5
2-14-68	321	3.1
3- 4-68	340	3.1
4- 2-68	369	2.6
6- 7-68	435	2.1

\* Time in days since startup of the extended aeration unit.

#### SUMMARY AND CONCLUSIONS

The results obtained during this investigation throw some light on the efficiency and duration of successful operation of extended aeration activated sludge plants. During the period of operation it was shown that the process could handle slug organic loadings. The high residual COD values observed are attributable to the fact that only a small amount of supernatant was discarded under this type



of operation. The buildup of residual COD may be due at least in part to organic metabolic products which are biologically nondegradable. It may also have been due in part to a buildup of inorganic salts from the synthetic waste. During the present investigation, no attempt was made to determine the nature of the residual COD during batch operation in view of the fact that the continuous flow operation (of major interest in field application) yielded such excellent COD removal efficiency under both steady and shock loading conditions.

Comparison of Figures 4B through 11B show, in general, that for similar shock loadings the sludge during periods of continuous flow operation responded more successfully than under the period of batch operation. This finding is substantiated by comparison of the specific substrate removal rates calculated for the comparable low-solids studies in Table II. Furthermore, the data presented in Table II indicate that the continuously fed systems could be expected to handle higher quantitative shock loads (as step increases) than those employed in this study. It can be seen from Figure 1 that biological solids did not steadily increase under conditions of no sludge wasting (extended aeration process) as would be expected if the sludge were not subject to some autooxidation. The specific substrate removal capability of the sludge was indeed lower than values we have observed in other work for systems in which sludge wastage was practiced. However, if the lower specific

substrate removal rate is due to a high fraction of biologically inert sludge, this biologically inert fraction (as assessed by specific substrate removal rate) did not increase during the period from November 30, 1967, to June 7, 1968. Apparently during the year of operation prior to initiating the shock loading studies, the sludge had attained a low but relatively stable capacity to remove the carbon source. Our research on the total oxidation process is still in progress, and the laboratory unit has now been in operation for nearly eighteen months. At the time of preparing this manuscript (August, 1968), it is still yielding excellent COD removal efficiency (and is operating under continuous flow conditions). While there is much work yet to be reported on our findings, and while there is much research yet to be done on the system, the findings herein reported would seem to us to augur well for the process.

#### ACKNOWLEDGEMENT

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## LITERATURE CITED

1. Porges, N., L. Jasewicz, and S. R. Hoover, "Biochemical Oxidation of Dairy Wastes. VII. Purification, Oxidation, Synthesis and Storage." Proceedings, 10th Industrial Waste Conference, Purdue University, 135-146 (1955).
2. Kountz, R. R., "Total Oxidation Treatment." Proceedings 11th Industrial Waste Conference, Purdue University, 157-159 (1956).
3. Forney, C., Jr., and R. R. Kountz, "Activated Sludge Metabolism." Proceedings 13th Industrial Waste Conference, Purdue University, 313-320 (1958).
4. Symons, J. M., and R. E. McKinney, "The Biochemistry of Nitrogen in the Synthesis of Activated Sludge." Sewage and Industrial Wastes, 30, 874-890 (1958).
5. Kountz, R. R., and C. Forney, Jr., "Metabolic Energy Balances in a Total Oxidation Activated Sludge System." Sewage and Industrial Wastes, 31, 819-826 (1959).
6. Busch, A. W., and N. Myrick, "Food Population Equilibria in Bench-scale Bio-oxidation Units." Sewage and Industrial Wastes, 32, 949-959 (1960).
7. Washington, D. R., and J. M. Symons, "Volatile Sludge Accumulation in Activated Sludge Systems." Journal Water Pollution Control Federation, 34, 767-789 (1962).
8. McWhorter, T. R., and H. Heukelekian, "Growth and Endogenous Phases in the Oxidation of Glucose." Advances in Water Pollution Research, 2, 419-436. The MacMillan Company, New York (1964).
9. Standard Methods for the Examination of Water and Waste Water, 12th Edition, American Public Health Association, New York (1965).
10. Gaudy, A. F. Jr., "Colorimetric Determination of Protein and Carbohydrate." Industrial Water and Wastes, 7, 17-22 (1962).

**APPENDIX 3**

**Gaudy, A. F. Jr., Yang, P. Y., and Obayashi, A. W., "Studies on the Total Oxidation of Activated Sludge With and Without Hydrolytic Pretreatment." Journal Water Pollution Control Federation, 43, 40-54 (1971).**

STUDIES ON THE TOTAL OXIDATION OF ACTIVATED SLUDGE WITH AND  
WITHOUT HYDROLYTIC PRETREATMENT

By

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In the fall of 1966, studies pertinent to metabolic behavior of heterogeneous microbial populations (activated sludges) after completion of the growth and substrate removal phases were undertaken in the authors' laboratories. This phase of the metabolic cycle is usually termed the "endogenous" phase. However, it is emphasized that when this term is applied to heterogeneous populations, it cannot be considered simply as a period for utilization of internal cellular carbon sources by individual cells, since a heterogeneous population represents an extremely complex ecosystem in which various species may serve as food for other members of the population.

Recently, Thabaraj and Gaudy have studied some effects of the past growth history of sludge on biochemical changes and kinetic behavior during its aerobic decomposition in the subsequent endogenous phase (1). It was observed that under prolonged endogenous metabolism, total oxidation of an amount of sludge equal to that synthesized in the previous growth phase was possible. These studies have obvious ramifications toward understanding the basic metabolic and ecological principles and

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concepts underlying the so-called total oxidation or extended aeration process. Indeed, a part of the reason for making the studies cited above was to gain better understanding of the operational behavior of an extended aeration process pilot plant which has been in operation for a long period of time in the authors' laboratory.

Portions of the results of the pilot plant studies have been previously presented. Long-term pilot plant studies on the ability of an extended aeration process to withstand quantitative shock loads have been reported (2). Also, a continuing pilot plant study on the operational stability of an extended aeration process has been under way since March, 1967, and operational results as well as those of auxiliary experiments through January 13, 1969, have recently been reported (3).

At the start of the pilot plant experimentation, the purpose was to determine whether biological solids would continue to accumulate in the system and, if so, whether such accumulation would lead to increased inactivity of the biological sludge to such an extent as to cause a biochemical failure of the system. It was anticipated that the problem of inadvertent loss of solids in the plant effluent, which is often encountered in the field and which has been encountered in laboratory pilot plant studies as well, could also be a problem in the present study. In order to provide for positive retention of suspended solids, the effluent was routinely collected and passed through a centrifuge. Thus, all solids were returned to the aeration chamber. The amount of solids removed for various analyses and auxiliary experiments was less than 0.2 per cent.

It was found after nearly two years of operation that the system provided excellent biochemical efficiency, and that the sludge

concentration did not continue to build up in the system. A so-called equilibrium solids concentration was not attained, and the solids concentration passed through cycles of increase and of decrease. During periods of decreasing solids concentration, the COD removal efficiency of the system was not grossly impaired. Since no solids were wasted, either inadvertently or purposely, the only logical interpretation of these results was that during the decreasing period of solids concentration, those biological solids which had previously been accumulating were serving now as carbon source for other members of the population. A number of natural biological causations and theoretical possibilities for periodic acceleration of the autodigestive process of the biomass were presented and augmented by analysis of the results of other workers in the pollution control field, of workers in the basic microbiological field, and results obtained in the authors' laboratory. The results indicated that the theory of "total oxidation" was not inconsistent with sound microbiological theory. While there are indeed engineering problems, for example, retention of solids in the system during times of high solids concentration, etc. these may be subject to various engineering solutions; whereas, if the concept were not biochemically or metabolically sound, there would be little scientific justification for its continued or accelerated use.

Because of the results obtained and the tentative conclusions concerning the theoretical soundness of the process as well as the important ramifications its use has toward solving the problem of sludge disposal, it seemed that extension of the scope of applicability of the extended aeration process, especially in the industrial waste field, was a definite possibility. There were three avenues for continued

research which were of immediate importance. First it was desirable to continue the pilot plant operation and the auxiliary experiments on a routine basis for further assessment of its operational characteristics. Also, the sludge which had been developed over such a long period of time represented a unique and complicated ecological system which it was felt should be perpetuated for further examination. Secondly, it was essential to initiate experiments to determine the ability of various heterogeneous microbial populations to metabolize various fractions of other cells, since such studies could give insight into natural causation for the periods of accelerated autodigestion which had been observed in the pilot plant operation. Thirdly, it was essential to initiate studies to devise and to test possible bioengineering process innovations for operational control of the system. In the present report, results pertinent to these three investigational approaches are presented.

#### MATERIALS AND METHODS

The continuous flow pilot plant employed in these studies has been described in detail in a previous report (3). The total operating volume was 9.4 liters, separated into a 6.2-liter aeration chamber and a 3.2 liter settling chamber. The overall detention time was twenty-four hours (16 hr aeration, 8 hr settling). The composition of the synthetic waste was the same as that previously used. The carbon source was glucose at 500 mg/l, providing an organic loading of 50 lbs COD/1000 cu ft aeration capacity. Effluent from the settling chamber was collected in a holding tank (see Figure 1).

Once or twice daily the contents of the effluent holding tank were passed through the Sharples centrifuge, and all solids were returned to



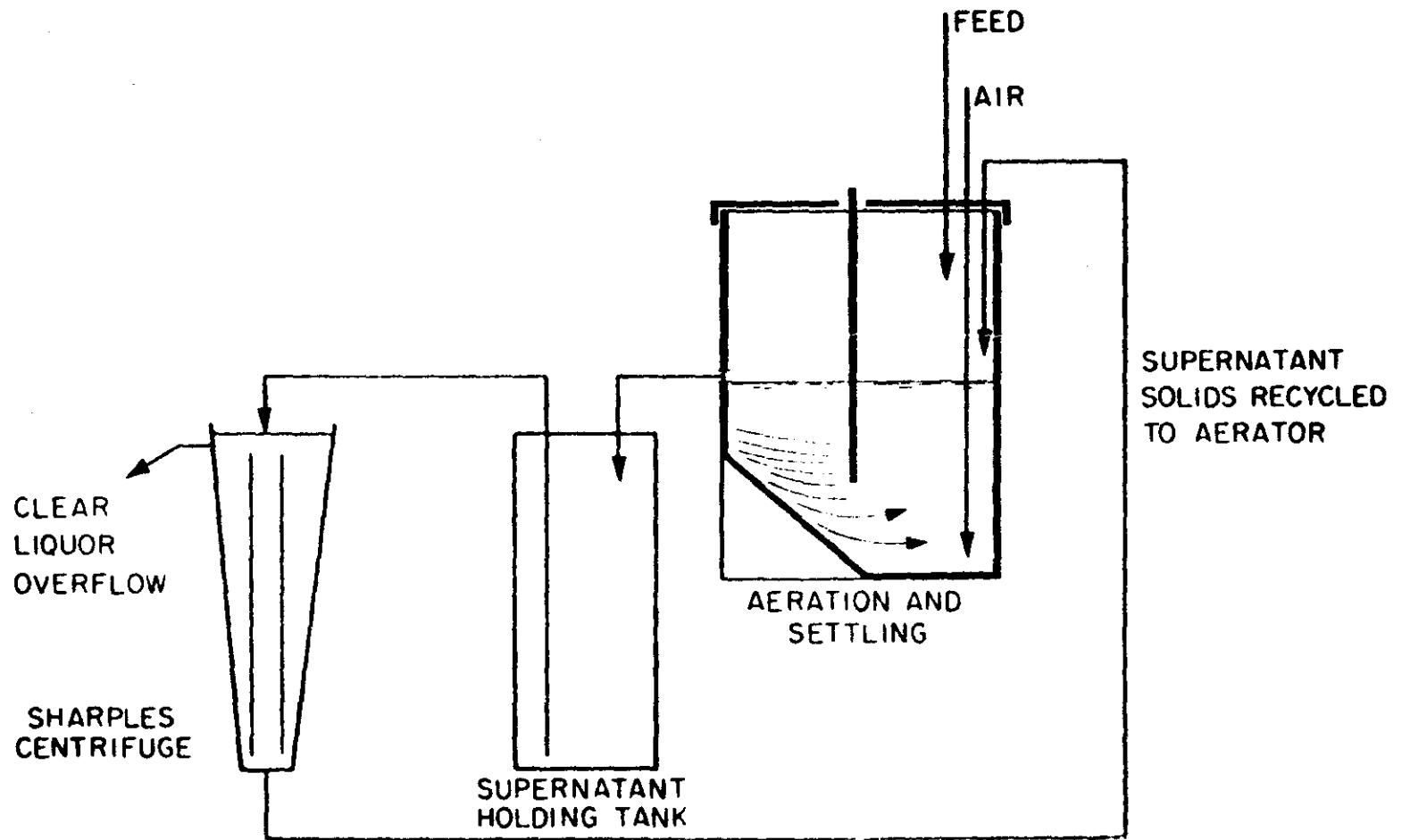


Figure 1 - Continuous flow extended aeration pilot plant.

the aeration chamber. In this way all biological solids were retained in the system. The only solids removed from the system were those taken out during sampling, and a small amount taken out for auxiliary experiments (no more than 0.2 per cent). At two to three-week intervals the unit was run in batch operation for one day to assess the substrate removal rate. At this time a small sample of sludge was also taken for a batch experiment to determine patterns of substrate removal and growth for a low initial inoculum of the population. Also, at about two to three-week intervals a small sample of sludge was taken to determine the endogenous  $O_2$  uptake rate of the extended aeration sludge.

Analyses performed on the system to assess operational behavior included biological solids concentration in the unit and in the effluent [membrane filter technique (3)], filtrate COD (3), protein and carbohydrate content of the sludge (3). At times, COD's were also run on the unfiltered effluent. At times, samples of the effluent were analyzed for  $NH_3-N$ ,  $NO_2$ , and  $NO_3$ , but such analyses were not performed on a routine or regular basis. Periodically, DO concentrations in the aeration chamber and in the settling compartment were measured electrometrically. Also, the pH of the system was assessed daily. Special experimental procedures to assess the availability of cellular materials as carbon source for the cell population are given along with the results pertaining to experiments on sludge "sonicate" and sludge hydrolysate.

## RESULTS

### Laboratory Pilot Plant Studies

In the previous report, the performance data for the extended

aeration pilot plant were plotted for the first 655 days of operation(3). In the present report, performance data are plotted beginning with day 600 (see Figure 2).

A brief review of the history of the system during the first 600 days of operation should aid in evaluating the results presented herein. Solids concentration in the system after 285 days of operation was slightly above 8000 mg/l. The solids concentration then decreased, and by day 310 was between 2000 and 3000 mg/l. Again, it is emphasized that solids were not "wasted" purposely or inadvertently. After completion of this downward cycle, the sludge concentration in the system followed an irregular but generally increasing trend. COD removal efficiency (filtrate) remained at approximately 90% or above throughout the decreasing and increasing cycle, and sludge settleability was very good (see days 600-630, Figure 2). After nearly two years of operation, the protein content of the sludge had decreased to between 20 and 25%, and the carbohydrate content had risen to approximately 20%. Also, as seen in Figure 2, the sludge retention in the settling chamber began to decrease (see days 637-655). As always, these solids were collected and returned to the aeration tank. All of these data were reported in detail previously (3).

Between day 655 and 679, the system behaved much as it did between day 637 and 655. Unfortunately, the data for days 655 and 679 had not been transferred to the summary sheet log for the pilot plant and the raw data sheets for this period were lost. One biological solids determination and one sludge protein value had been noted elsewhere and were available for plotting. Also, the solids concentration on January 27, 1969 (day 669, not plotted) had been noted in the previous report; it

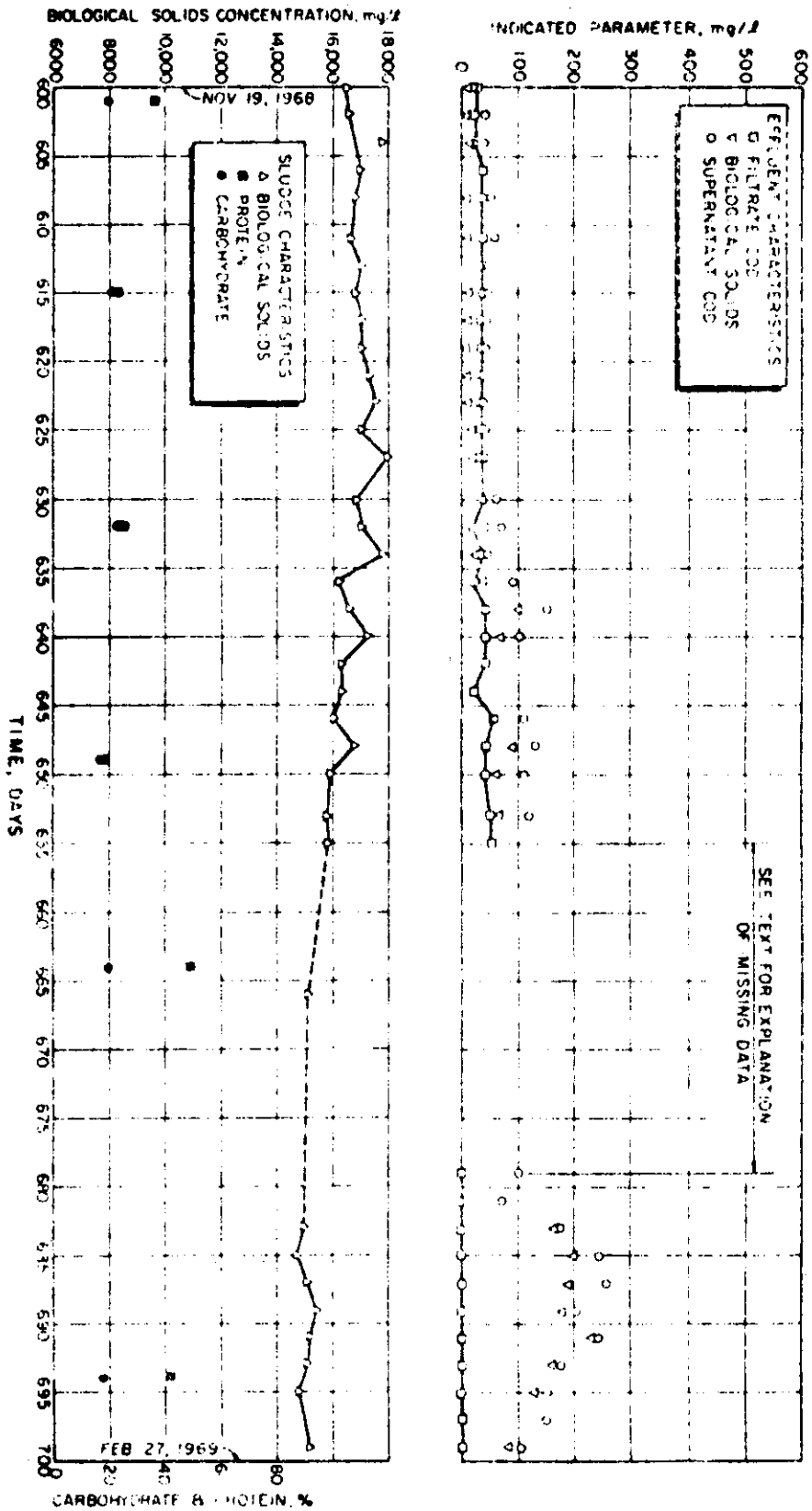


Figure 2 - Performance data continuous extended operation pilot plant 11/19/68 to 2/27/69

was 14,600 mg/l. Between day 680 and 700, the biological solids in the effluent increased to approximately 200 mg/l, and then began to decrease. During this period, the biochemical efficiency of the system (as measured by filtrate COD) was extremely high. The biological solids concentration in the system remained at approximately 15,000 mg/l.

In Figure 3 the performance of the plant for days 700 to 800 is shown. By day 706 rather good sludge separation was regained; the system efficiency as measured by supernatant COD was approximately 90%; as measured by filtrate (biochemical efficiency) over 99% of the COD was being removed. It is seen that in the days following, the system underwent a serious disruption with regard to effluent solids. On day 718 the pH which had been running at the normal level, 7.0, began a slow decline. By day 729 it had dropped off to 6.0. Over the next 50 days the pH was adjusted gradually to 7.0 by adding small amounts of NaOH and by increasing the amount of buffer in the synthetic waste. During the period of excessive solids loss, portions of the sludge blanket in the settling chamber were carried over. The solids carryover was not due to dispersed growth, but to floc carryover. The floc had a rather light, fluffy character, but was not overly filamentous in nature. Biochemical efficiency remained close to 90%. Again, it is emphasized that the solids were not lost to the system, but were collected, centrifuged, and returned to the aeration chamber. During this period there was a noticeable decrease in protein content of the sludge, and carbohydrate content remained at approximately 20%. Supernatant COD is not plotted beyond day 730, since the effluent solids were rather high and they remained so thereafter. The system recovered from the severe solids carryover experienced between day 740 and 750, but it is seen that solids continued to

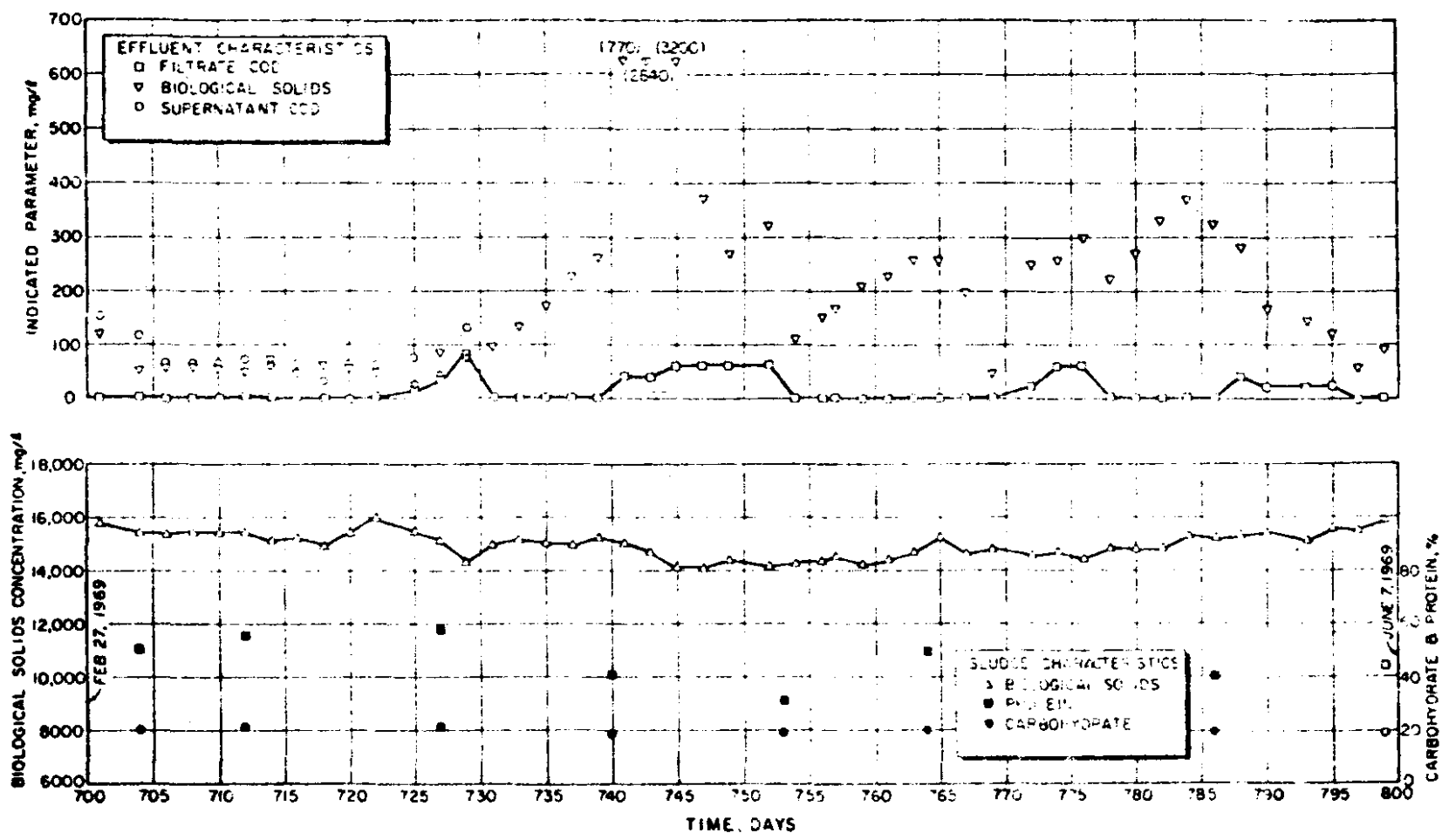


Figure 3 - Performance data continuous extended aeration pilot plant 2/27/69 to 6/7/69.

be carried over for an extended period. The biological solids concentration in the system during the 100-day period shown in Figure 3 fluctuated between 16,000 and 14,000 mg/l.

During the following 100 days of operation (see Figure 4), the solids concentration in the settling tank overflow fluctuated between 60 and 540 mg/l. These solids did not consist of carried-over floc, but of a mixture of dispersed growth and small floc particles, and both feeding and inactive protozoa were noted. The biochemical efficiency remained good, at times better than 99%. The sludge concentration decreased somewhat, but did not undergo an accelerated period of auto-digestion comparable to that experienced previously between days 285 and 310 (3). The carbohydrate content remained at approximately 20%, and there was an increasing trend in protein content.

In Figure 5 the operational behavior of the system from day 900 to day 1000 is shown. Early during this period, the biological solids concentration experienced a slight downward cycle, to nearly 12,000 mg/l. Thereafter the sludge concentration fluctuated, first increasing to nearly 16,000 mg/l, and then decreasing somewhat. The carbohydrate content remained at approximately 20% or lower, and the protein content was in general above 50%. The biochemical efficiency of the system averaged approximately 90%. During this time significant concentrations of biological solids were carried over in the settling tank effluent. Throughout this period, as in the previous 100 days of operation, the sludge level in the settling compartment was usually less than  $\frac{1}{4}$ -inch below the outlet line; thus the supernatant layer was extremely thin, and since the sludge concentration in the settling compartment had been previously shown to be generally the same as in the aeration compartment,

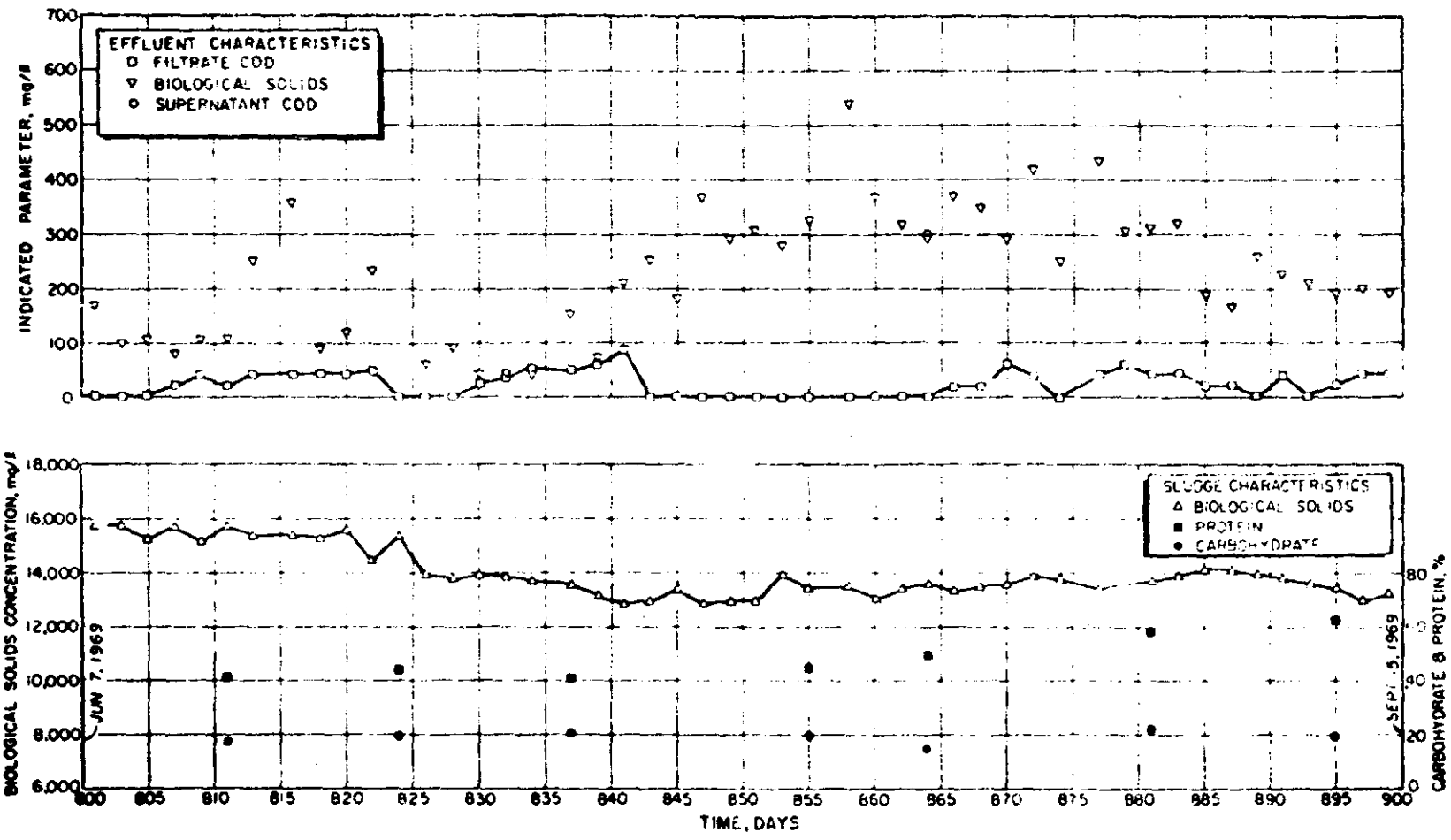


Figure 4 - Performance data continuous extended aeration pilot plant 6/7/69 to 9/15/69.



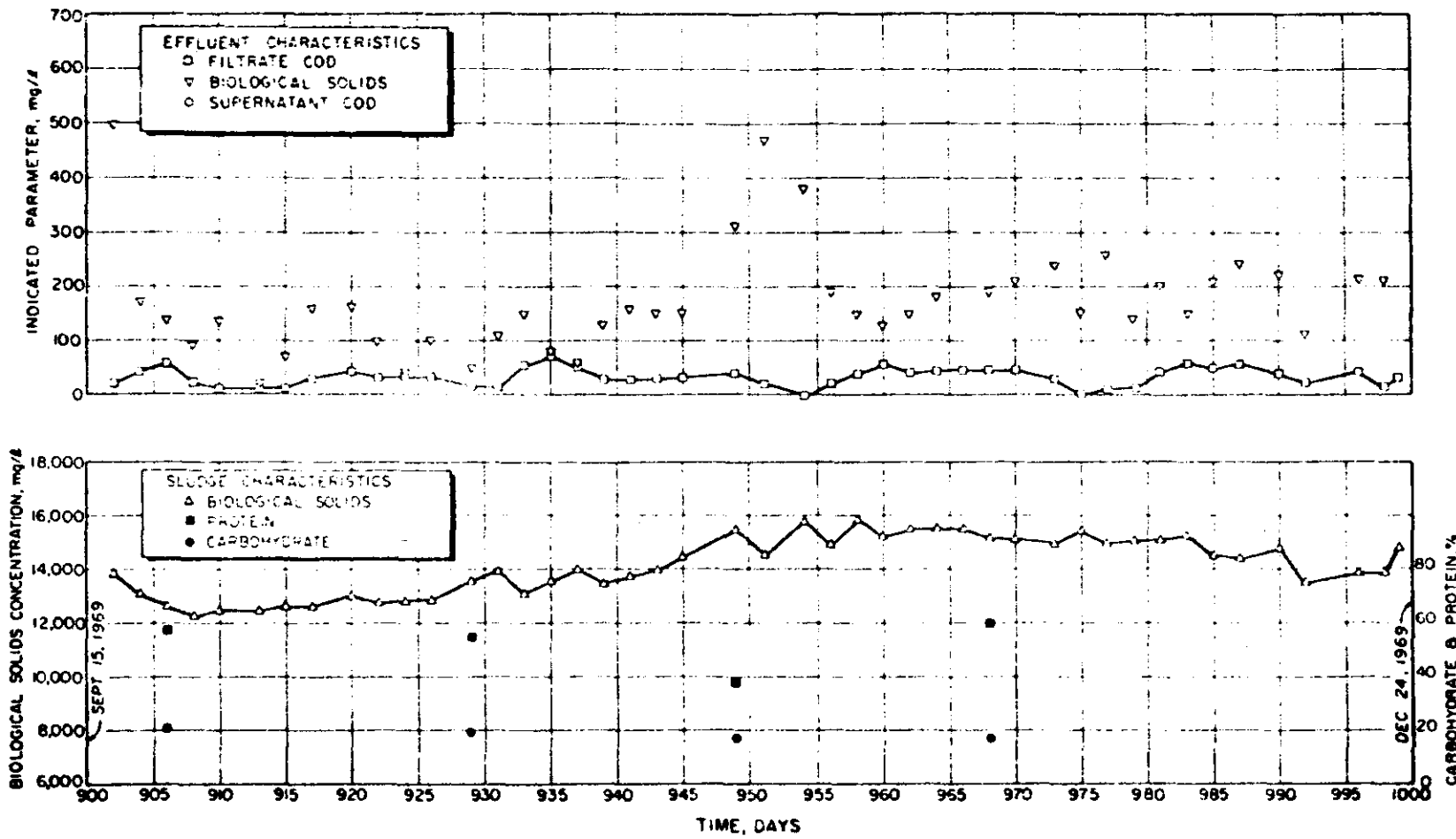


Figure 5 - Performance data continuous extended aeration pilot plant 9/15/69 to 12/24/69.

this high concentration, approximately 12,000 to 16,000 mg/l, militated against good mixed liquor separation.

It should also be mentioned here that routine checks on dissolved oxygen content of the mixed liquor in the aeration chamber and of the effluent from the settling compartment yielded values of 6 and 2 mg/l respectively. It is noted that excess nitrogen source is present in the synthetic feed to the pilot plant ( $COD/N = 530/53 = 10/1$ ). At times, although not on a routine basis,  $NH_3-N$ ,  $NO_2-N$ , and  $NO_3-N$  in the effluent were determined. These analyses were made primarily to determine whether the plant was producing a nitrified effluent. Nitrate-N in the effluent and, generally, traces of nitrite-N have always been observed. At times the sum of ammonia, nitrite and nitrate nitrogen has been equal, or nearly so, to the nitrogen concentration in the feed, providing some indication that the nitrogen source has at times been re-used by the sludge. On a few occasions volatile solids content of the sludge has been determined; the values range from 90 to 92 per cent.

Periodically, a small sample of sludge was removed from the system and its endogenous  $O_2$  uptake was determined manometrically using a Warburg respirometer at  $25^\circ C$  and 110 oscillations/min. Unit  $O_2$  uptake measurements ( $mg O_2/hr/gm$  sludge) for the first 650 days of operation were given in the previous report (3). In Figure 6 the values during the entire 1000-day period are shown; the early portions of the curve have been discussed previously. The rising trend in unit oxygen uptake corresponded to the period of accelerated autodigestion of the sludge previously reported (3). After 600 days of operation, it appeared that the endogenous respiration rate was attaining a lower limit. Since that

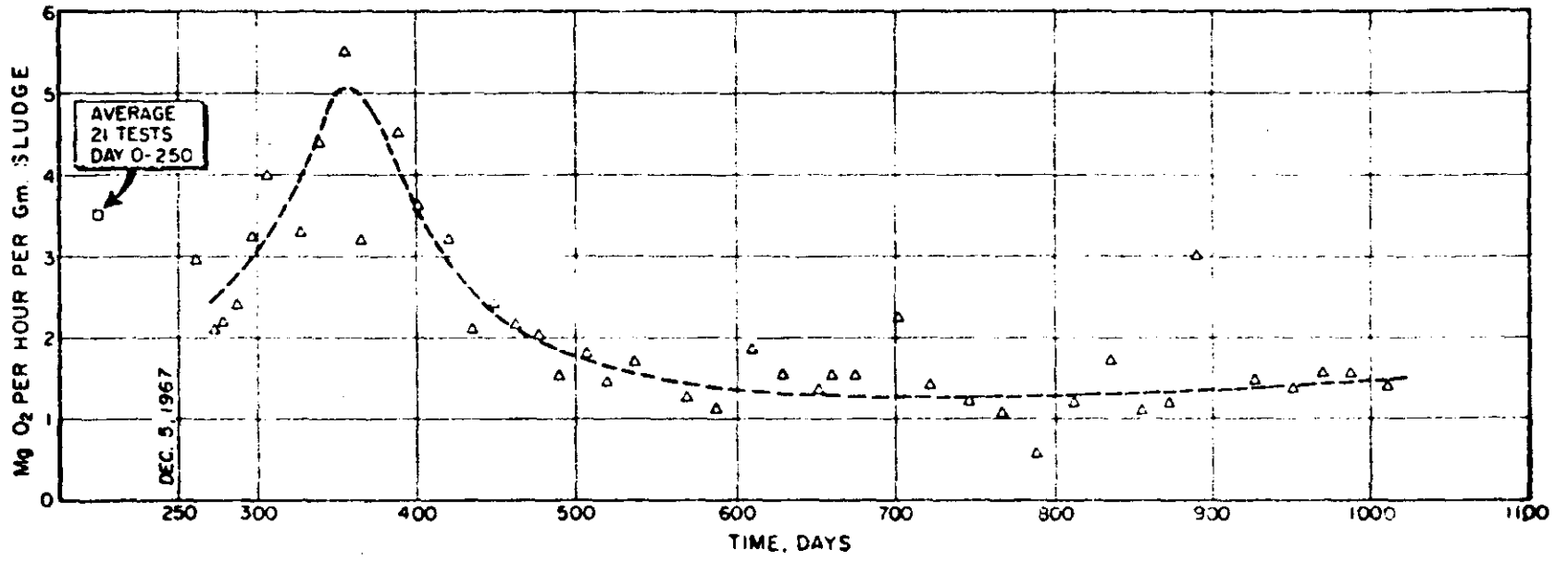


Figure 6 - Effect of aging on endogenous O<sub>2</sub> uptake of extended aeration activated sludge

time it can be seen that it has fluctuated somewhat, but the sludge has apparently attained a value of approximately 1.5 mg O<sub>2</sub>/hr/gm sludge. This value was previously found to be approximately 5 to 10% that for young cells grown from a small inoculum of cells taken from the aeration chamber.

At two to three-week intervals, the feed pump to the pilot plant was stopped and the feed was added on a batch basis to assess the rate of COD removal. As in similar studies during the first year and a half of operation, the carbon source was always removed rapidly. In Figure 7 the results for four of the batch studies made during the last 300 days of operation are shown. It can be seen in all four graphs that the feed COD was removed rather rapidly, within approximately 30 minutes. The COD removal rate per unit weight of sludge was approximately 70 mg/hr/gm. This value compares with rates ranging from approximately 300 to 700 mg/hr/gm, which can be calculated from data from more "normal" sludge systems, i.e., ones in which sludge wasting was practiced (4)(5)(6). Thus the substrate removal rate as well as the endogenous O<sub>2</sub> uptake rates are lower, in roughly the same proportion, for sludges developed under conditions of total solids retention (extended aeration) than for systems in which sludge is routinely wasted.

The major facts thus far delineated are that while the substrate removal rate per gram of sludge is rather low, and while the endogenous respiration rate is rather low, purification of the organic loading which was fed (which is rather high for extended aeration processes) by this three-year old sludge, even under shock (batch loading) conditions, proceeds in approximately 1/32 (0.5/16) of the detention time which is provided in the aeration chamber. It should be apparent from these

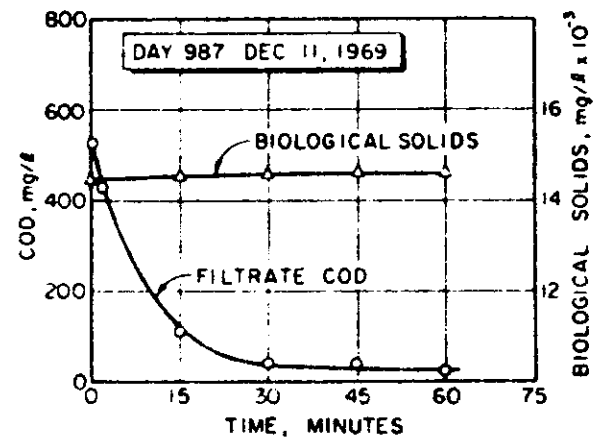
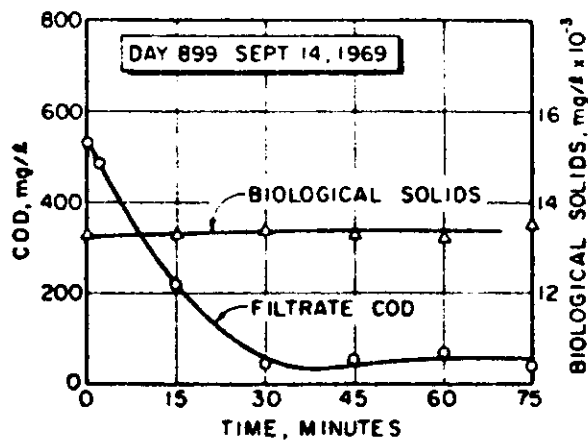
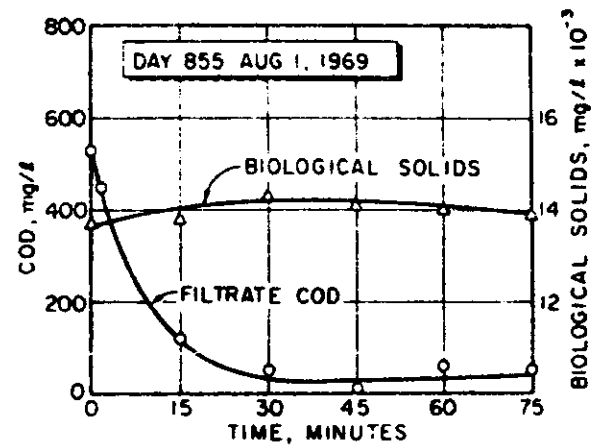
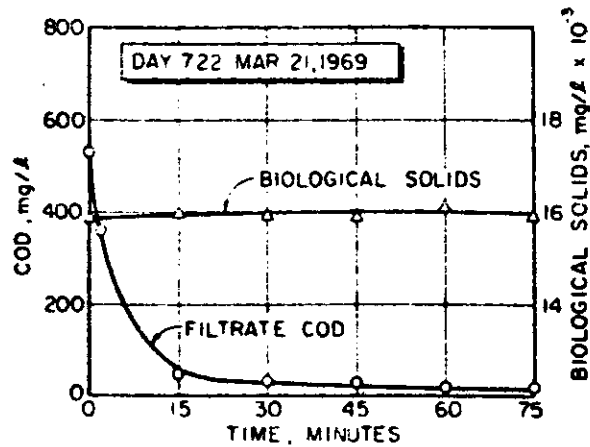


Figure 7 - COD removal capability of extended aeration activated sludge on the day indicated.

results as well as the operational data on the pilot plant, i.e., consistent attainment of approximately 90% COD removal based on filtrate, the lack of continual sludge buildup in the system, and the data on sludge composition, that after three years any significant inactive fraction has not built up to an extent which would cause the system to fail biochemically. These results, as well as previous data which have been presented (1)(3), reinforce the conclusion that the concept of total (biological) oxidation of sludge which has been biologically synthesized is not inconsistent with sound microbiological principles. As applied to the extended aeration process, the proof of the concept does not automatically prove the useful adaptation of the process, since the problem of retaining the solids is of prime consideration. It was seen in Figures 3, 4, and 5 that a large amount of solids would have left the system, had settling alone been the means of separating the cells. In view of the problems which a total oxidation process can solve, (e.g., oxidative disposal of the sludge), it would seem a wise expenditure of effort to seek engineering solutions to the problem of sludge retention rather than to overlook the advantages of the total oxidation process because there are settling problems from time to time. In order for the process to work, the particulate organic carbon synthesized in the system must be retained in the system and, as seen from the present results, if this can be done, cells which can degrade and metabolize other portions of the sludge will develop. Since in the extended aeration process the sludge itself is a major portion of the food supply, it is essential to gain information on the nature and ease of metabolism of this extremely complex "waste" and to seek possible engineering processes or treatments which can increase its metabolic availability. In the remainder of this

report, some of the studies undertaken by the authors along both of these avenues of investigation are presented.

#### Experiments on Sludge "Sonicate"

It is to be expected that the availability of various fractions or components of microbial cells as food material for other cells in the system will vary, depending upon the type of cells present (both as feeders and as feed material). The purpose of one phase of these investigations is to determine the relative ease or difficulty with which cell components can be used as carbon source(s). There are various ways to categorize the organic cell fractions; for the purposes of the present studies, three major categories of cell components are being examined. If the cell wall-membrane of the microbial cell were ruptured, the organic material inside the cell (most of which is soluble) could become a source of food for other cells. The non-soluble shell or sac may also represent a source of food, but it must first be hydrolyzed enzymatically. A third possibly significant category for carbon source might be the slime (largely polysaccharide) layer or capsule surrounding the cell wall.

If a cell is to become usable as a metabolic carbon source, leading to its eventual "total oxidation," these fractions must be acted upon in the preparatory metabolic stages in order to become natural sources of food for other species. Some bacterial species may be ingested directly by predators or grazers, but for one bacterial cell to become food for another, the three fractions mentioned above must be put into metabolizable form. In an extended aeration system, these fractions may be utilized sequentially or concurrently, or perhaps not at all, depending upon the ever-shifting predominance of species in the feeding population.

of experiments were run in which the soluble cell components were used as feed in systems in which the seed population consisted of a small inoculum of cells grown from sewage, or cells from the extended aeration unit. In some cases there was a rather long lag period followed by very rapid growth, whereas in others the sonicate COD was removed with no acclimation period but at a rather slow rate. Also, cell sonicate has been fed to the extended aeration pilot plant. Results for one such experiment are shown in Figure 8. For this experiment, a heterogeneous microbial population was grown from a sewage seed, with glucose as a carbon source. The cells were then harvested, washed, and broken by ultrasonic vibration (Branson Model S-75). The insoluble material was removed by centrifugation; the soluble organics were passed through a membrane filter and the soluble material was then used as a carbon source. For the results shown in Figure 8, this material was fed as a shockloading, slug dose to the extended aeration pilot plant during a batch experiment, i.e., an experiment similar to a batch run of the type shown in Figure 7, except that the "sonicate" rather than the normal feed, glucose, was used. The initial protein and carbohydrate content of the feed material were approximately 45 and 10 per cent, respectively. It is seen in the graph at the left-hand side of the figure that a portion of the sonicate COD was removed very rapidly; approximately 50 per cent was removed in 15 minutes.

Some difficulty was experienced in running the carbohydrate and protein content on filtrate during this experiment. Therefore these data are not shown, because at best they would represent rough approximations. However, it did not appear that the rapid removal during the first ten minutes was due only to protein or to carbohydrate uptake by



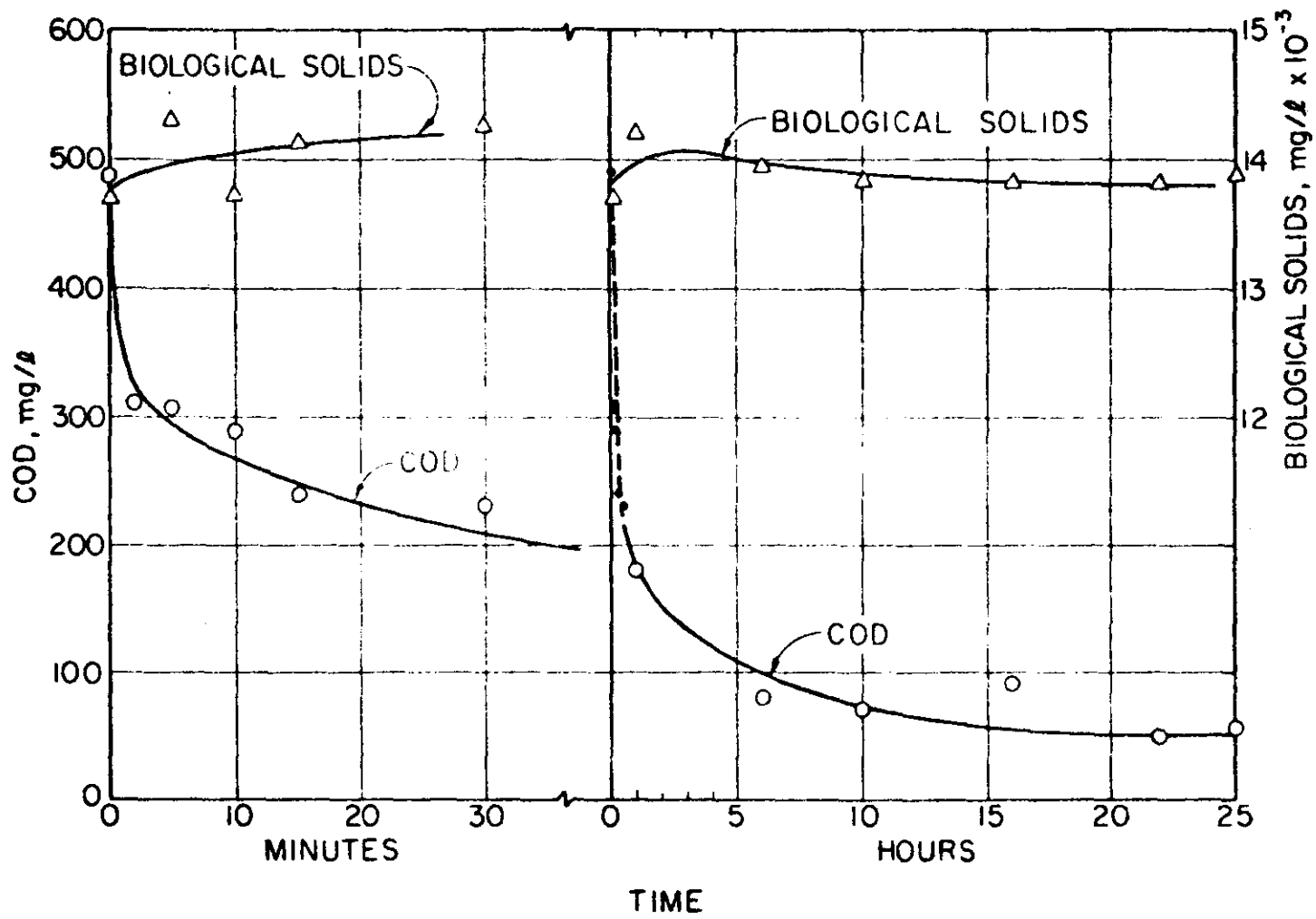


Figure 8 - Metabolism of soluble cell components (sonicate-COD) by extended aeration activated sludge.

the sludge since the relative concentrations of the two did not change appreciably. Also, the rapid removal of COD was not due to stripping of volatile organic cell constituents, since the same feed stock subjected to aeration in a system in which a low initial cell inoculum was employed showed no evidence of rapid COD removal. The graph in the right-hand portion of the figure shows the course of COD removal during the entire period of the experiment. The values which were plotted in the expanded scale (first thirty minutes) at the left are also shown (small solid circles) on this plot. It is seen that approximately ninety per cent of this organic matter was subject to biological removal (as adjudged by COD analysis), i.e., the degree of efficiency was approximately the same as that normally delivered by the system.

#### Experiments on Sludge Hydrolysate

Experiments of the type described above as well as others which are planned should do much to provide insight into the natural causation for the periodic phases of solids accumulation and subsequent de-accumulation or accelerated autodigestion. These studies may also give broader insight into understanding and possible control of natural aquatic ecosystems.

In a more immediately practical sense, it is also important to seek biological engineering practices which may allow control and enhancement of the autodigestive phenomenon. Since the results of experimentations on the extended aeration process have indicated that the concept of so-called "total oxidation" is not theoretically unsound or impossible from a microbiological standpoint, the major problem is one of controlling the autodigestive process or finding a way to retain the sludge

until a natural period of accelerated autodigestion which will reduce the sludge concentration occurs. Retaining the sludge in the system can at times be extremely difficult with present mixed liquor separation techniques, i.e., quiescent settling, since the solids concentration may undergo considerable buildup before autodigestion reduces the sludge concentration. While some innovations in settling tank design, possible chemical settling aids, etc., can help provide adequate separation of mixed liquors at high solids concentrations, there is yet another way to approach the problem, i.e., to control sludge concentration so that it is maintained in a range which does not militate against separation in the settling tank. At first consideration, such procedure would not seem consistent with the aim of retaining this organic matter in the system; however, it was noted in Figure 7 that although the detention time in the aeration chamber was sixteen hours, when the feed was added on a batch basis to ascertain the rate of COD removal, the COD was removed in approximately thirty minutes. The carbon source in the synthetic waste was glucose, which is admittedly a most readily-metabolized substrate, but the results shown in Figure 7 certainly indicate that there was excess aeration time insofar as removal of the organic loading in the incoming waste is concerned. However, the major organic loading in an extended aeration system is indeed the sludge itself, and the long aeration time is desirable because the organic materials contained in the sludge do indeed represent a complex carbon source, and the extra aeration may be needed for various autodigestive processes. It also seems reasonable to postulate that the most difficult metabolic steps in the autodigestive process are those involved in making this organic material available as a source of carbon, i.e., the necessary preparatory

steps; for example, establishment of predominance of predatory cells and induction of enzymes by one cell which could lyse various other cells, thus releasing the interior portions of the cell to the surrounding medium, the development of the necessary enzyme complement to hydrolyze various slime or capsule layers of some cells, and development of the necessary enzyme complement in order for some cells to solubilize the cell wall and membrane of other cells. The pilot plant results previously presented (3) as well as those presented herein indicate that these processes do occur naturally in the complicated ecosystem represented by the extended aeration process. However, it seemed, from a standpoint of engineering expediency and control of sludge concentration in extended aeration processes, that these preparatory metabolic steps which are believed to be the most difficult for the biological system to perform, could be overcome if a "chemical assist" to the system were provided. If these large molecules were made essentially soluble by chemical hydrolysis, the remainder of the metabolic autodigestion of the sludge might be very easily facilitated by the sludge itself. Thus, one can envision a mode of operation wherein during periods when the biological solids concentration was accumulating to such an extent that settling was impaired, some of the sludge could be withdrawn from the underflow of the settling tank, hydrolyzed, and the neutralized hydrolysate, i.e., chemically prepared cell material, could then be recycled or bled back to the aeration chamber to be used as carbon source by the extended aeration sludge, thus hastening the process of its eventual total oxidation, i.e., conversion of the organic carbon to  $\text{CO}_2$ .

Thus far this concept has been tested by performing experiments in which cells grown up from an initial seed taken from the extended

aeration pilot plant have been hydrolyzed under conditions which solubilize the sludge, and this material has been fed as a source of carbon to the extended aeration plant under batch shockloading conditions similar to those in the feeding studies shown in Figures 7 and 8. The results of such an experiment are shown in Figure 9. If one compares the course of COD removal for this hydrolyzed cell material, which represents an extremely complex carbon source, with the results shown in Figure 7 for which the feed consisted of the synthetic waste entering the extended aeration plant, it can be seen that the sludge had no difficulty whatsoever in metabolizing the hydrolyzed sludge. For the experiment shown in Figure 9, the sludge was hydrolyzed under acid conditions, pH = 1, in a laboratory autoclave (five hours at 15 psi, 121°C), and after neutralization to pH 7, it was added as a slug dose to the pilot plant aeration tank. In the work thus far, materials balances have been made on cell COD before and after acid hydrolysis at the conditions described above for four different sludges. For two of them, the COD of the cells before hydrolysis was the same as the COD of the hydrolyzed sludge. In one, 23 per cent of the COD was removed by the hydrolysis procedure, and in another, 16 per cent was removed. It is interesting to note that under the hydrolysis conditions employed, all cell components are generally solubilized. After hydrolysis, there is usually a small amount of a fluffy precipitate of unknown composition but which is apparently inorganic in nature since the COD of the neutralized hydrolysate before and after passage through a Millipore filter (0.45  $\mu$ ), which retained the small amount of precipitate formed, was the same. The conditions of hydrolysis employed do not break the cell protein down to amino acids. Approximately 50-60

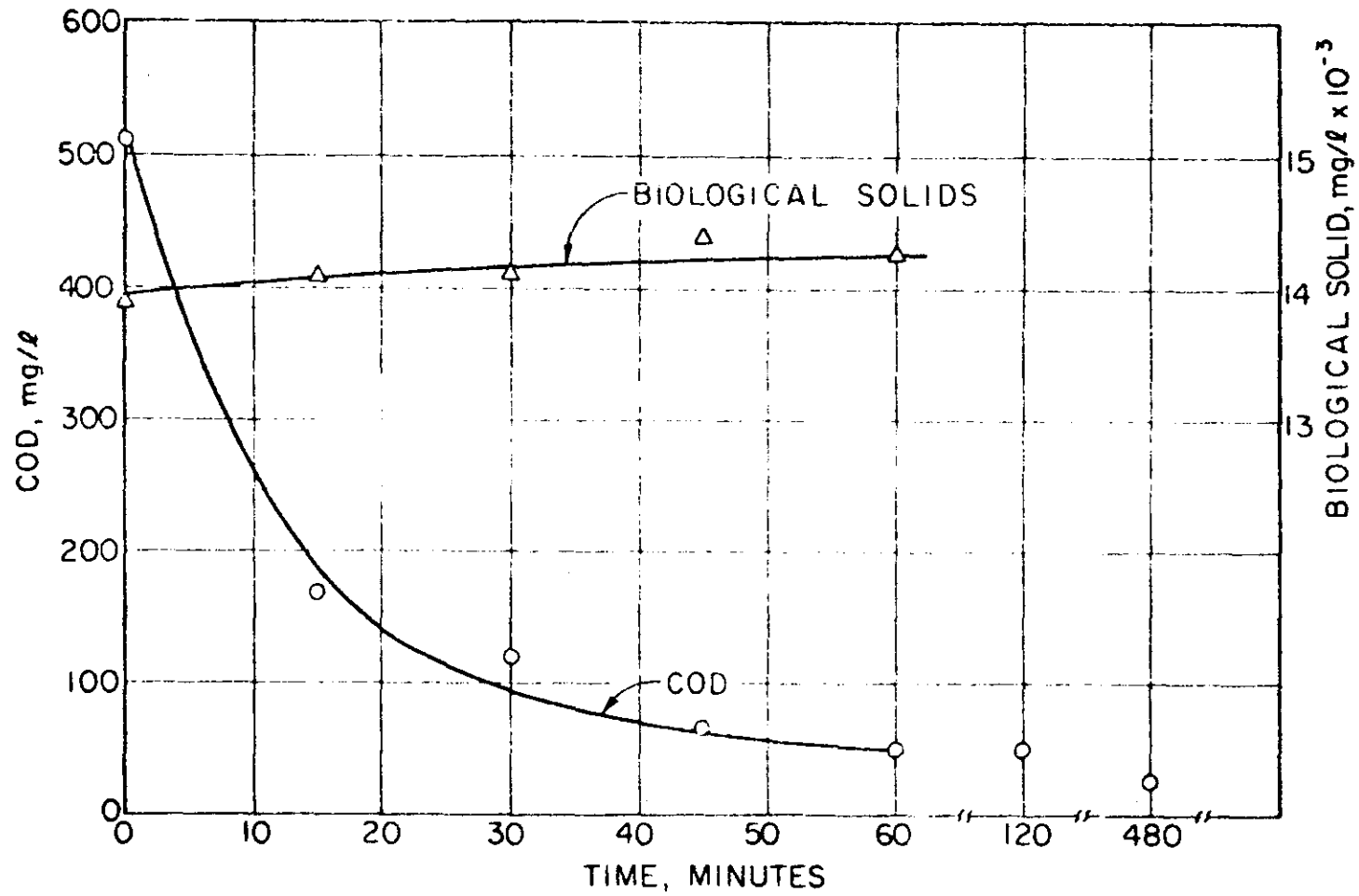


Figure 9 - Metabolism of hydrolyzed biological sludge by extended aeration activated sludge.

per cent of the hydrolysate COD equivalent as protein (nitrogen test), and  
10 per cent as volatile solids (dry weight) (nitrogen test).

### Conclusions and Proposed Process Modification

From the results which have been presented, it is possible to suggest the rather innovative process modification or mode of operation shown in the flow diagram in Figure 10. A typical extended aeration process unit consisting of aeration chamber and settling tank with internal solids recycle to the aeration tank is depicted in the top portion of the figure. The lower portion represents the proposed hydrolysis plant wherein sludge drawn from the settling tank underflow is acidified and subjected to hydrolysis. The hydrolysate, rich in COD, may then be neutralized and bled back to the aeration chamber. With such a process it would not be necessary to determine ways and means to retain sludge until the ecology of the system brought about conditions of biological hydrolysis. Such a system as shown in the diagram utilizes both chemical and biological methods of sludge disposal to best advantage. It does chemically a function which is biologically difficult, and does biologically a function which is difficult and costly to do chemically.

The total system, i.e., extended aeration plant plus the hydrolysis plant to aid the autodigestive process, can provide for treatment of the organic waste and for sludge disposal. The results to date indicate that such a system could be successfully operated and that the process being proposed could be a very useful one which could do much to alleviate the sludge disposal problem and permit the wider use of extended aeration for high strength industrial wastes. The hydrolysis portion of the plant could be operated intermittently only when solids

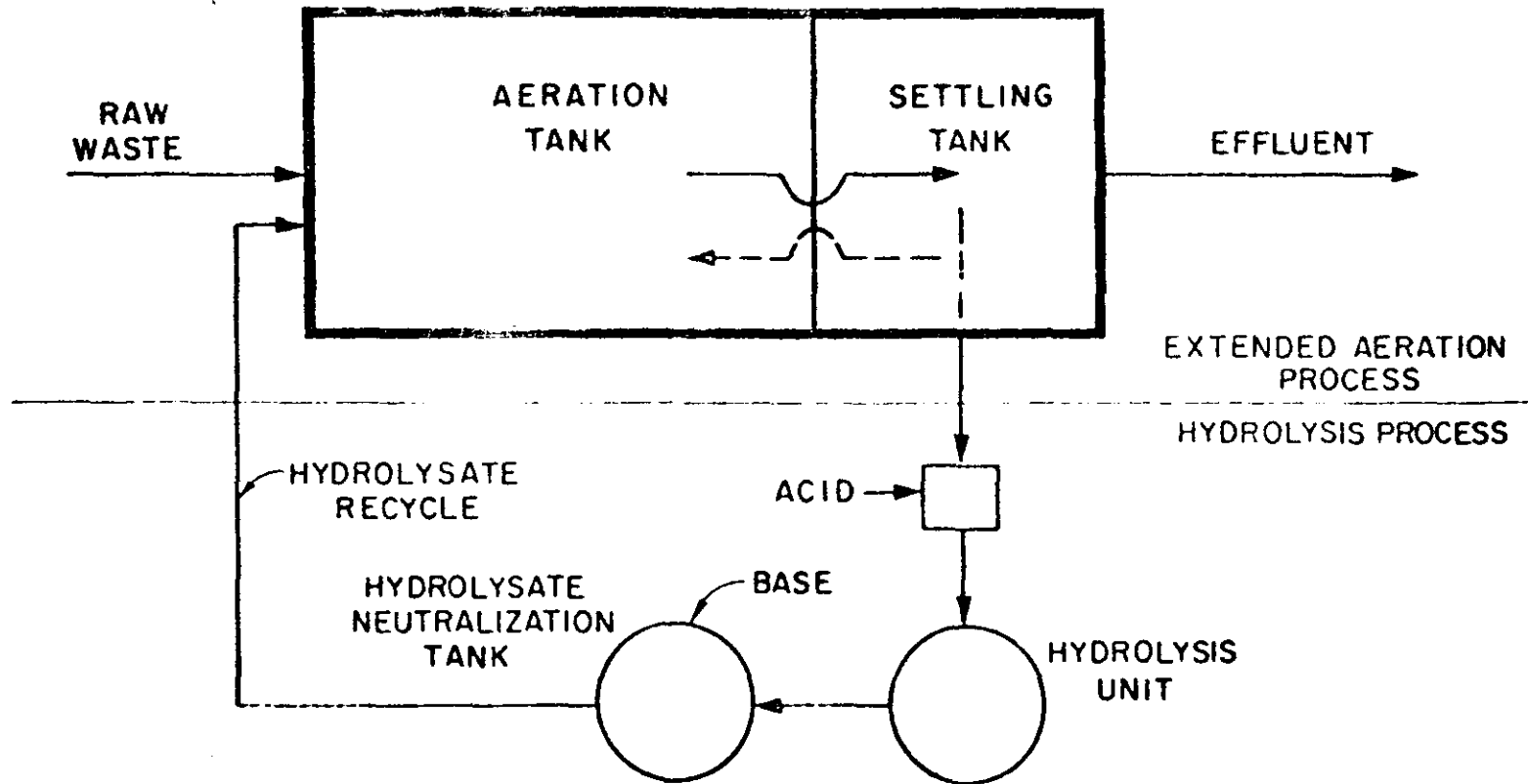


Figure 10 - Proposed extended aeration activated sludge process incorporating chemical hydrolysis for control of sludge concentration.



in the settling tank were building up to such an extent that settling was endangered. Employing acid hydrolysis, the pH of the hydrolysate would be low enough so that this material could be stored for bleeding back into the aeration chamber without serious danger of its decomposition in the holding tank. Alternatively, the hydrolysis unit could be operated often enough to permit continual recycle of hydrolysate. If the particular industrial waste being treated was one which was originally deficient in nitrogen, this portion of the sludge disposal operation would also permit considerable re-use of the nitrogen supplement, thus cutting nitrogen costs; phosphorus supplementation might similarly be re-used. On the other hand, if the waste contained an overabundance of nitrogen, that portion of the nitrogen retained in the sludge might be removed by employing more severe conditions, e.g., partial chemical digestion, which could convert the amino groups of the proteins to ammonium ion. Adjustment of the pH to the alkaline range would permit the nitrogen to be stripped from the hydrolysate. By the same token, if the waste contained an excess of phosphorus, that portion of it taken up in the sludge could be removed by alkaline precipitation prior to neutralization and channelling the hydrolysate to the aeration tank for removal of the soluble organic matter of the sludge. Such operations for nitrogen and phosphorus removal of these constituents taken up by the sludge could require only a minor modification to the proposed flow sheet shown in Figure 10.

The present research to elucidate the natural ecological mechanisms for the biological or wet combustion of the sludge which occurs during the operation of the extended aeration process, and engineering studies on combined chemical hydrolysis and biological oxidation of the sludge

concurrent with removal of the organic loading in an inflowing waste, are continuing. However, it seems reasonable to conclude that the results to date indicate that the process described in Figure 10 and its various modifications discussed in the text, offer distinct advantages over present practices, and that the process holds considerable promise for future development and wider application of the extended aeration process. The concept is a simple one and it is believed to be a workable one. It circumvents present lack of knowledge regarding natural ecological hydrolysis of cell components by performing this particular operation chemically, and permits biological (wet) combustion of the chemically hydrolyzed sludge concurrently with removal of the organic carbon sources in the incoming waste stream. Furthermore, in the case of a nutrient-deficient industrial waste, it permits useful recycling of nutrients, thus cutting operational cost while aiding in the "total" oxidation of the sludge.

#### ACKNOWLEDGEMENTS

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## LITERATURE CITED

1. Thabaraj, G. J., and Gaudy, A. F. Jr., "Effect of Initial Biological Solids Concentration and Nitrogen Supply on Metabolic Patterns During Substrate Removal and Endogenous Metabolism." Presented at the 42nd Annual Conference, Water Pollution Control Federation, Dallas, Texas, October 5-10 (1969).
2. Ramanathan, M., Gaudy, A. F. Jr., and Ragthaidee, W., "Responses of Extended Aeration Activated Sludge to Quantitative Shock Loads." Proceedings, 19th Oklahoma Industrial Waste Conference, Oklahoma State University, Stillwater, Oklahoma (1968).
3. Gaudy, A. F. Jr., Ramanathan, M., Yang, P. Y., and DeGeare, T. V., "Studies on the Operational Stability of the Extended Aeration Process." Journal Water Pollution Control Federation, 42, 165-179 (1970).
4. Gaudy, A. F. Jr., and Engelbrecht, R. S., "Basic Biochemical Considerations During Metabolism in Growing versus Respiring Systems." Advances in Biological Waste Treatment, II, Pergamon Press, New York (1963).
5. Rao, B. S., and Gaudy, A. F. Jr., "Effect of Sludge Concentration on Various Aspects of Biological Activity in Activated Sludge." Journal Water Pollution Control Federation, 38, 794-811 (1966).
6. Krishnan, P., and Gaudy, A. F. Jr., "Substrate Utilization at High Biological Solids Concentrations." Journal Water Pollution Control Federation, Research Supplement, 40, R54-66 (1968).

Yang, P. Y., "Studies on the Operational Stability of the Extended  
Aeration Process." Master's thesis, Oklahoma State University,  
Stillwater, Oklahoma (1969).

APPENDIX 4

**STUDIES ON THE OPERATIONAL STABILITY OF THE  
EXTENDED AERATION PROCESS**

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Title of Study: STUDIES ON THE OPERATIONAL STABILITY OF THE EXTENDED  
AERATION PROCESS

Pages in Study: 86

Candidate for the Degree of Master of Science

Major Field: Bioenvironmental Engineering

Scope and Method of Study: The performance of an extended aeration process (both batch and continuous feeding) operated without sludge wastage, was investigated. Glucose was used as the sole carbon source in the synthetic waste. The parameters measured during this study were COD, biological solids concentration, sludge composition, and endogenous  $O_2$  uptake.

Findings and Conclusions: The results of the study indicated that an extended aeration activated sludge process can provide high purification efficiency without a steady increase in biological solids concentration. Periodic decreases in biological solids concentration were observed. During such periods, purification efficiency remained high. It was also found that the substrate utilization capability of the sludge did not deteriorate with sludge age, although the endogenous oxygen uptake of the aged sludge did attain very low levels.

ADVISER'S APPROVAL \_\_\_\_\_

STUDIES ON THE OPERATIONAL STABILITY OF THE  
EXTENDED AERATION PROCESS

Thesis Approved:

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Thesis Adviser

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Dean of the Graduate College

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## CHAPTER I

### INTRODUCTION

The activated sludge process was developed in 1913 in England, and remained essentially unchanged during the next thirty years. During the past twenty-five years, many modifications of the process have been developed. The extended aeration process is one of the more recent modifications. It has been claimed to be simple in operation, low in cost, and stable to environmental changes. The increase in the number of extended aeration plants used in the United States is impressive. Because of the ever-increasing demand for water pollution control, many small communities and commercial establishments have found that the extended aeration activated sludge system is the desirable one for treatment of their organic wastes. However, some prominent researchers have concluded that the operation of an extended aeration system (without wasting sludge) is theoretically impossible. Regardless of this objection, the process has been used to an increasing extent.

The study herein reported was designed to determine how long a system in which all biological solids were returned to the aerator could be operated before the "inactive" fraction built up to such an extent that the biochemical efficiency was destroyed or greatly retarded. The work of previous investigators indicated that such systems should in time undergo biochemical failure. However, there

were no data which provided unequivocal proof that failure would actually occur, nor were there any data indicating how long it would take for such a system to lose its substrate removal capability.



## CHAPTER II

### LITERATURE REVIEW

Since the end of World War II, many modifications have been proposed to the activated sludge process. The process has been improved through years of research. Its use will be accelerated because of new pollution control laws. The extended aeration process (or total oxidation process) is one of the modifications of the activated sludge treatment process which is extremely attractive from an economic point of view. This kind of treatment process, if it could be successfully operated, could attain complete mineralization of organic wastes and obviate the need for separate sludge digestion facilities. It would be particularly useful for treatment of soluble industrial wastes.

Although the extended aeration process would seem to have advantages, it has been the subject of much controversy during the past years. The major concern is over the accumulation of inert metabolic products and inactive biological solids. Regardless of these concerns, the increase in the number of extended aeration plants used in the United States is impressive; from three in 1950, to 1,224 in 1960, and to more than 2,600 in 1962. Were it not for the published conclusions of various research engineers warning against use of the process on the basis of its theoretical unsoundness, it might enjoy even wider use today.

The extended aeration process is designed to provide a detention

time in an aeration chamber of 16 to 24 hours, and a detention time in a final settling tank of four to eight hours. Long aeration periods are provided so that the organisms in the extended aeration process have sufficient time to assimilate the exogenous substrate into the cell components and then to oxidize these components. Many microorganisms can survive for a "considerable period" in the absence of nutrients (1). Reserve materials within the cell can be oxidized and provide the energy to make the cell survive for a "considerable period" of time. However, the extended aeration system is one in which low organic loadings are employed. The organism in this system must have the competence to take the food (organic wastes), but most of the organisms would be expected to be in a state of starvation because of the low substrate loading in the extended aeration system. Under such conditions, some cells might not survive very long but might die, thus releasing food materials for other cells. If extended aeration is to be a workable process, such autodigestion of the sludge must go on concurrently with metabolism of the carbon source in the incoming waste water.

Porges, et al. (2) reported that an equilibrium between the net autoxidation and net synthesis of new biological sludge had been established in the treatment of dairy waste. A portion of the soluble organic waste (skim milk) was converted to cell material and then autoxidized in the later phases of aeration. Porges, et al. concluded that the waste could be treated without any accumulation of sludge. This report stimulated a considerable amount of research on the process by workers in the water pollution control field.

Most of the findings led to the general conclusion that from a

theoretical standpoint total oxidation is not possible, since a portion of the soluble organic substrate is channelled into synthesis of so-called "permanently inert materials" (3)(4)(5)(6). Thus it was concluded that if no sludge was wasted, the extended aeration process could not perform successfully. It must be emphasized that in all of the studies (3)(4)(5)(6) from which the above conclusion was made, soluble organic substrates were used. Thus the inert materials referred to are inert biological materials produced by the microorganisms responsible for removal of the exogenous substrate.

Based on the pure culture study of Dawes and Ribbons (1), the endogenous substrates thus far recognized in bacteria include glycogen, lipid, poly- $\beta$ -hydroxybutyric acid, protein, and RNA; inorganic polyphosphate (volutin) can serve as an endogenous store of phosphate. Most of these endogenous substrates can be utilized by various microorganisms. However, in addition to endogenous metabolism of individual species it must be remembered that the activated sludge system is a mixed population, i.e., a rather complicated ecosystem, and one species may cause death of another and may use the structural components of the other species as carbon source.

The report of McCarty and Broderick (7) is particularly interesting. These workers suggested that solids accumulation must be considered in the design of extended aeration systems. They felt that if no facilities for disposal of excess sludge were provided, the system will accumulate solids and discharge the excess suspended solids in the effluent. Thus, the purification efficiency of the extended aeration system could be expected to decrease as a result of the continual increase in solids. The sludge accumulated in the unit will include

the synthesized biological solids and the biologically nondegradable suspended solids which were originally present in the influent waste (e.g., small grit particles or certain biologically resistant organics such as lignin and cellulose). Therefore they suggested that industrial, soluble organic wastes and municipal wastes must be considered separately when designing extended aeration systems. In addition, they suggested that the efficiency of operation of extended aeration processes was closely related to the effectiveness of the settling tank in retaining the suspended solids. They also noted that nitrification will cause false values for BOD removal efficiency, the dropping of pH, and rising sludge in the settling tank. Nitrification is the result of excess aeration (long detention time) and low organic loading, which favor the growth of nitrifying bacteria. In wastes containing an abundance of nitrogen it would seem that nitrification could present a problem. They explained that the lowering of pH was caused by the formation of nitric acid. If pH dropped in the system, it would appear that filamentous organisms might come into predominance and a bulking sludge could develop as well as, or instead of, a rising sludge.

In a more recent study, Westrick, et al. (8) observed the operation of two extended aeration plants in Ohio. Both of these plants had no equipment to control the wasting sludge, i.e., all of the settled sludge was returned except the solids which escaped in the effluent. From their results they concluded that when the influent loading value was close to the design value and no facilities for the removal of sludge or some efficient means of solids separation were provided, the extended aeration plants could not produce the desired purification efficiency. Thus, their observation is much the same as that of McCarty and

Broderson (7), i.e., that an extended aeration plant must be designed with an oversized settling chamber to avoid the escape of solids in the effluent.

From the above review it can be seen that the results of other workers indicate that biologically inert organic solids will gradually accumulate, and that biological solids will escape in the settling chamber effluent. Both of these events can cause drastic reduction in purification efficiency. The buildup of biologically inert material is deemed to be the foremost consideration in assessing future possibilities for the process, since even if ways and means could be found to avoid loss of solids in the effluent, thus assuring retention of all biological solids, the increasing inert fraction could cause the system to fail biochemically. The complicated nature of the ecosystem which exists in an activated sludge, and the possibilities for predatory activity made it difficult to accept the concept of continual buildup of inert organic matter, and encouraged the initiation of the present investigational effort. Studies on the extended aeration process have been under way in the bioengineering laboratories of Oklahoma State University for nearly two years. Recently, studies on the ability of the process to accommodate quantitative shock loads have been reported (9). Also, some of the research included in this thesis has been recently reported (10).

## CHAPTER III

### MATERIALS AND METHODS

#### 1. Experimental Apparatus

The bench scale extended aeration pilot plant employed in these studies was essentially the same as the one used in the shock loading studies reported previously (9). A flow diagram showing the aerator and settling chambers, the supernatant holding tank, and the Sharples centrifuge through which the holding tank liquor was processed is shown in Figure 1. The total volume of the system is 9.4 liters (6.2 liters, aeration chamber, and 3.2 liters, settling chamber). A sliding baffle was used to separate the aeration and settling chambers. Compressed air provided not only mixing and oxygen supply to the biological solids, but also provided suction to recycle settled solids from the settling chamber. Airflow rate was maintained at 2000 cc/min/l. This airflow rate provided for a dissolved oxygen concentration in the aeration chamber, which was always above 6 mg/l, and a DO concentration in the settling chamber which averaged slightly above 2 mg/l. Temperature was maintained at  $23 \pm 2^{\circ}\text{C}$ .

Figure 2 shows the apparatus used in studies to determine the growth and substrate removal characteristics of cells from the extended aeration unit. For these studies a small inoculum of cells from the extended aeration unit was placed in the growth apparatus along with

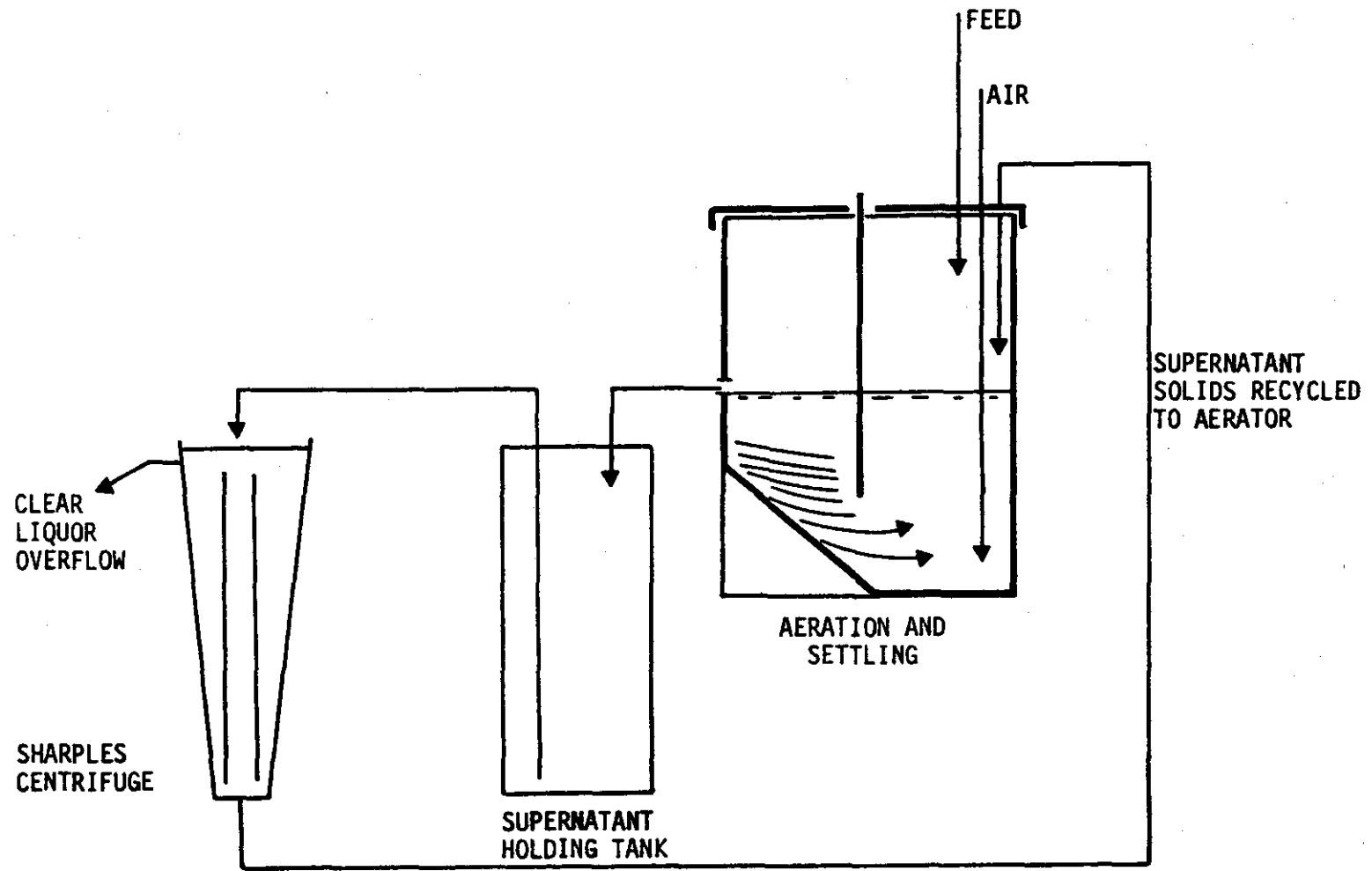


Figure 1 - Continuous flow extended aeration pilot plant.

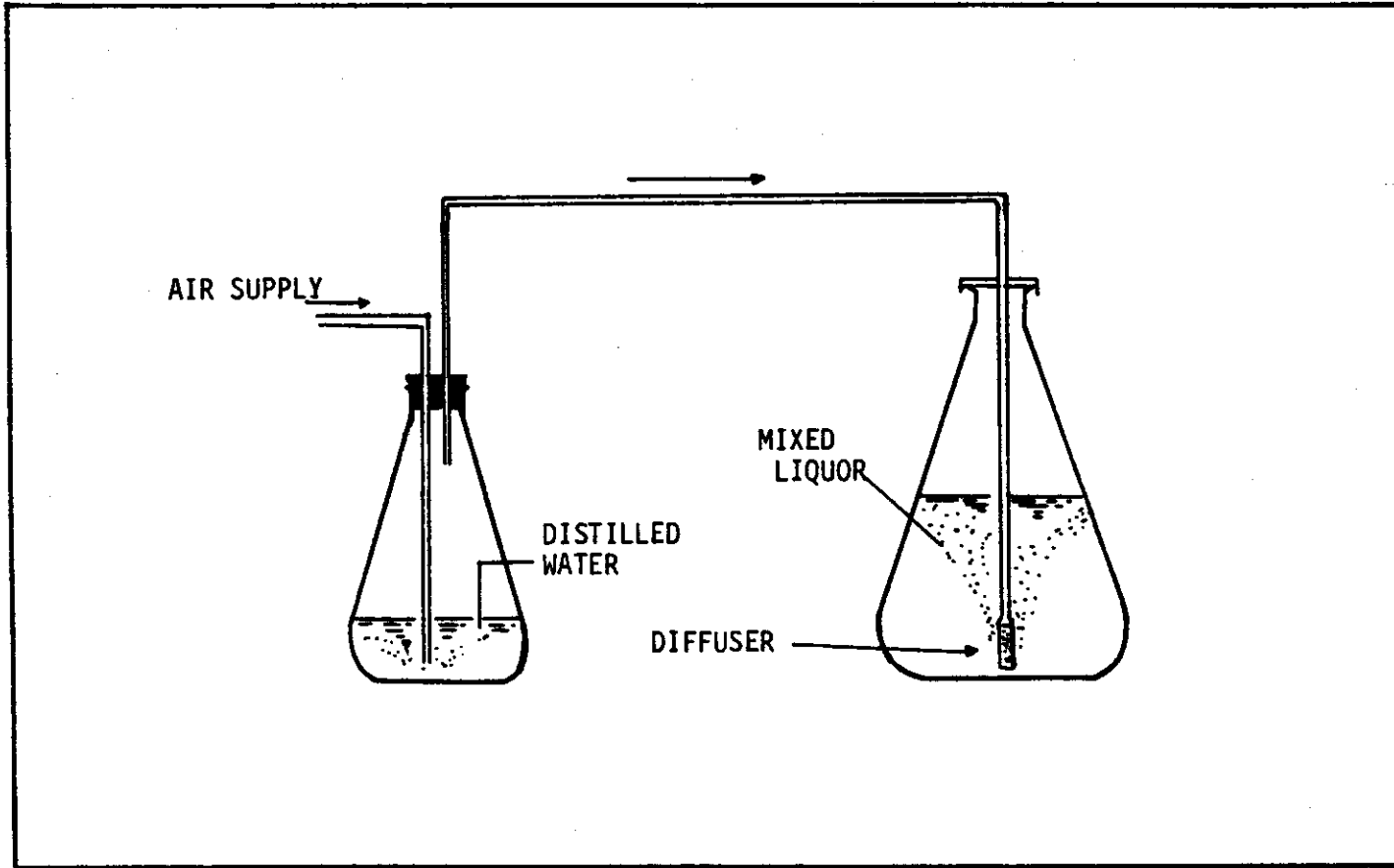


Figure 2 - Experimental setup for substrate utilization rate with low initial solids concentration.



fresh medium, and samples were withdrawn periodically for analysis.

## 2. Composition of the Synthetic Waste

Table I shows the composition of the synthetic waste during the continuous feeding operation. During periods of batch operation, the concentration of each constituent was increased 9.4-fold. The organic loading was approximately 50 lb COD/1000 cu ft aeration capacity (32 lb BOD<sub>5</sub>/1000 cu ft) during continuous flow operations, and 32 lb COD/1000 cu ft (22 lb BOD<sub>5</sub>/1000 cu ft) on the basis of total volume of the system (or aeration volume) during batch operation.

The composition of the synthetic waste used in the separate batch studies to assess rate of growth and substrate removal was the same as that shown in Table I.

TABLE I

### COMPOSITION OF FEED FOR 500 mg/l GLUCOSE AS SUBSTRATE

Glucose	500	mg/l
(NH <sub>4</sub> ) <sub>2</sub> SO <sub>4</sub>	250	mg/l
MgSO <sub>4</sub> ·7H <sub>2</sub> O	50	mg/l
FeCl <sub>3</sub>	0.25	mg/l
CaCl <sub>2</sub>	3.75	mg/l
MnSO <sub>4</sub> ·H <sub>2</sub> O	5	mg/l
Phosphate Buffer, 1.0 M (KH <sub>2</sub> PO <sub>4</sub> + K <sub>2</sub> HPO <sub>4</sub> )	5	ml/l
Tap Water	50	ml/l

### 3. Procedure

During the operation under continuous flow conditions, the feed rate was set to provide an overall detention time of 24 hours (approximately 16 hours aeration and eight hours settling). During periods of batch operation, the baffle separating the two chambers was removed and the unit was fed once daily. During this period a 23-hour aeration period was employed. The sludge was permitted to settle for one hour after the daily sample (15 ml) was taken. After the one-hour settling period, one liter of supernatant was removed, centrifuged, and the biological solids were returned with one liter of feed solution to the reactor, thus the unit was again brought back to the operating level (9.4 liters). During continuous flow operation, the sterile feed solution was channelled to the aeration chamber through a dual unit positive displacement pump arrangement (minipump Model MM2-B-96R). The feed line was also cleaned (sterilized) regularly. There were never any indications of contamination of the feed solution. The effluent collected in the holding tank was periodically (once or twice daily) passed through the centrifuge (Sharples Superspeed), and any solids which had been carried over from the settling chamber were scraped from the bowl and returned to the aeration chamber. It is emphasized that no biological solids were either inadvertently or intentionally lost from the system throughout this study. The only solids "lost" from the system were those taken for samples to assess operational behavior.

Daily (or nearly so) 15 ml samples of mixed liquor were removed for analysis. At less frequent intervals (approximately twice per month) a total of 20 ml samples of mixed liquor were removed for measurement of endogenous  $O_2$  uptake, sludge composition (carbohydrate

and protein) and periodic examination of substrate removal rate. The samples for the measurement of biological solids concentration were taken in such a way that the data represent those in the total system, not in the aeration chamber alone. The procedure for sampling was as follows: Just prior to taking a sample, the settling chamber outlet was stoppered, and the feed was momentarily shut off. The dividing baffle was then lifted and the contents of the settling chamber were allowed to mix completely. Also while the solids were mixing, the solids in the effluent which had been passed through the centrifuge were returned to the system. Then the 15 ml sample was pipetted from the unit and the dividing baffle was again inserted into place. The settling chamber effluent was reopened, and the feed restarted. This procedure allowed a direct assessment of the course changes in solids concentration in the system.

#### 4. Analytical Methods

Biological solids concentration was determined by the membrane filter technique (Millipore Filter Co., Bedford, Mass., HA 0.45 $\mu$ ) as outlined in Standard Methods (11). Chemical oxygen demand (COD) was also run on the filtrate (11). At periodical intervals, COD's were also run on the effluent from the settling chamber. Protein and carbohydrate contents of the sludge were determined, respectively, by the biuret and anthrone (12) tests. At various times the  $\text{PO}_4^{\equiv}$  (13),  $\text{NH}_3\text{-N}$ ,  $\text{NO}_2\text{-N}$ , and  $\text{NO}_3\text{-N}$  (11) were determined on the filtrated effluent. In some batch experiments, the anthrone test (in addition to COD's) was run on the membrane filtrate. Endogenous  $\text{O}_2$  uptake of the sludge was determined for a cell suspension obtained from a 10-15 ml sample of reactor mixed liquor (washed twice in 0.1 M phosphate buffer solution). The

rate of the endogenous  $O_2$  uptake was measured in a Warburg respirometer using a reaction suspension of 40 ml, and 1.5 ml 20 per cent KOH in the center well. The system was maintained at 25°C and 90 osc/min. Periodically throughout the experimentation, dissolved oxygen in the aeration chamber mixed liquor and in the settling chamber effluent was measured by a galvanic cell oxygen analyzer, in accordance with the procedures given in the operating instructions supplied with the instrument (14).

## CHAPTER IV

### RESULTS

The effluent, from the primary clarifier of the municipal sewage treatment plant at Stillwater, Oklahoma, was used as initial seed for the development of the extended aeration sludge. After a few days of "batch" growth to allow acclimation and solids accumulation, the unit was put into operation on March 30, 1967, under continuous flow feeding with 1000 mg/l glucose as the substrate, and all biological solids contained in the effluent were returned to the aerator except a small portion (15 ml daily) taken for analysis. On October 12, 1967 (196 days after starting the unit), approximately half of the system was transferred to a second unit of the same type. Both systems were diluted to 9.4 liters with tap water. Since the biological solids concentration was now approximately halved, the feed concentration was reduced proportionately (500 mg/l glucose). One system was used for studies on the long-term behavior of the extended aeration process; the other was used for studies on the shock load behavior of the extended aeration process. Some of the shock load studies (quantitative shock) have recently been reported (9). The work reported herein pertains to the long-term behavior of the other unit which was operated under non-shock loading conditions. Portions of this work have also been reported at a recent industrial wastes conference (10). The early

phases of the experimental effort were conducted by Mr. T. V. DeGeare (spring and summer, 1967).

### 1. Daily Performance Characteristics of the Extended Aeration Pilot Plant

From the initial time forward the performance data are shown in Figures 3 to 17. A considerable number of parameters were assessed and are plotted on these graphs. Also, the days when the various batch studies were made are indicated on the graphs. The following identification key applies to Figures 3 to 17:

- = protein content of the biological solids
- = supernatant COD (unfiltered)
- △ = carbohydrate content of the biological solids
- ▲ = supernatant biological solids
- △ = biological solids in the system
- = filtrate COD
- ⊙ = sample taken for endogenous O<sub>2</sub> uptake (Warburg)
- ▲ = sample taken for "low initial solids" batch experiments
- = "high solids" batch experiment

At the time of initiating the experimental work, the feed consisted of 1000 mg/l glucose, and salt concentrations were proportionally increased (i.e., double those shown in Table I). During the first thirty days of operation, the biological solids concentration rose gradually from 2000 mg/l to slightly over 6000 mg/l, and then decreased to 4700 mg/l by the thirty-ninth day of operation. It must be emphasized that the decrease in solids concentration was due, not to the loss of solids in the effluent or purposely wasting sludge, since all

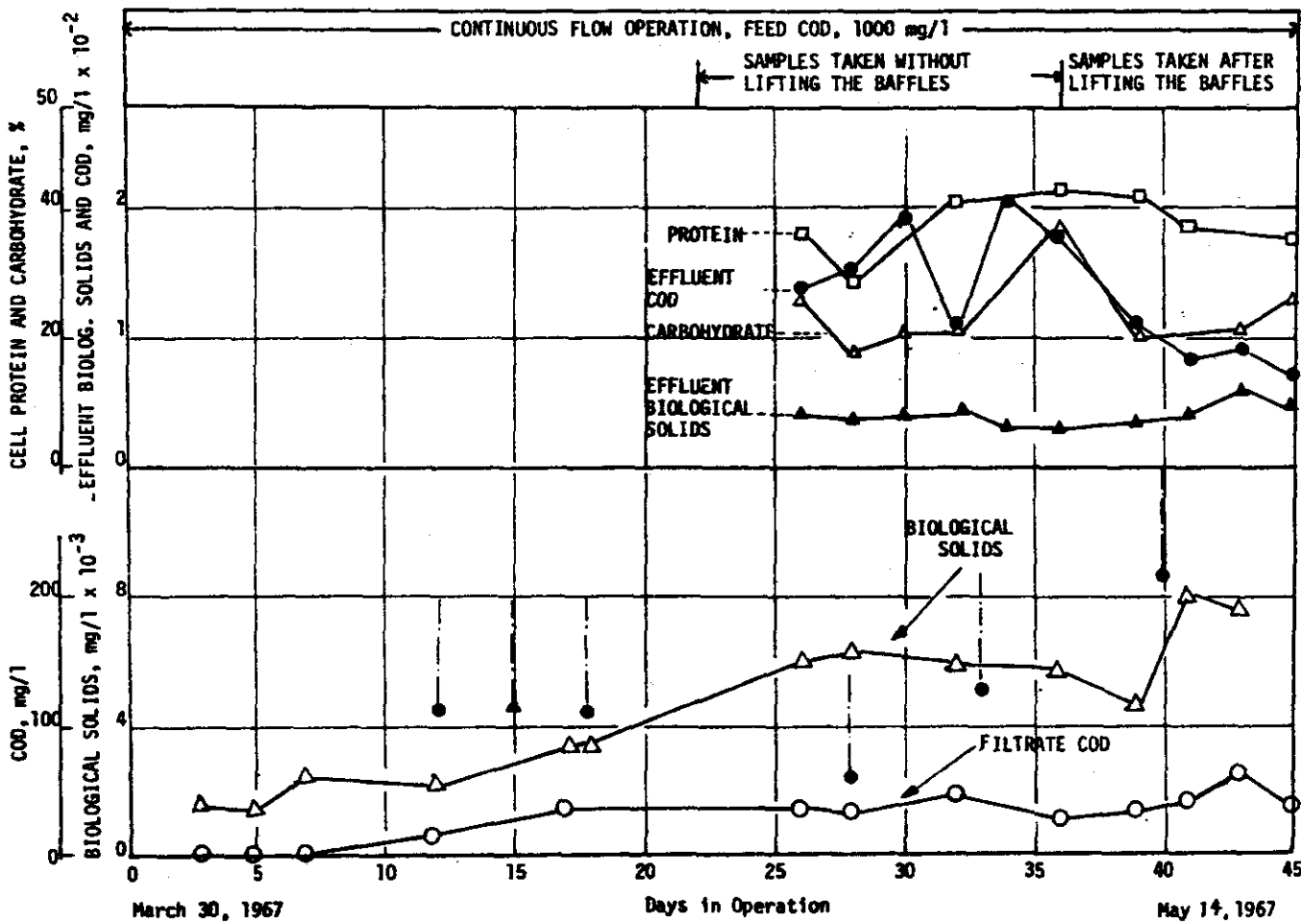


Figure 3 - Operational characteristics, March 30, 1967, to May 14, 1967.

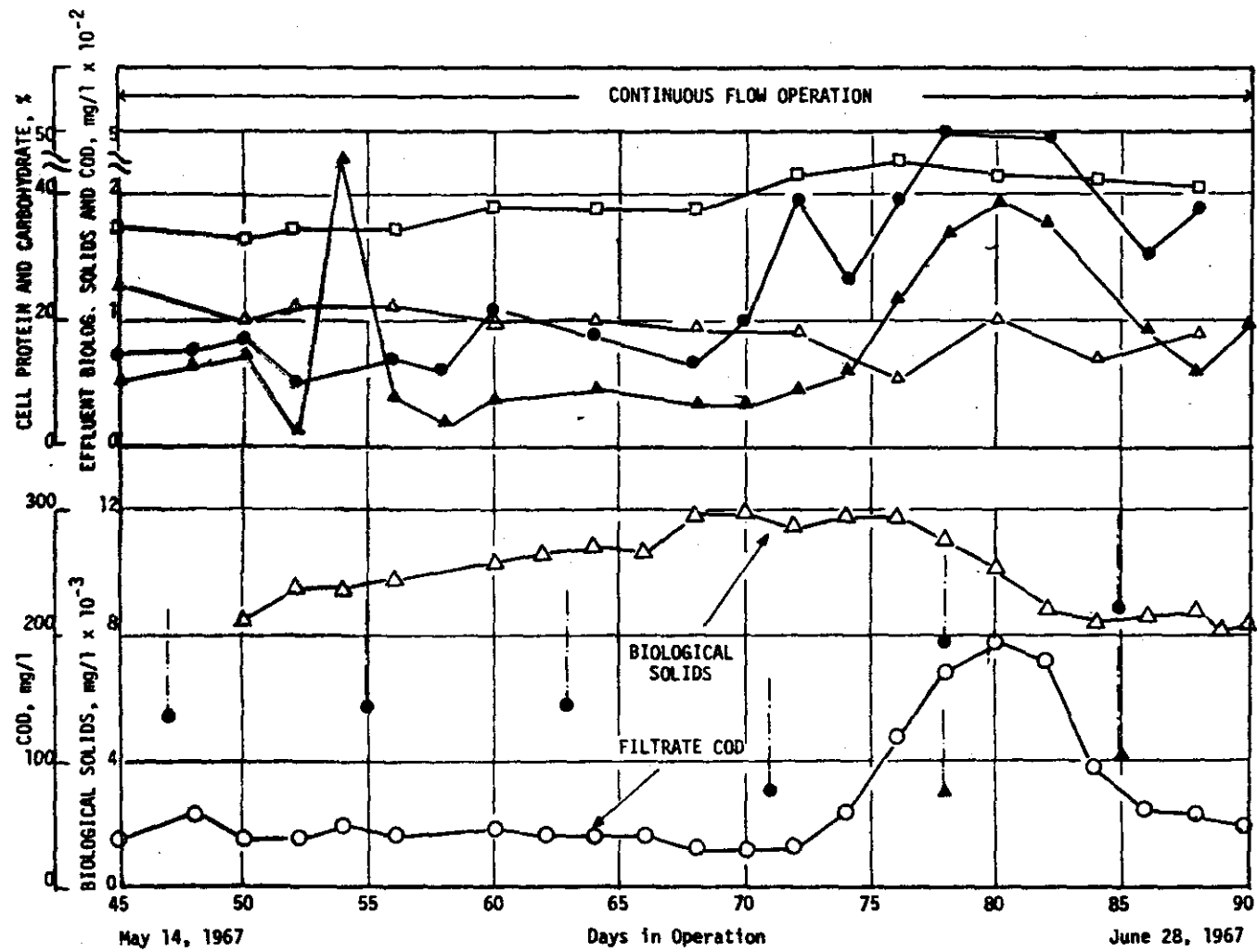


Figure 4 - Operational characteristics, May 14, 1967, to June 28, 1967.



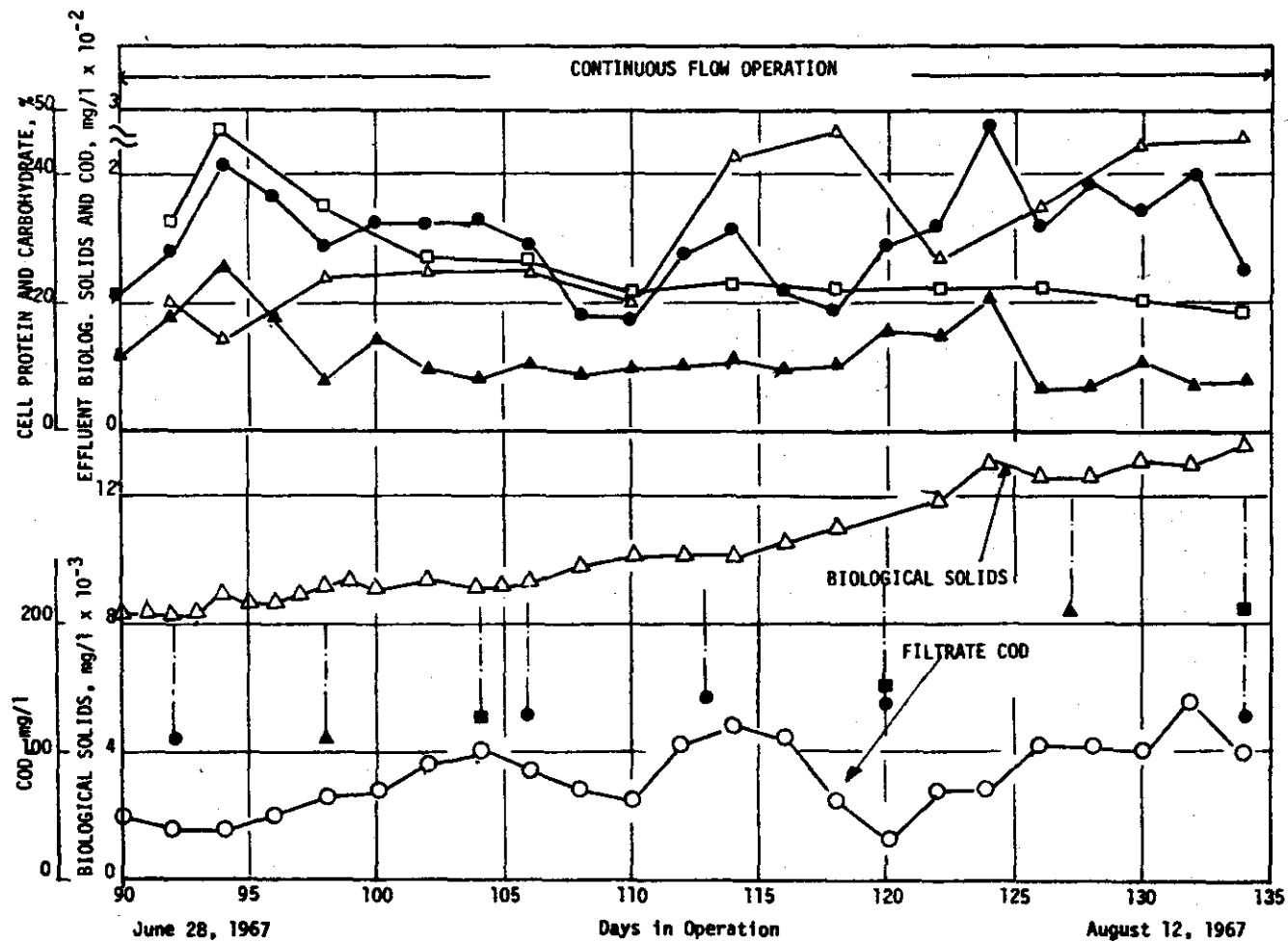


Figure 5 - Operational characteristics, June 28, 1967, to August 12, 1967.

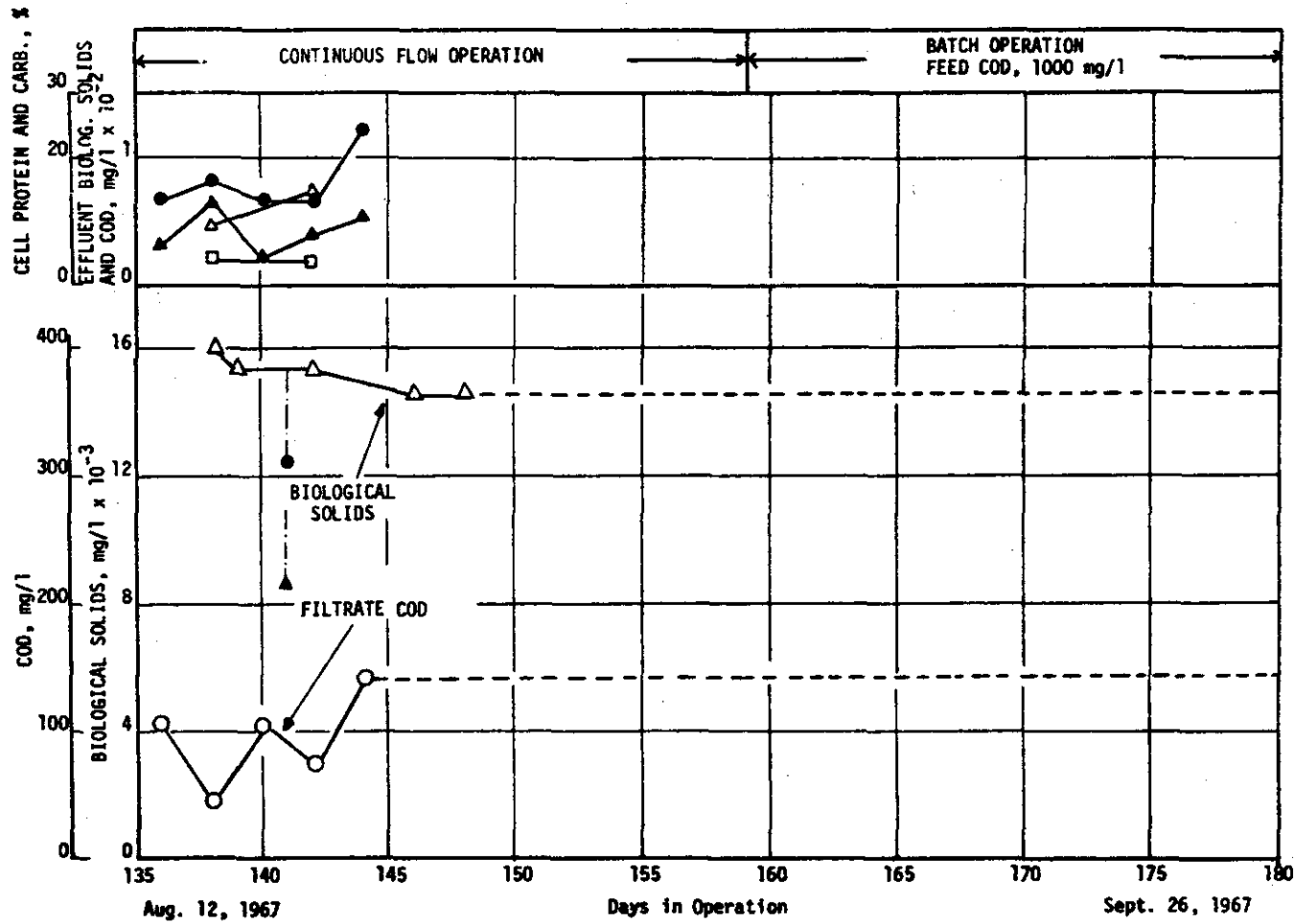
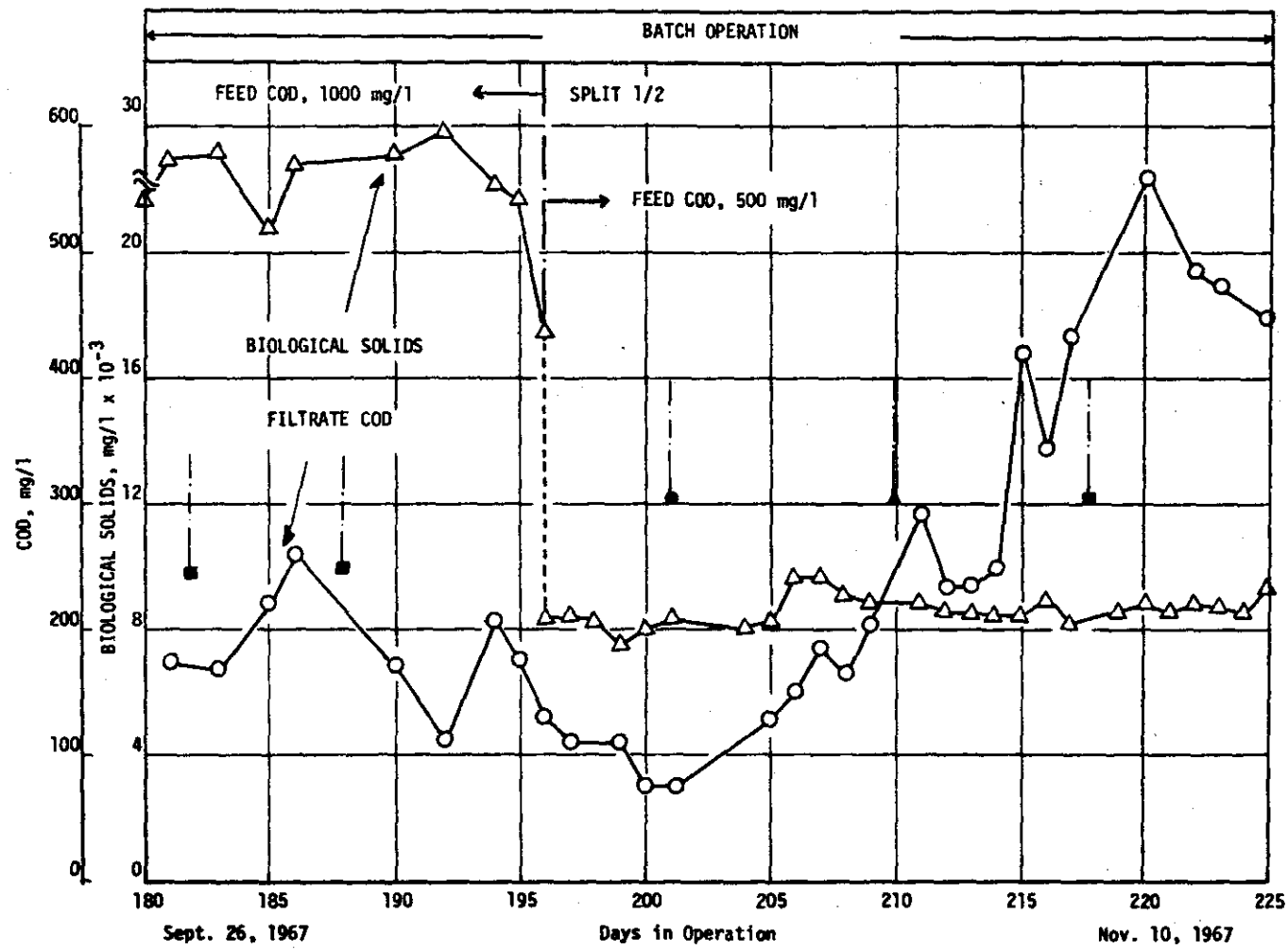


Figure 6 - Operational characteristics, August 12, 1967, to September 26, 1967.



Operational characteristics, September 26, 1967, to November 10, 1967.

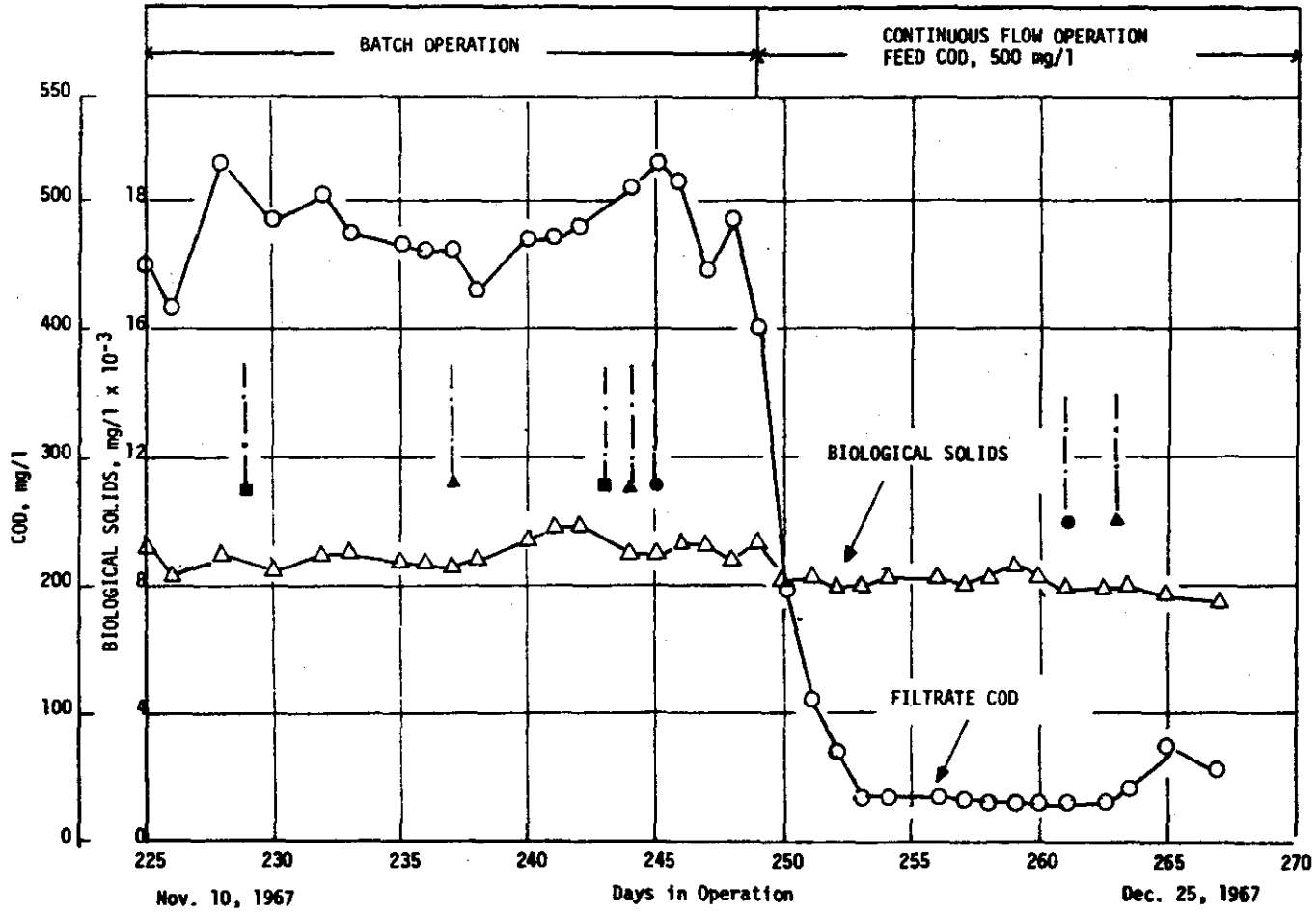


Figure 8 - Operational characteristics, November 10, 1967, to December 25, 1967.

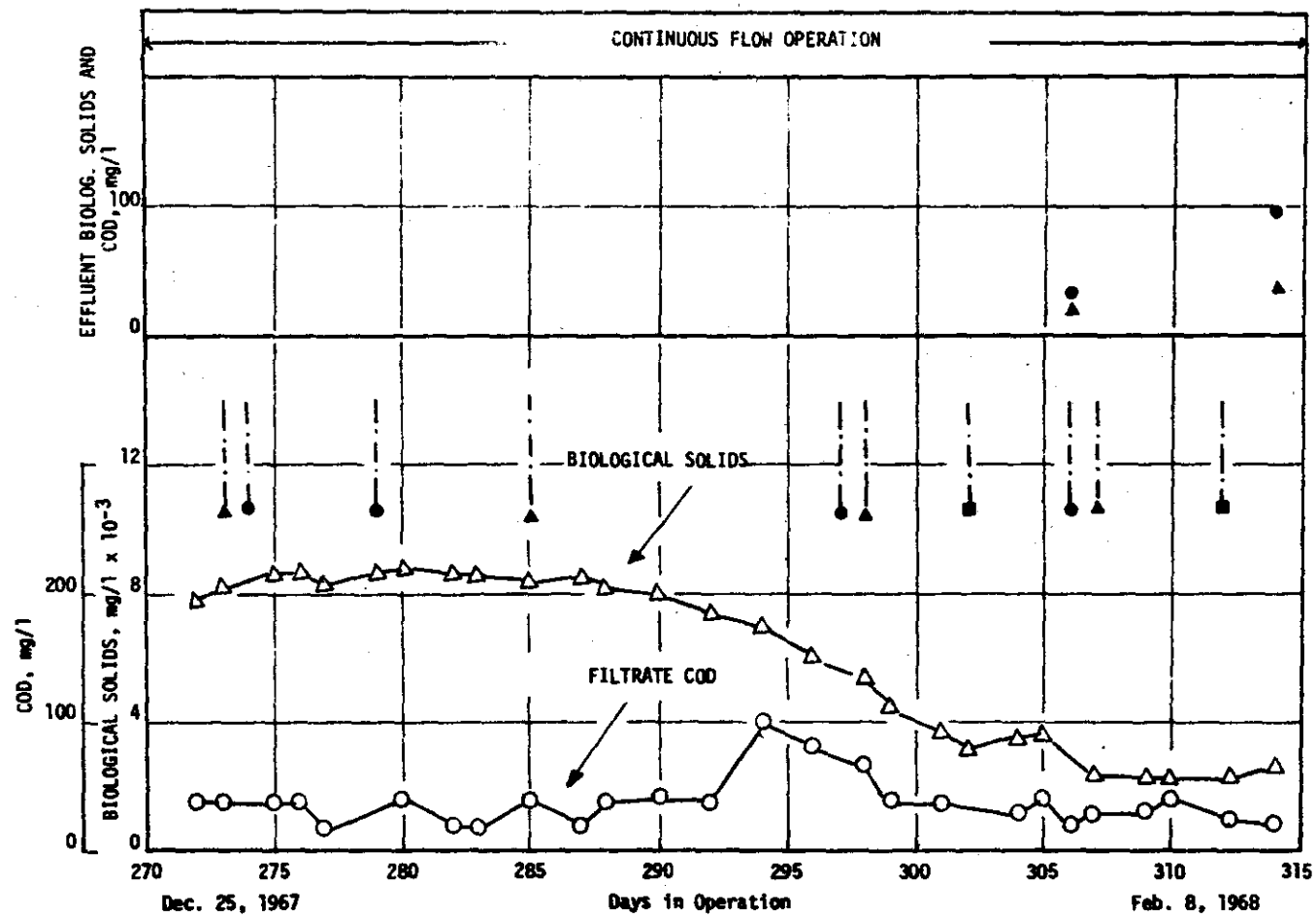


Figure 9 - Operational characteristics, December 25, 1967, to February 8, 1968.

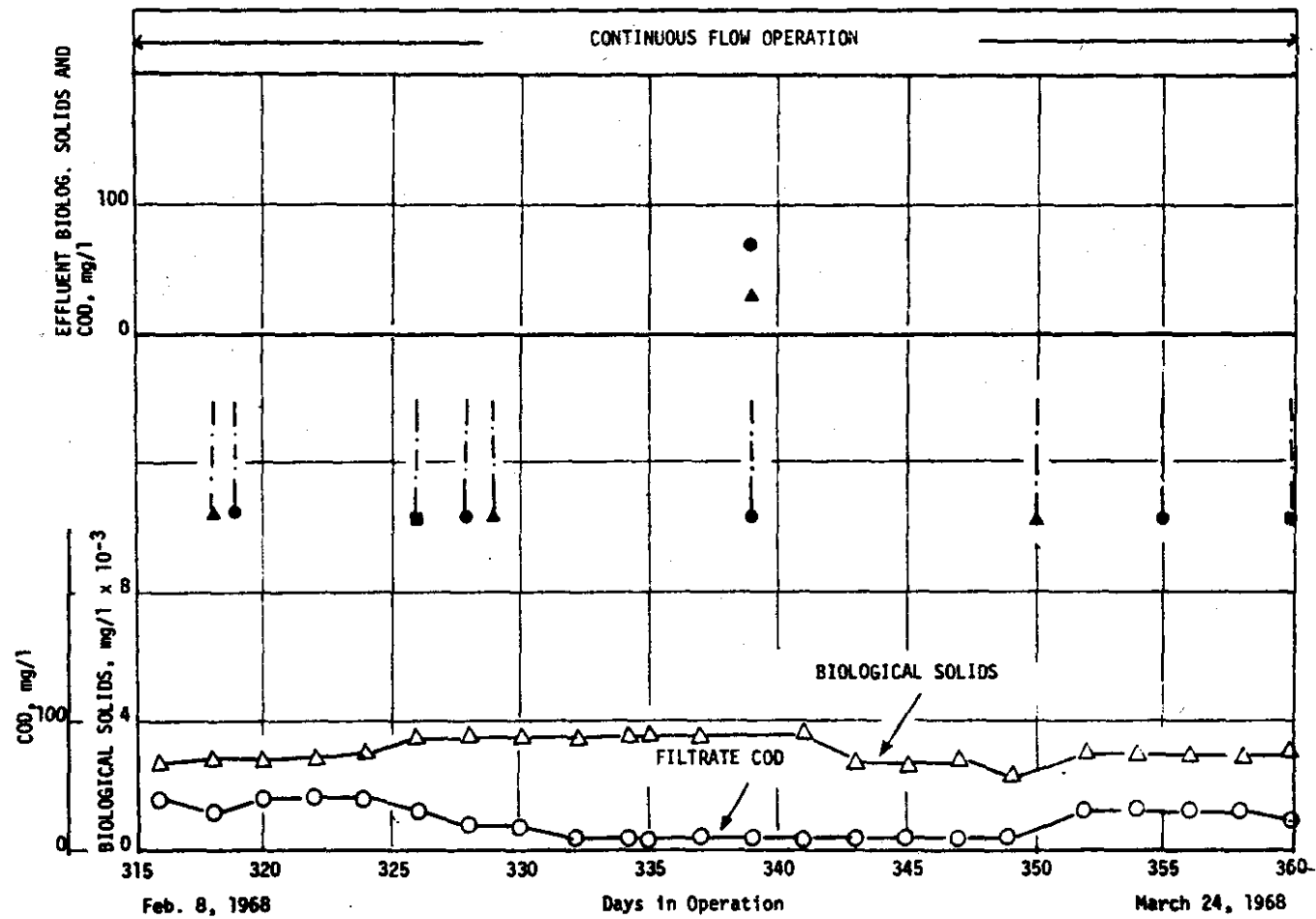


Figure 10 - Operational characteristics, February 8, 1968, to March 24, 1968.

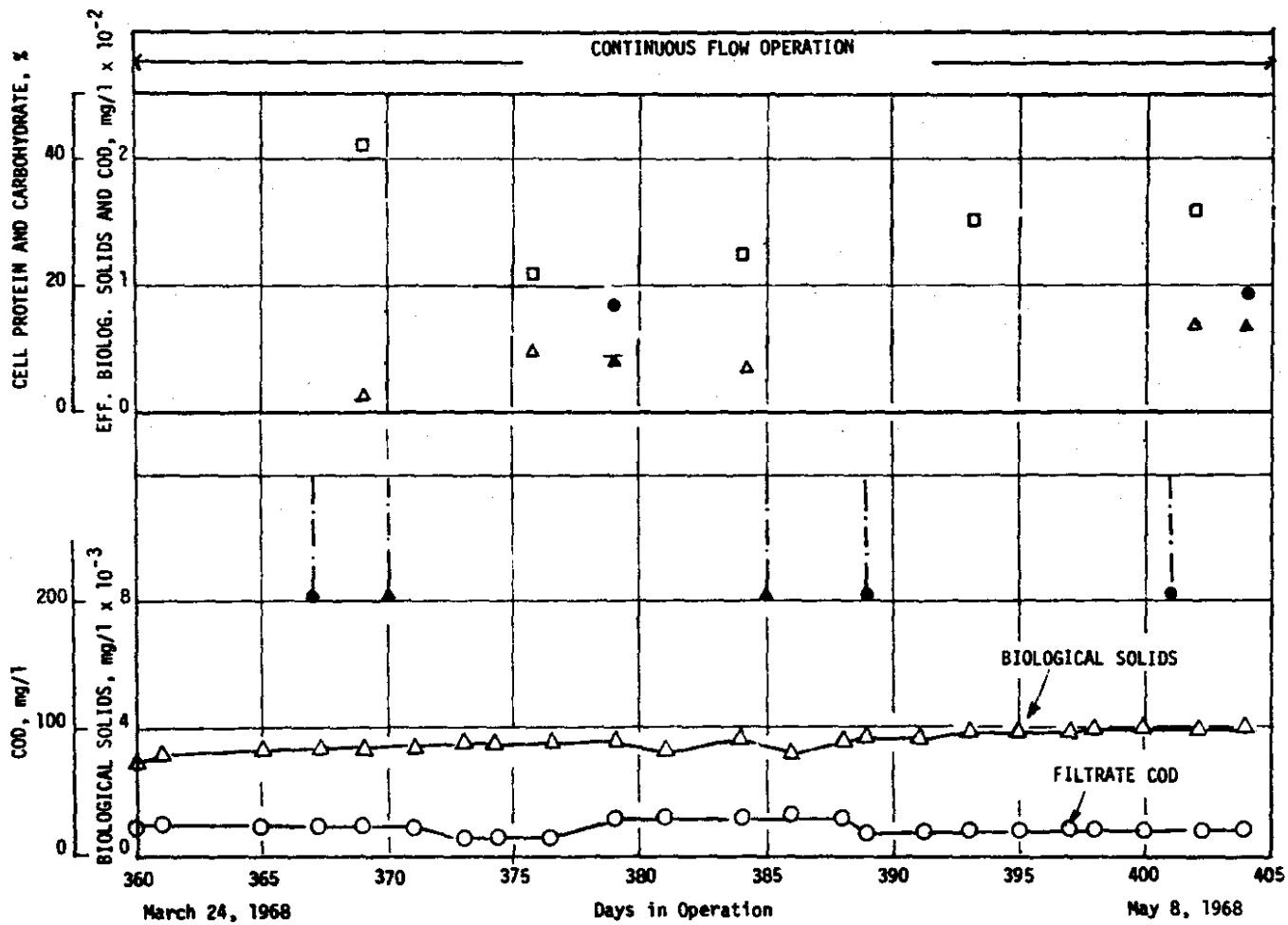


Figure 11 - Operational characteristics, March 24, 1968, to May 8, 1968.

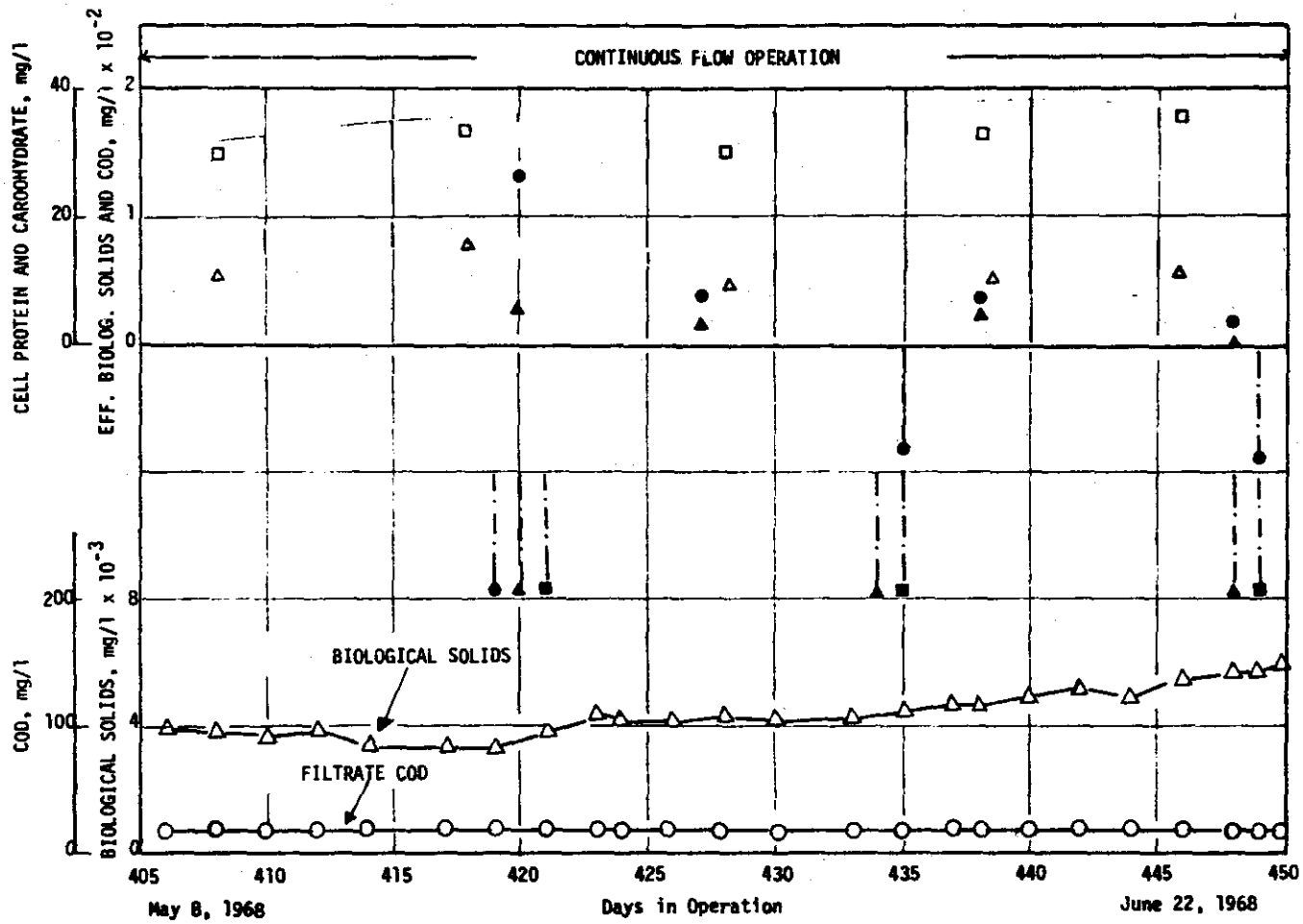


Figure 12 - Operational characteristics, May 8, 1968, to June 22, 1968.



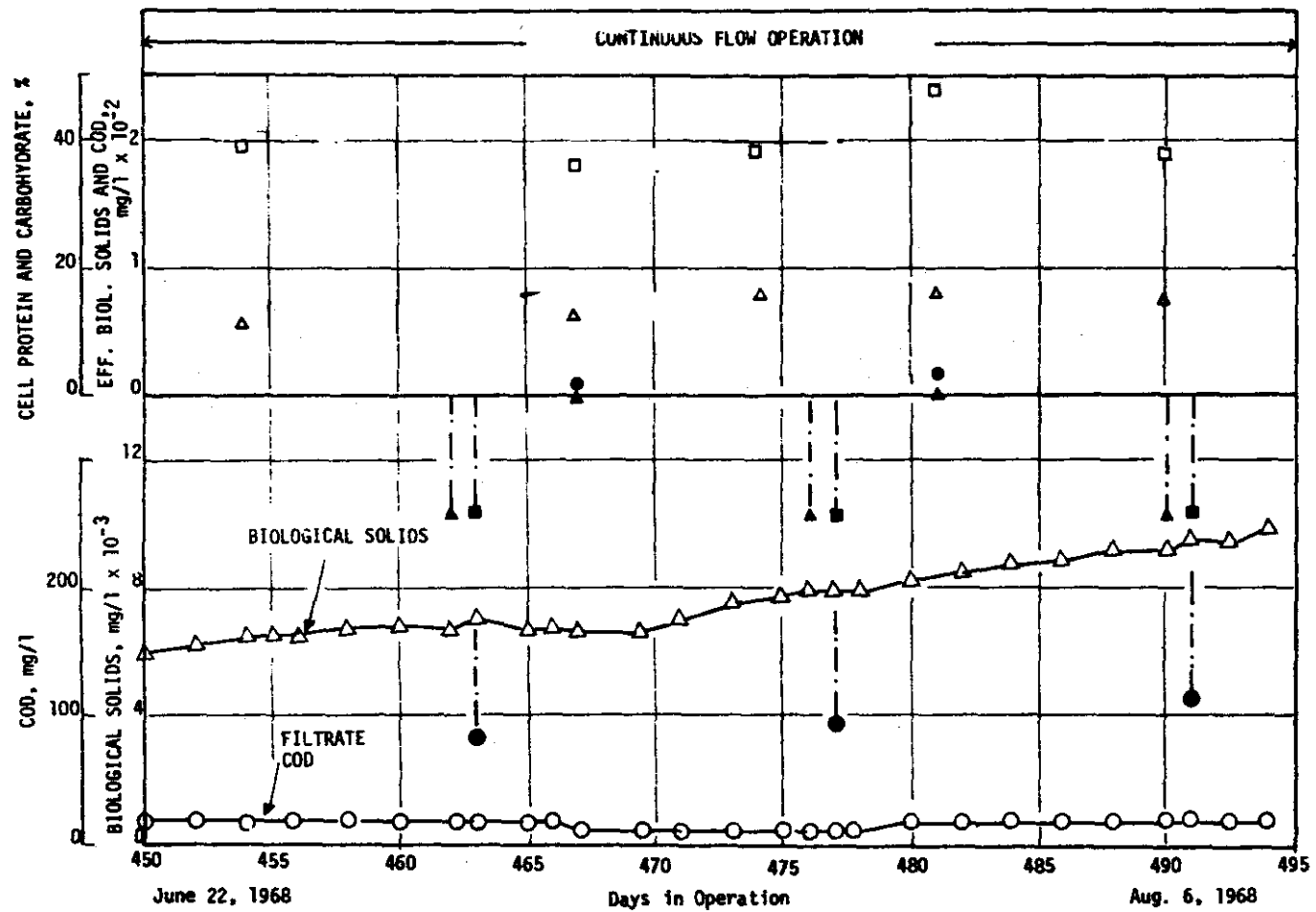


Figure 13 - Operational characteristics, June 22, 1968, to August 6, 1968.

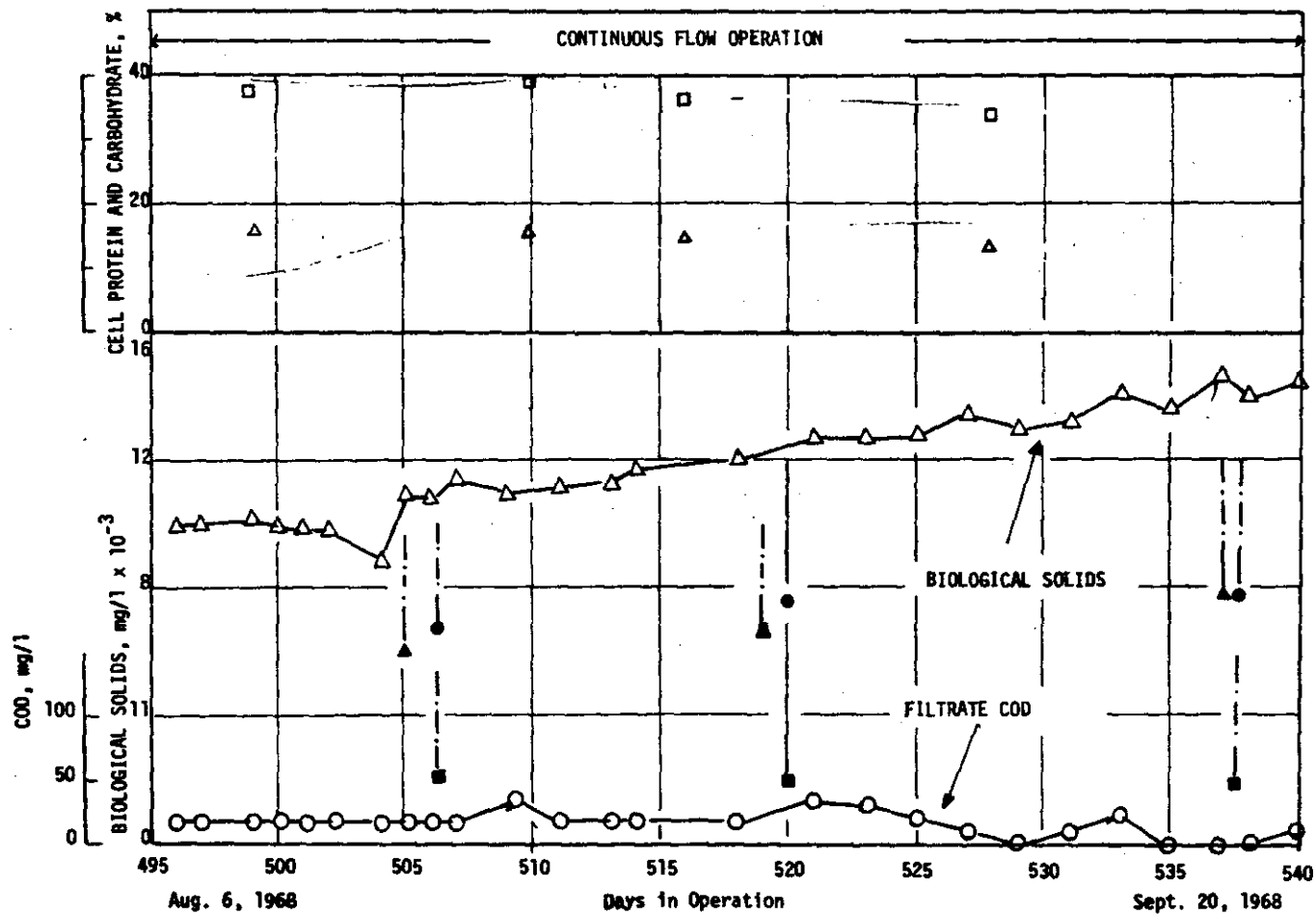


Figure 14 - Operational characteristics, August 6, 1968, to September 20, 1968.

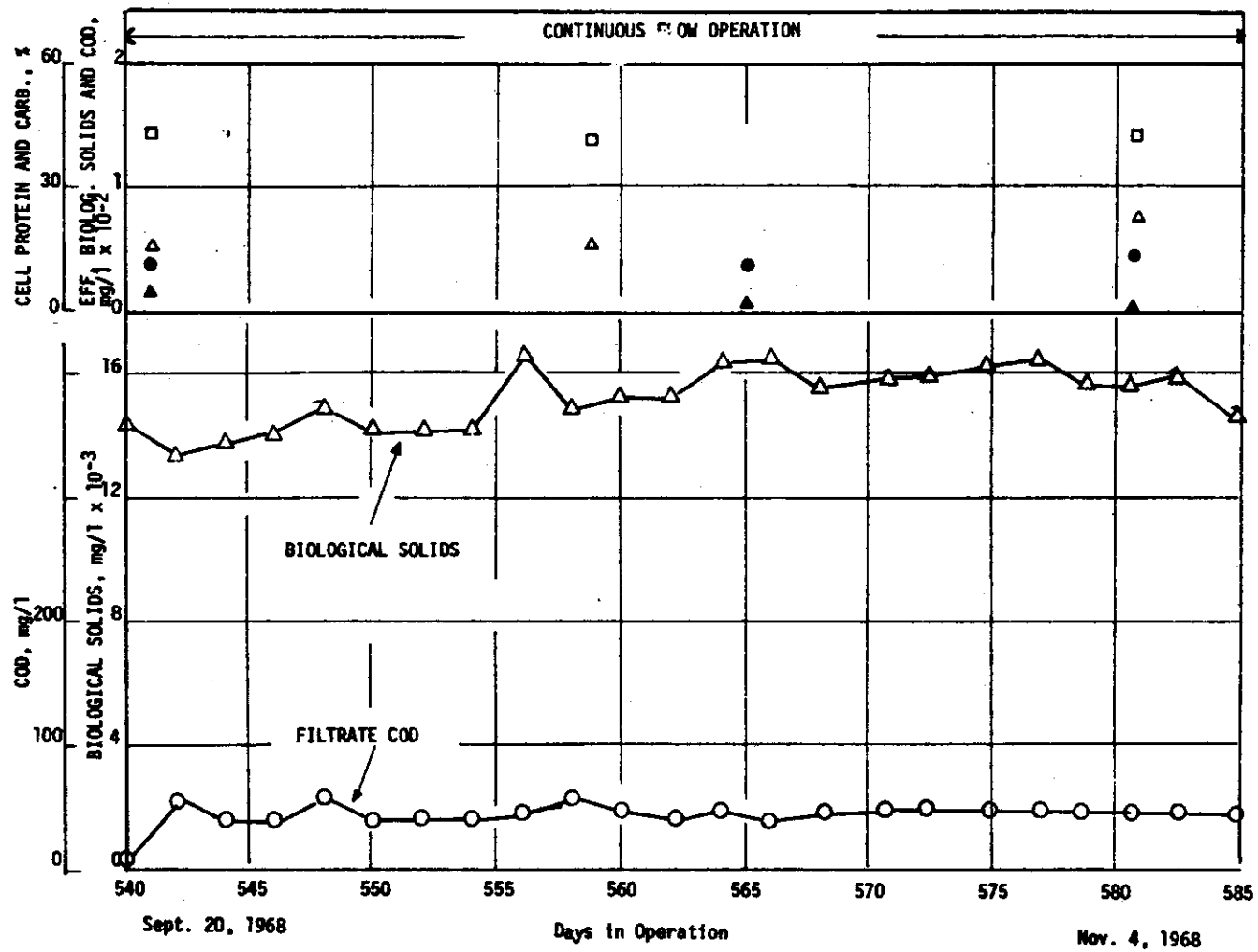


Figure 15 - Operational characteristics, September 20, 1968, to November 4, 1968.

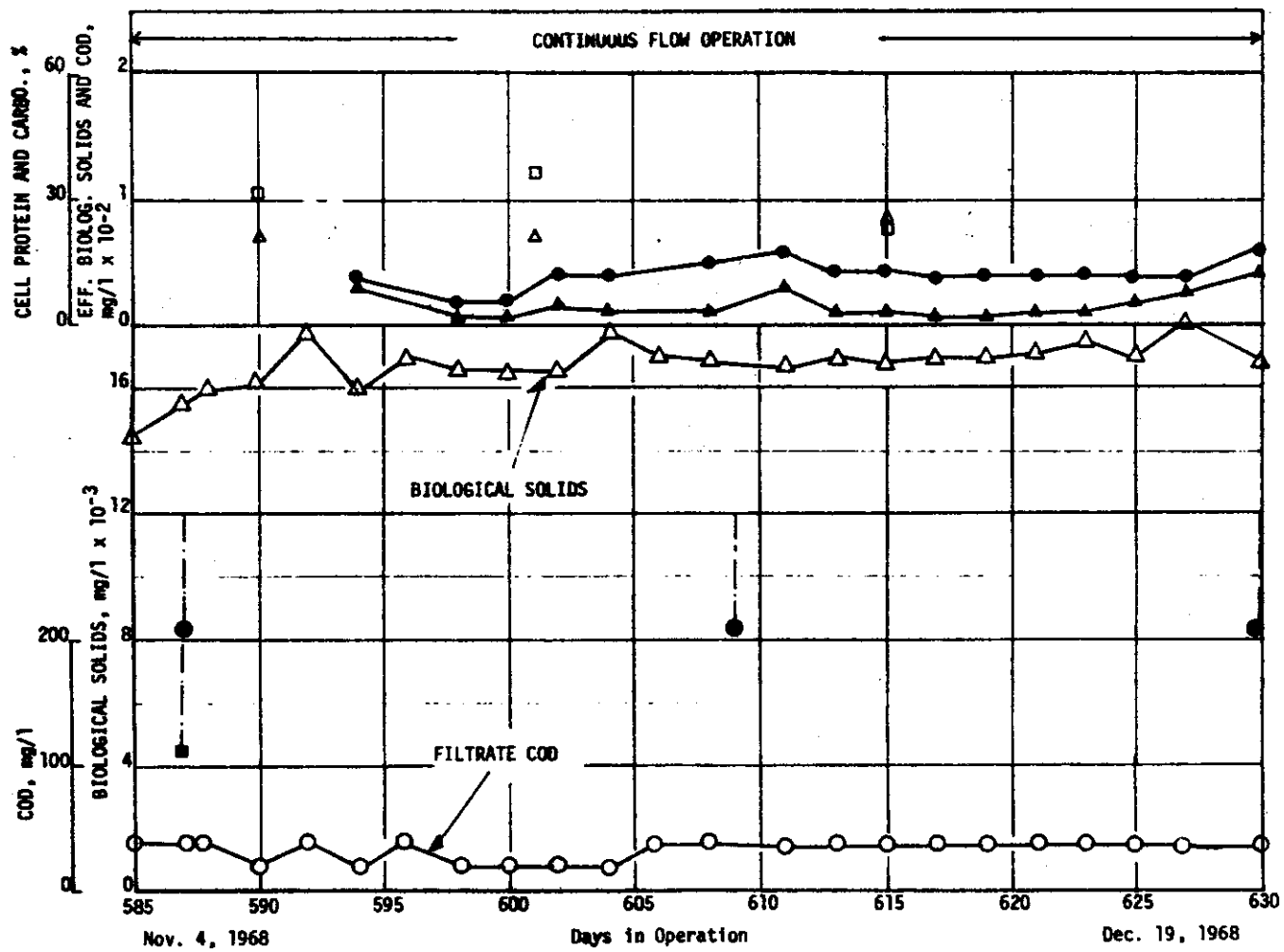


Figure 17 - Operational characteristics, November 4, 1968, to December 19, 1968.

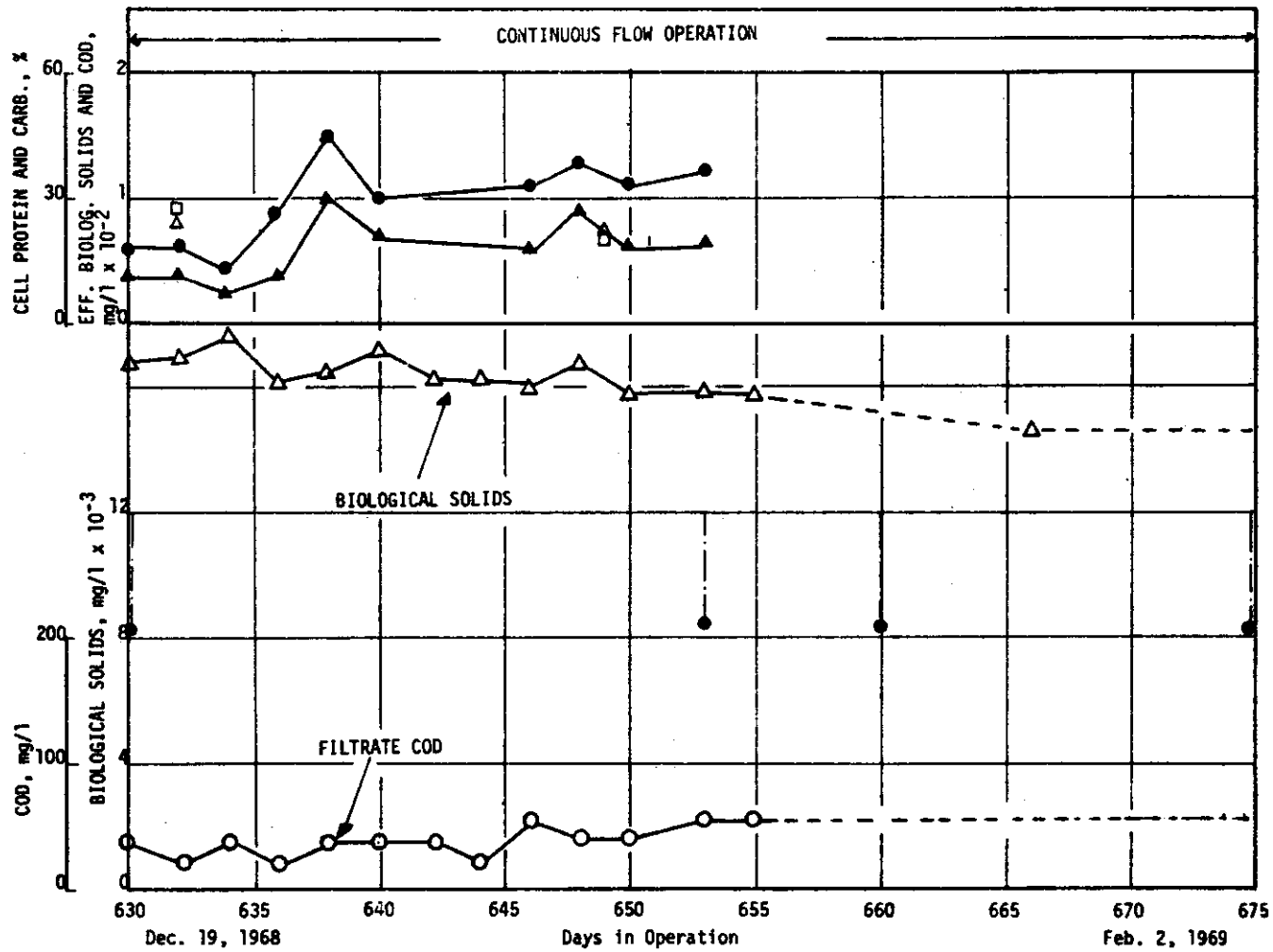


Figure 17 - Operational characteristics, December 19, 1968, to February 2, 1969.

of the biological solids in the effluent were passed through the Sharples centrifuge and returned to the aerator. The purification efficiency was 95 per cent during this period. From day 40-70 (Figures 3 and 4), the biological solids concentration rose steadily to approximately 12,000 mg/l. During this period the purification efficiency was also approximately 95 per cent.

Between days 70 and 80, biological solids concentration decreased from 12,000 mg/l to 8500 mg/l. During this period the effluent solids concentration increased, and there was also a rise in filtrate COD. The COD removal efficiency dropped to between 80 and 85 per cent. Again it is emphasized that the decrease in solids concentration of the mixed liquor was due, not to the wasting of solids (either purposely or accidentally). However, during the decrease of solids concentration, some froth was formed on the surface of the liquor in the aeration chamber, and it appeared that a portion of the sludge had undergone a gradual lysis. The solids concentration levelled off at 8500 mg/l, and within five days (by day 85) the COD removal efficiency had returned to 95 per cent. During the next few months of operation the solids concentration gradually increased, and COD removal efficiency varied between 90 and 95 per cent (Figure 5).

Beginning on September 5, 1967 (day 159), the mode of operation was changed from continuous flow to batch (Figure 6). This was done partially as a convenience, and partially because it seemed desirable to observe the behavioral patterns under once daily rather than continuous feeding. By the 192nd day of operation (October 8, 1967)(see Figure 7), the biological solids concentration had built up to nearly 30,000 mg/l. At this solids concentration the sludge did not settle,

but the biochemical efficiency of the unit was not impaired. At this time plans were made to divide the unit in order to run another total oxidation unit, so that the shock load responses of such sludge could be studied (9). On day 193 there was a slight decrease in biological solids concentration in the unit. By day 196, the solids concentration had decreased to 17,600 mg/l. In a period of four days the system experienced a one-third decrease in biological solids concentration--a change equal in magnitude to that which had previously occurred; substrate efficiency remained high. A portion of the sludge appeared to be undergoing lysis, and the lysis products were metabolized by the remaining cells.

On October 12, 1967 (day 196), the sludge was parted into two portions (the original unit was used for the present study, while another similar unit was employed for the shock loading studies). The original unit was operated on a batch feeding cycle until December 5, 1967 (day 249), at which time continuous flow operation was resumed. In order to maintain a relatively constant organic loading (on the basis of COD per unit weight of sludge in the original unit), the feed concentration was halved; i.e., 500 mg/l rather than 1000 mg/l was fed.

During the period of batch operation, the residual COD in the unit rose gradually to approximately 500 mg/l (see Figures 7 and 8). This high residual COD does not necessarily represent a deterioration of COD removal efficiency. In a later portion of this chapter, experimental results in defense of the above statements will be presented. The buildup of residual COD is believed to be due to the mode of batch operation. Only a small portion of the daily residual soluble COD was taken from the unit, thus the residual COD in the unit gradually

accumulated. The fact that it did not continue to rise throughout the period of batch operation suggests that even this residual material was eventually subject to some degree of biological degradation.

In mid-January, 1968 (see Figure 9), the system began another cycle of decreasing solids concentration. From day 285 to 307, the biological solids concentration decreased from approximately 8500 to 2400 mg/l. The filtrate COD which had been averaging approximately 40 mg/l (i.e., approximately 95 per cent COD removal efficiency), rose to 100 mg/l during the early period of the decrease in solids concentration, but in less than a week returned to its previous level. This cyclic decrease in solids concentration was a more severe depression in solids concentration than those previously observed, but it was more gradual, requiring over 20 days. Again, it is emphasized that the solids were not lost or wasted; also there was no operational change (pH, temperature, etc.) which could have caused the decrease. This cyclic decrease in solids concentration can be attributed only to natural causation, and was brought about by the biological system itself. As before, there was some amount of froth on the surface of the aeration liquor. The general appearance was observed when microbial cells underwent lysis. It is apparent that if the lysis did occur, the organic products released upon cell disruption or dissolution were metabolized by the intact cells, since the COD removal efficiency was scarcely interrupted during this period.

The biological solids concentration remained at approximately 3000 mg/l through February and late March (see Figure 10) and then (see Figures 11, 12, 13, and 14) began a gradual rise during the following days, and values of approximately 16,000 were attained in October, 1968



(Figure 15). During this period both the biochemical and total purification efficiency were excellent. Spot checks on the biological solids concentration and COD concentration in the supernatant indicated that at times the overall efficiency was approximately 80 per cent or below (see days 379, 404, and 420), and at times the overall efficiency was close to or as good as biochemical efficiency (see days 427, 448, 467, 481, and 541). The protein content of the sludge was approximately 30-40 per cent, and carbohydrate content ranged between 10 and 20 per cent. When the solids concentration started to increase more constantly after 470 days of operation, the protein content of the sludge attained values of approximately 40 per cent (see Figures 13 and 18, days 481, 490, 499, 510, 516, and 528). The characteristic behavior of the process from November 4, 1968, through January, 1969, is shown in Figures 16 and 17. The biological solids concentration remained between 16,000 and 17,000 mg/l, and early in 1969 began a slight decline. Through the fall and winter months the biochemical efficiency remained at approximately 90 per cent, and sludge settleability and COD removal efficiency were excellent, as may be seen by the low supernatant COD and the biological solids concentration in the effluent. Late in 1968, the solids carried over in the settling tank supernatant COD rose. Again it is emphasized that the solids were not lost but were harvested by centrifugation and returned to the aerator. It is interesting to note that during the latter part of 1968 and into 1969 the protein content of the sludge decreased to between 20 and 25 per cent, whereas the carbohydrate content approached 20 per cent. The system is still in operation, and at the time of preparing this report (March, 1969), the biological solids concentration is

14,500 mg/l - 15,500 mg/l, and the COD removal efficiency is near 100 per cent.

## 2. Endogenous $O_2$ Uptake, Substrate Removal Rate (High and Low Initial Sludge Concentration)

In this section, various batch experiments made in order to gain further insights into the metabolic capability of the extended aeration sludge, as its age increased, are presented.

Periodically, samples of the sludge were taken from the unit, worked free of substrate, suspended in phosphate buffer, and  $O_2$  uptake was determined (over a period of six hours) for a known concentration of sludge (gm/l). The unit  $O_2$  uptake (mg/l accumulated  $O_2$  utilization + gm/l initial sludge concentration) per hour was calculated and recorded as the endogenous  $O_2$  uptake, mg  $O_2$ /gm sludge/hr.

Small samples were also withdrawn from the aerator and used as seed in the apparatus (shown in Figure 2). In these experiments, the course of biological solids accumulation and COD removal were assessed. In some of these experiments, removal of the substrate (glucose) was also measured using the anthrone test. As a mean of facilitating comparison of the results of each experiment, the total amount of COD removal (mg/l) was divided by the average biological solids concentration (gm/l), i.e., initial + final + 2. This value was then divided by the time period (hrs) of the experiment. The resultant value was recorded as "specific substrate removal rate, mg COD/gm sludge/hr. It is realized that this type of calculation provides only a rough comparative parameter, since substrate removal and sludge accumulation in these "low incident solids" systems could not often be approximated with kinetics of zero order.

At various times, experiments of the type described above were conducted in the extended aeration unit itself. Such experiments were easily facilitated during a routine daily feeding period when the unit was being batch fed and during periods of normally continuous flow operation, and the system was batch fed on the day a substrate removal rate was run. For these "high initial solids" experiments, the specific substrate removal rate was determined as for the "low initial solids" experiments.

The values for endogenous  $O_2$  uptake and specific substrate removal rate (both low and high initial solids) are shown in Figure 18. The endogenous  $O_2$  uptake rate fluctuated rather widely during the first 250 days of operation. From day 30 to day 60 it dropped sharply as biological solids continued to accumulate, and it rose sharply when the biological solids concentration in the system experienced a decrease. Unfortunately, during the rapid decrease in solids concentration which took place just prior to 200 days of operation, no  $O_2$  uptake data was taken. The most significant trend took place after day 270. Just prior to and during the gradual decrease in solids concentration (see Figures 9 and 18), the  $O_2$  uptake rate followed a rising trend. It remained rather high, and as biological solids gradually built up in the system, the  $O_2$  uptake rate followed a gradual decreasing trend and appeared to level off between one and two mg  $O_2$ /gm sludge/hr. After attainment of this low endogenous rate it was of interest to determine the endogenous rate of new or young cells grown up from a small inoculum of cells taken from the unit. Endogenous rates between 10 and 18 mg  $O_2$ /hr/gm sludge were observed. Thus, the endogenous activity of the extended aeration sludge is approximately 5 to 10 per cent of

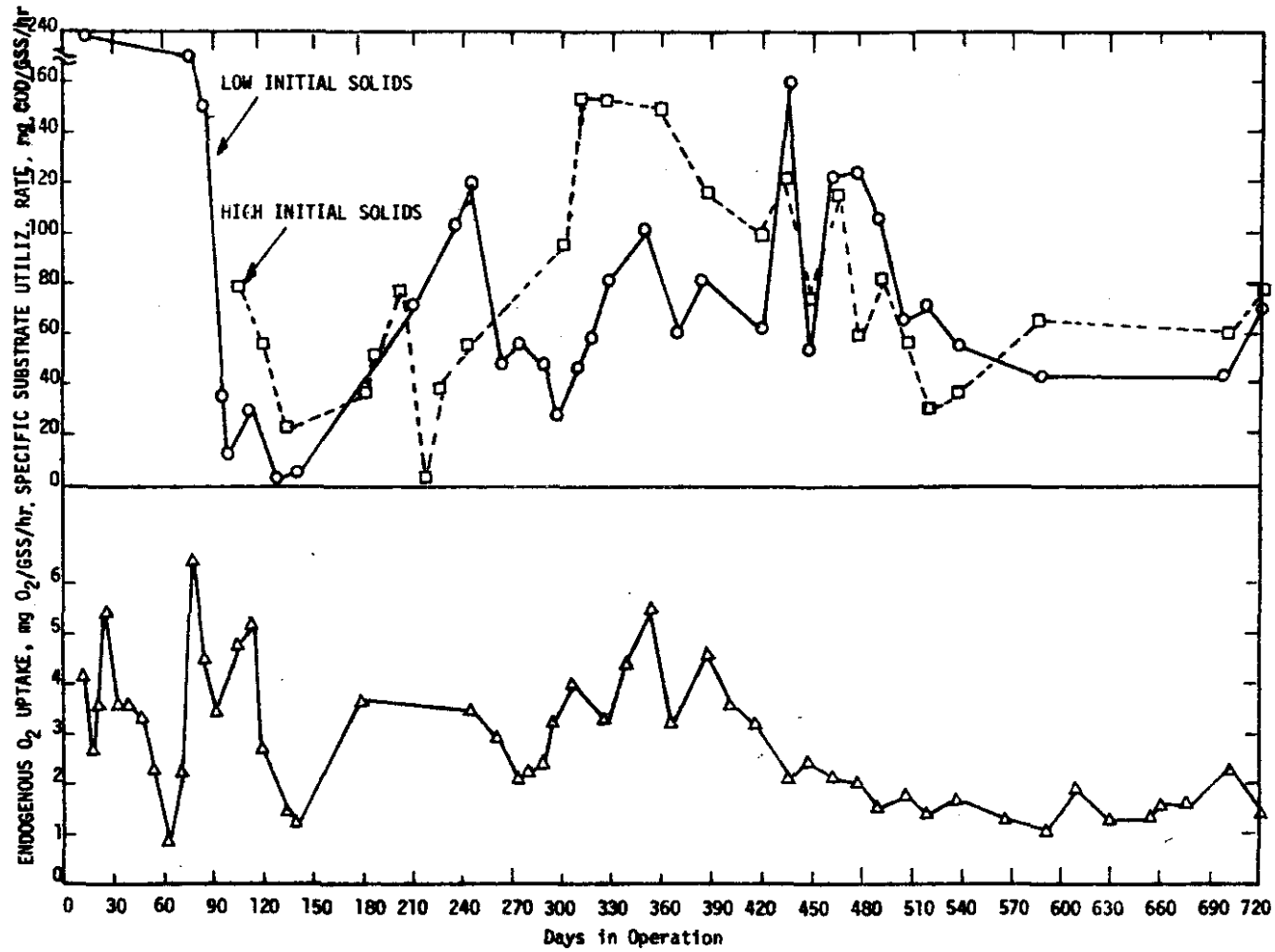


Figure 18 - Specific substrate utilization rate and endogenous O<sub>2</sub> uptake of extended aeration.

that for young cells of the same origin.

From Figure 18 it is seen that the specific substrate utilization rate for high initial solids and low initial solids systems tended to follow similar patterns. However, the high initial solids substrate utilization rate dropped to 8 mg COD/gm solids/hr on day 218, then gradually rose. The low removal rate value was obtained a few weeks after the sludge had been parted in order to run the shock load unit. Also, the unit had been recently switched from continuous flow to batch operation. During the later period of operation the values for substrate removal rate at both high and low initial solids concentration appear to have levelled off at values in the range 40-60 mg COD/gm sludge/hr.

The course of substrate removal and biological solids accumulation for the batch experiments at both high and low initial biological solids concentrations are presented in Figures 19 through 53. Since the significance of the specific substrate removal rate may be subject to debate, these figures are presented in order that a reader may scrutinize the actual experimental result rather than have only the calculated parameter upon which to base a judgement. These figures can be used in conjunction with Figures 3 through 17, as well as with Figure 18.

The low value for the specific substrate utilization rate shown in Figure 18 for day 218 (8 mg COD/gm sludge/hr) was calculated from the results shown in Figure 32. From Figure 7 it is seen that this run was made approximately three weeks after the unit had been switched from continuous flow to batch operation. During this period the residual COD in the unit after twenty-three hours of aeration was

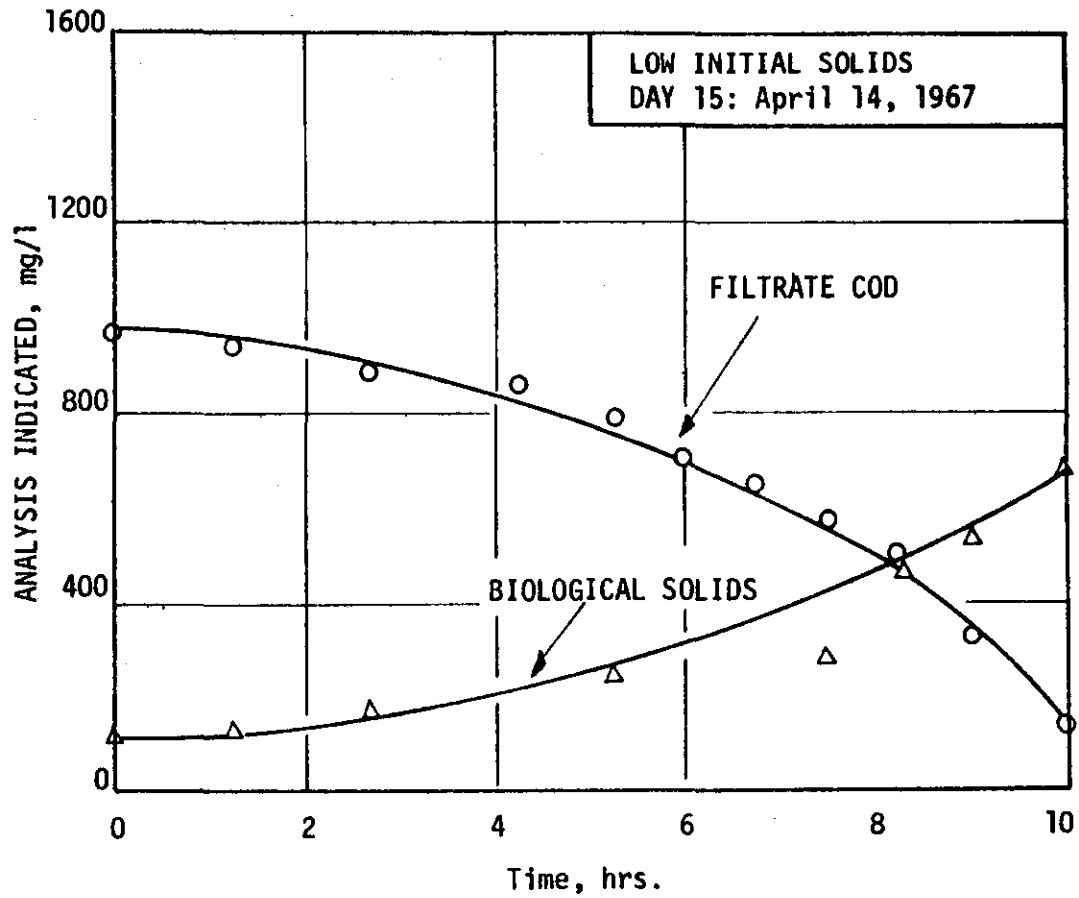


Figure 19 - Response of the extended aeration activated sludge to slug dosage of glucose after 15 days of operation.

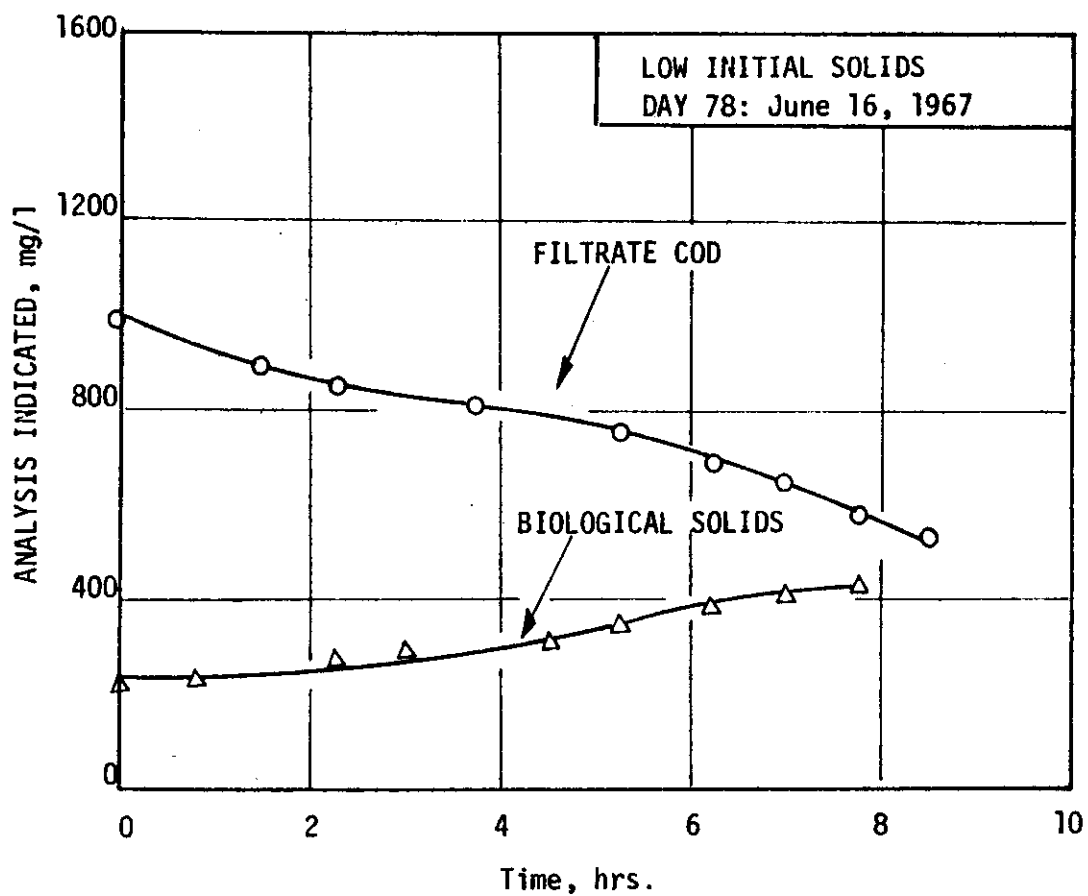


Figure 20 - Response of the extended aeration activated sludge to slug dosage of glucose after 78 days of operation.

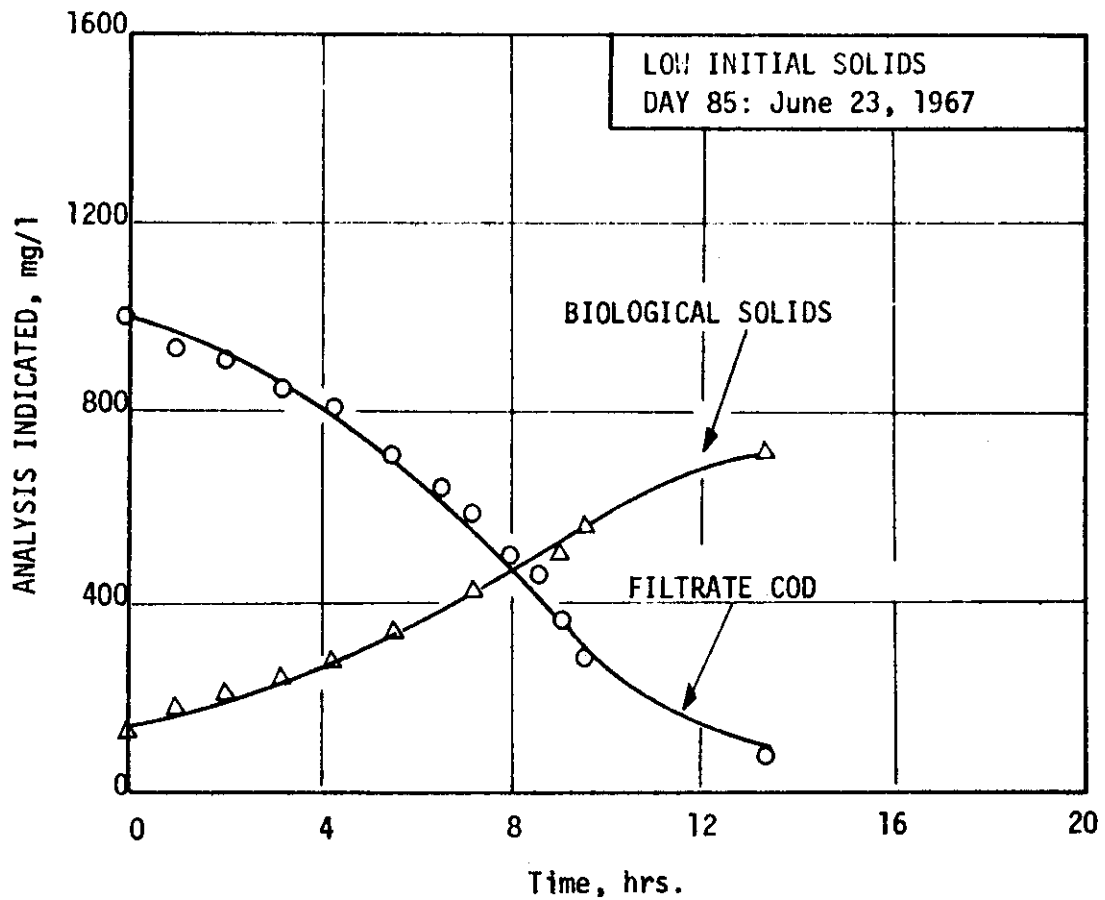


Figure 21 - Response of the extended aeration activated sludge to slug dosage of glucose after 85 days of operation.



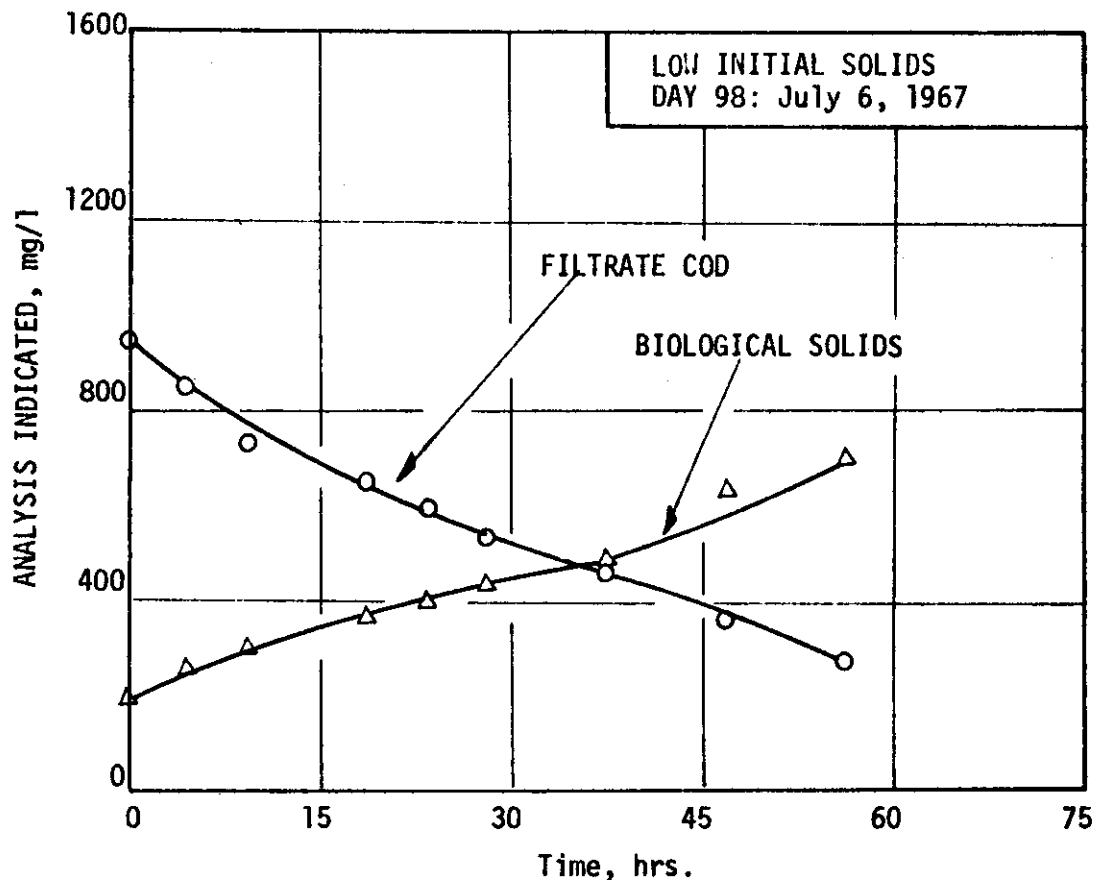


Figure 22 - Response of the extended aeration activated sludge to slug dosage of glucose after 98 days of operation.

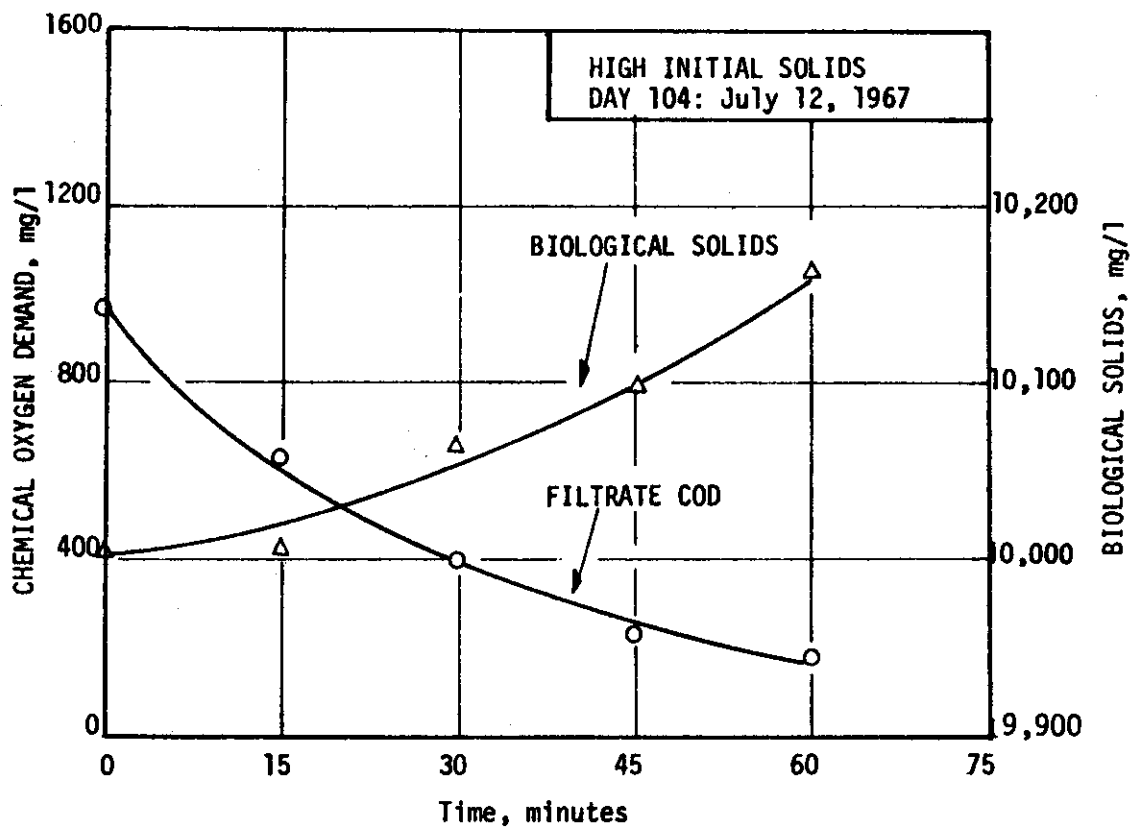


Figure 23: Response of the extended aeration activated sludge to slug dosage of glucose after 104 days of operation.

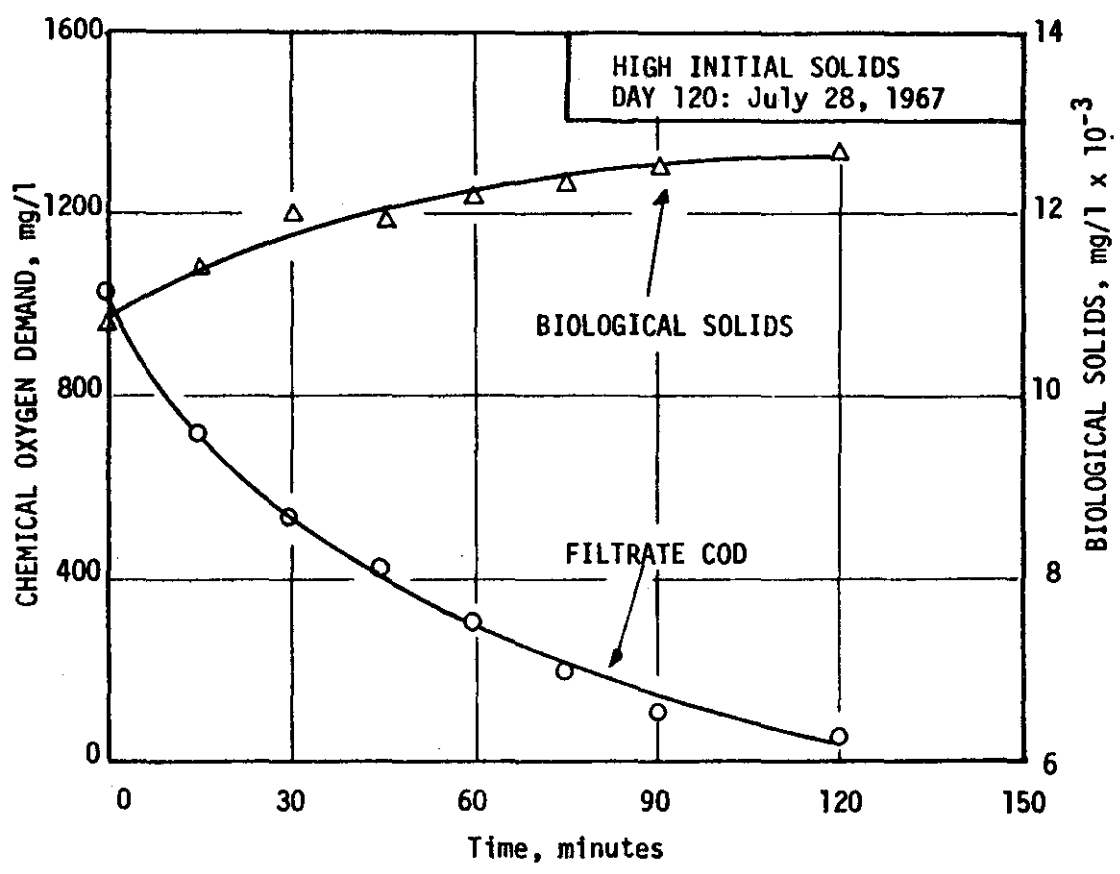


Figure 24 - Response of the extended aeration activated sludge to slug dosage of glucose after 120 days of operation.

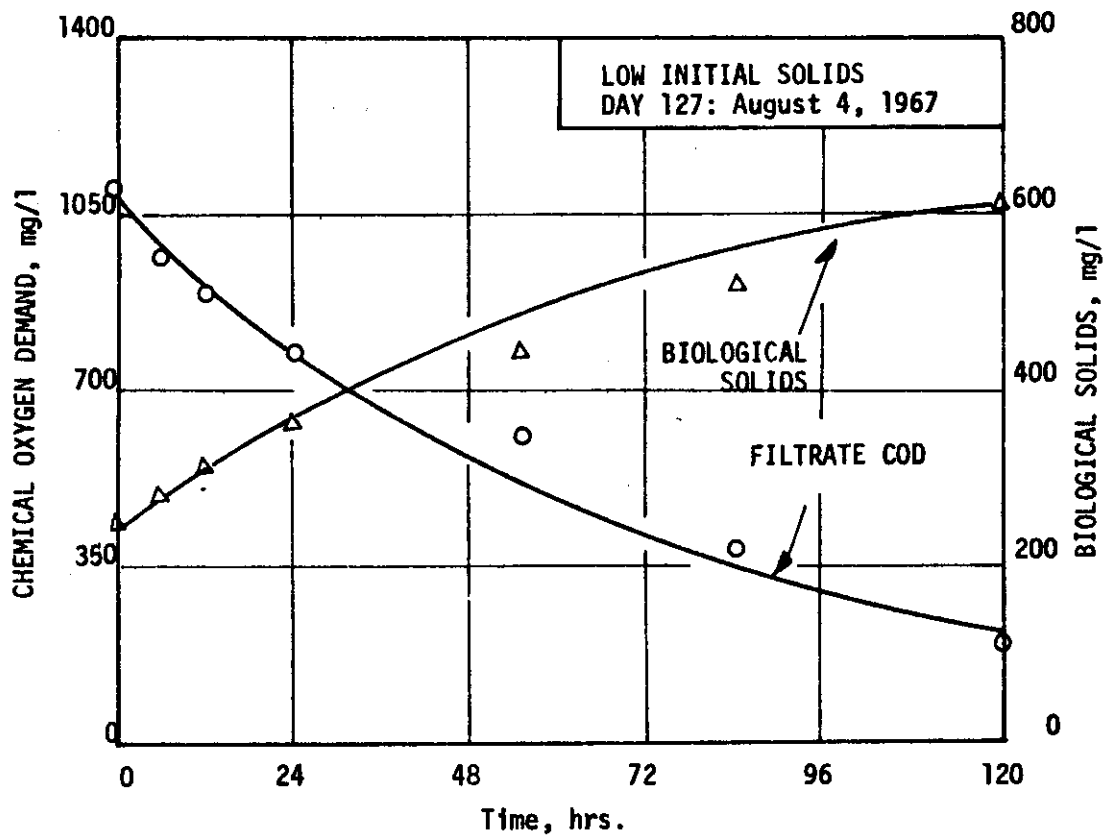


Figure 25 - Response of the extended aeration activated sludge to slug dosage of glucose after 127 days of operation.

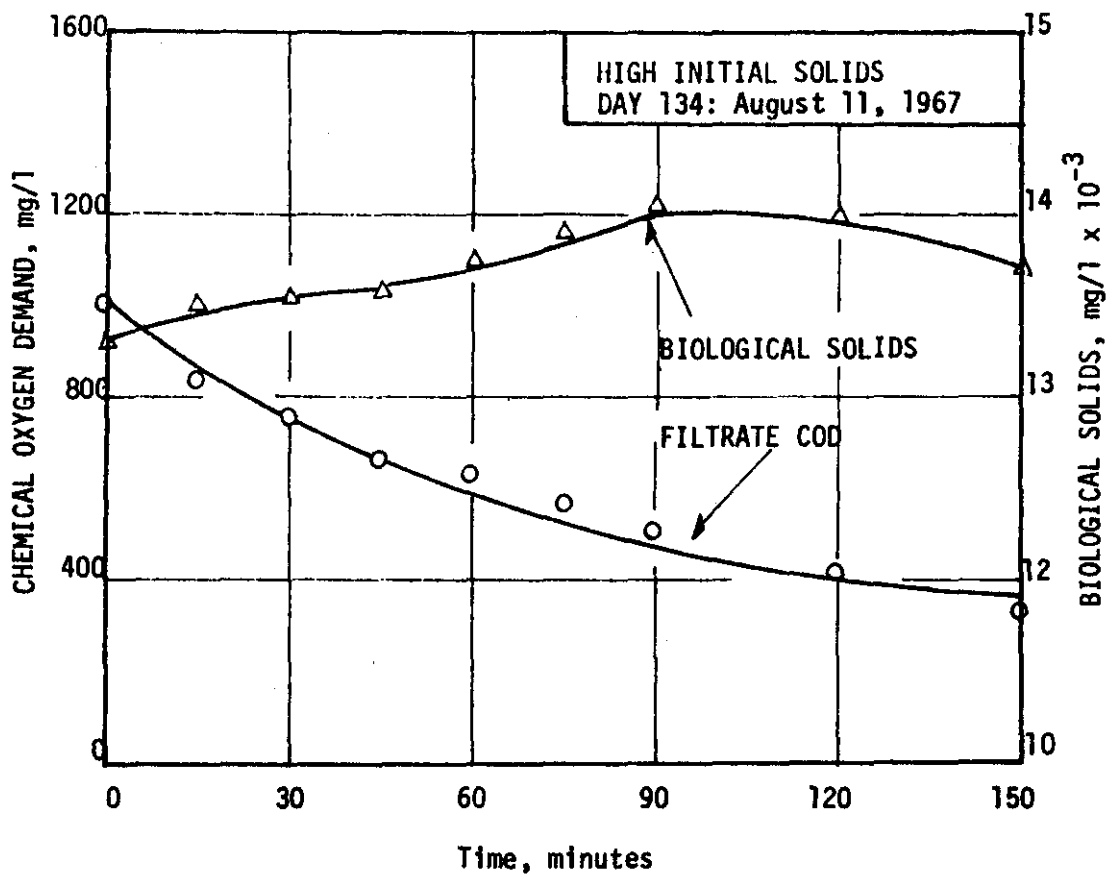


Figure 26 - Response of the extended aeration activated sludge to slug dosage of glucose after 134 days of operation.

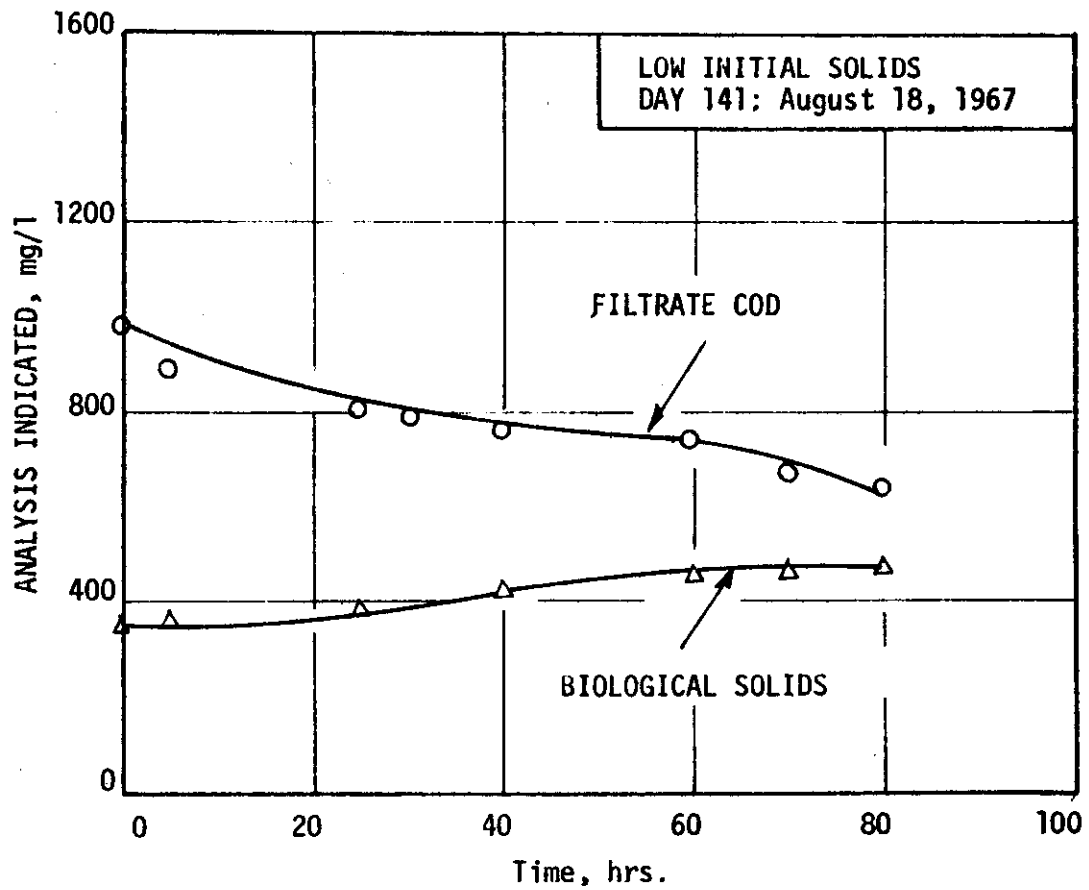


Figure 27 - Response of the extended aeration activated sludge to slug dosage of glucose after 141 days of operation.

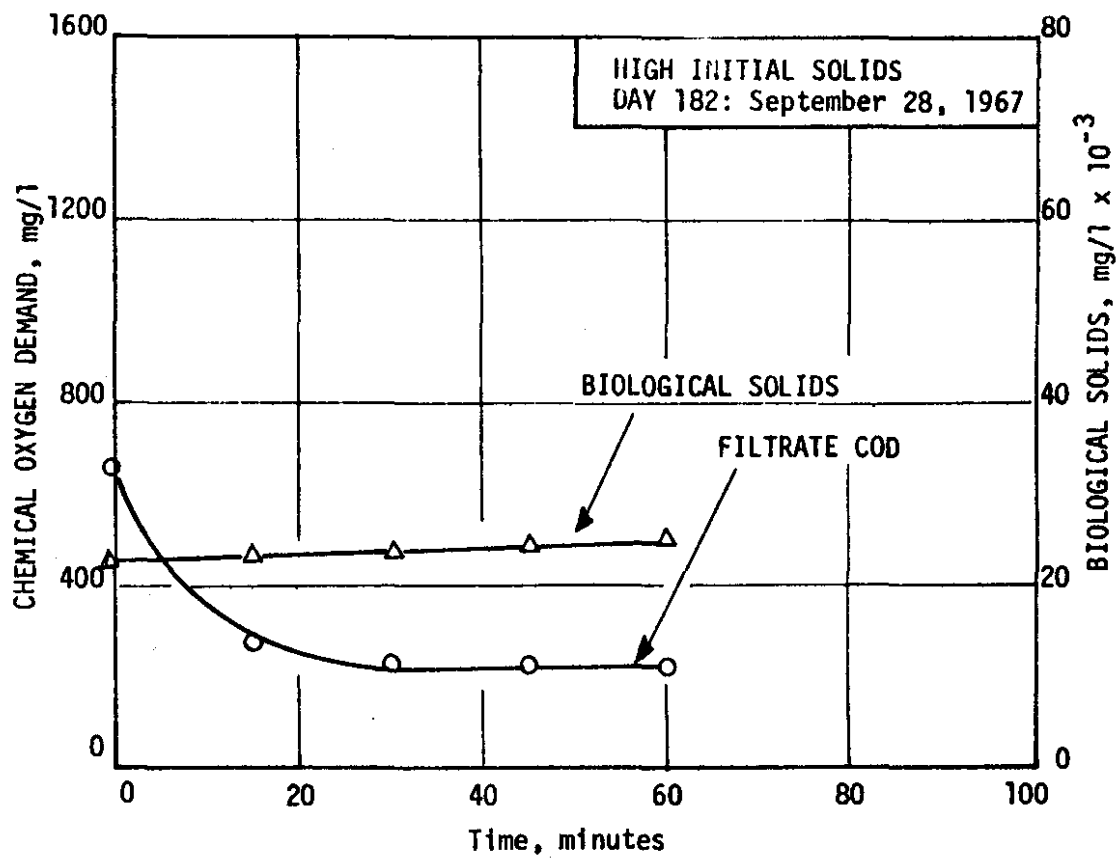


Figure 28 - Response of the extended aeration activated sludge to slug dosage of glucose after 182 days of operation.

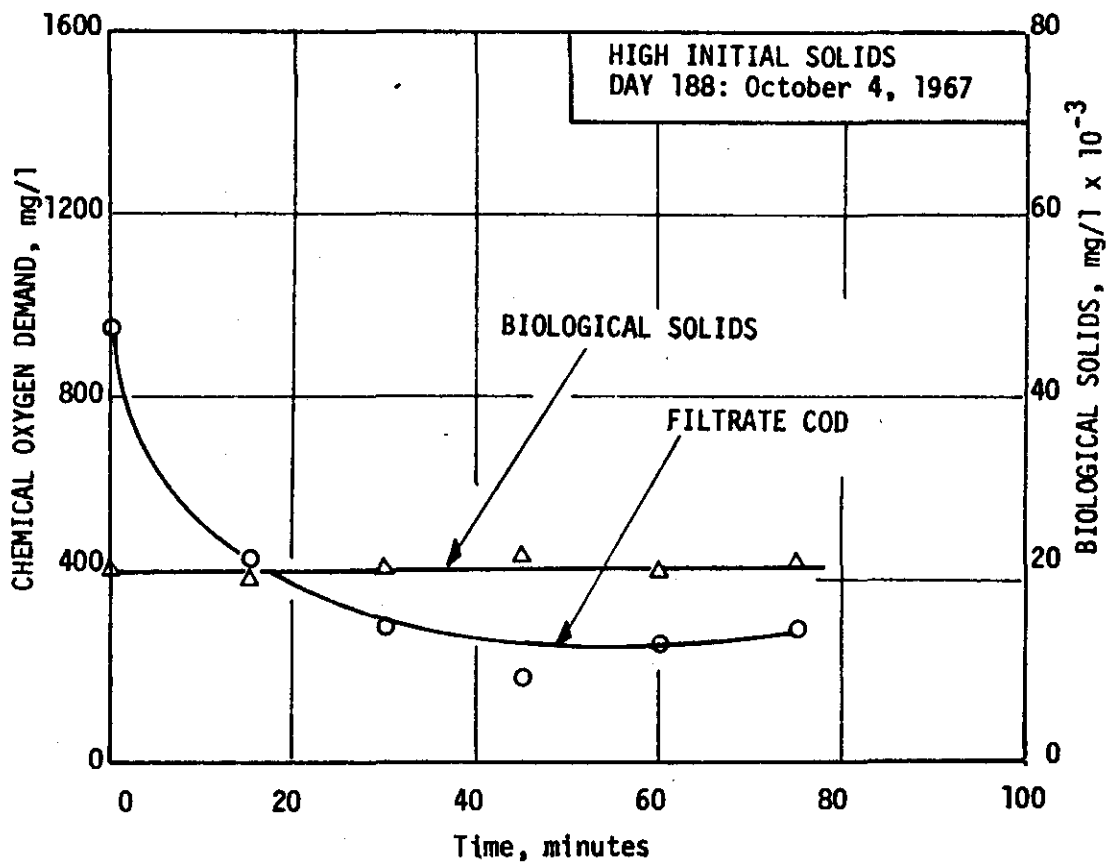


Figure 29 - Response of the extended aeration activated sludge to slug dosage of glucose after 188 days of operation.



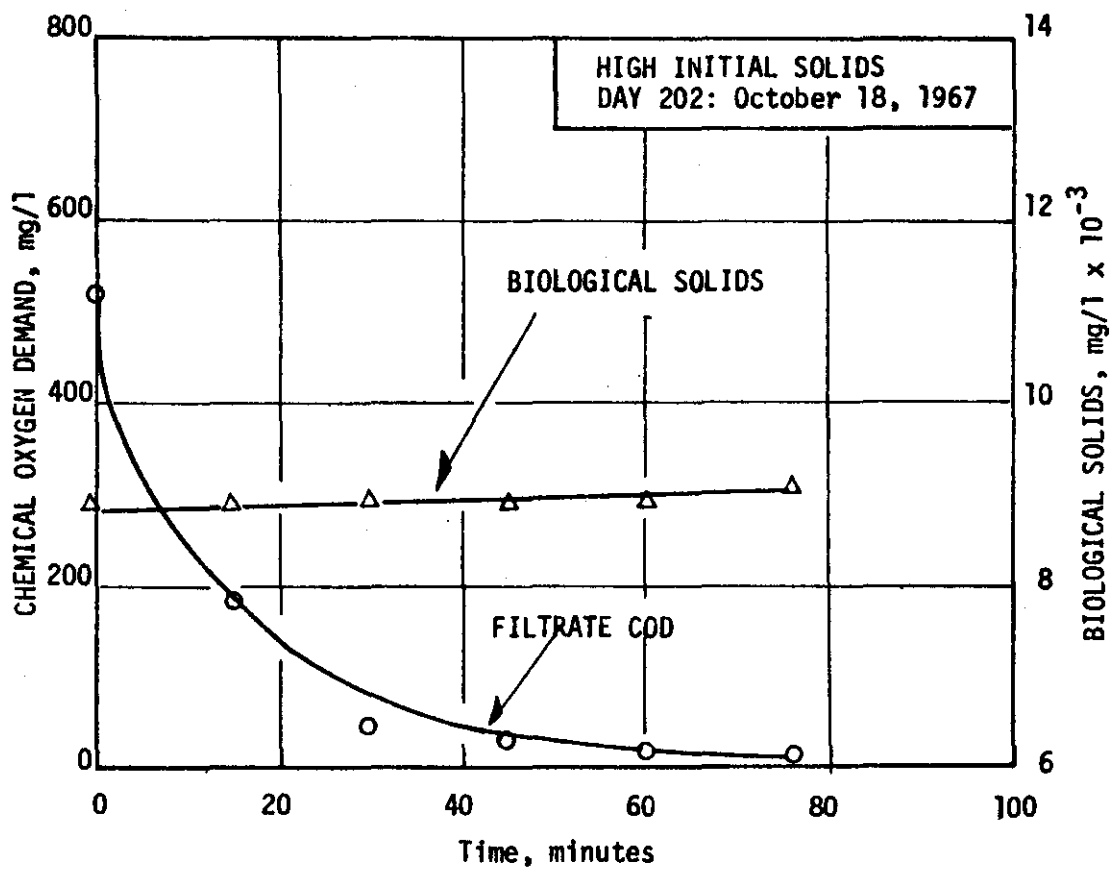


Figure 30 - Response of the extended aeration activated sludge to slug dosage of glucose after 202 days of operation.

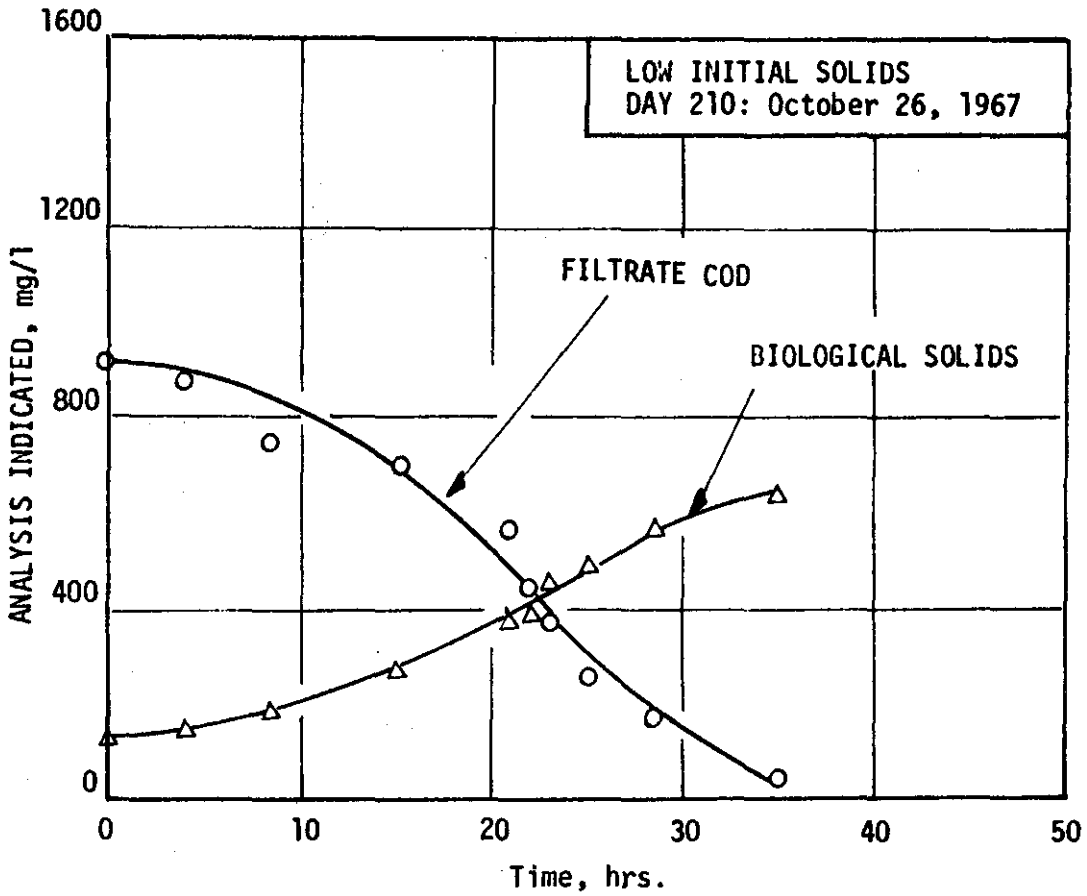


Figure 31 - Response of the extended aeration activated sludge to slug dosage of glucose after 210 days of operation.

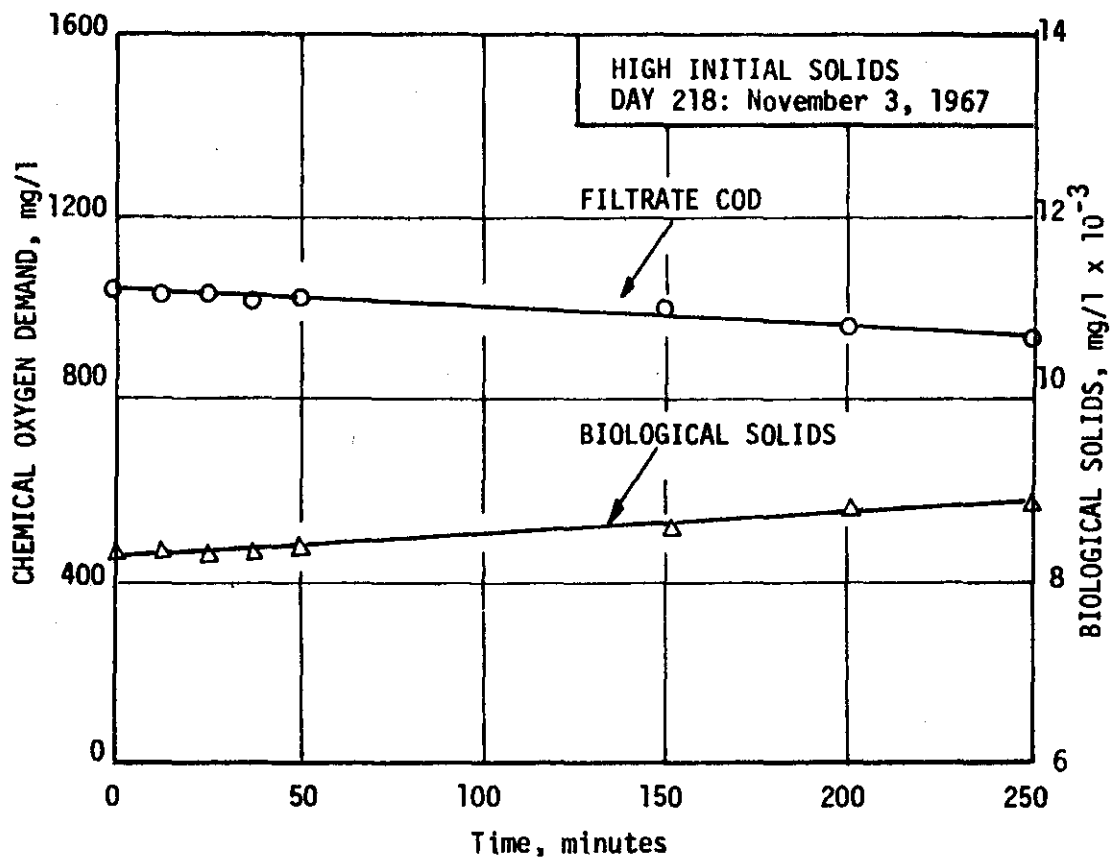


Figure 32 - Response of the extended aeration activated sludge to slug dosage of glucose after 218 days of operation

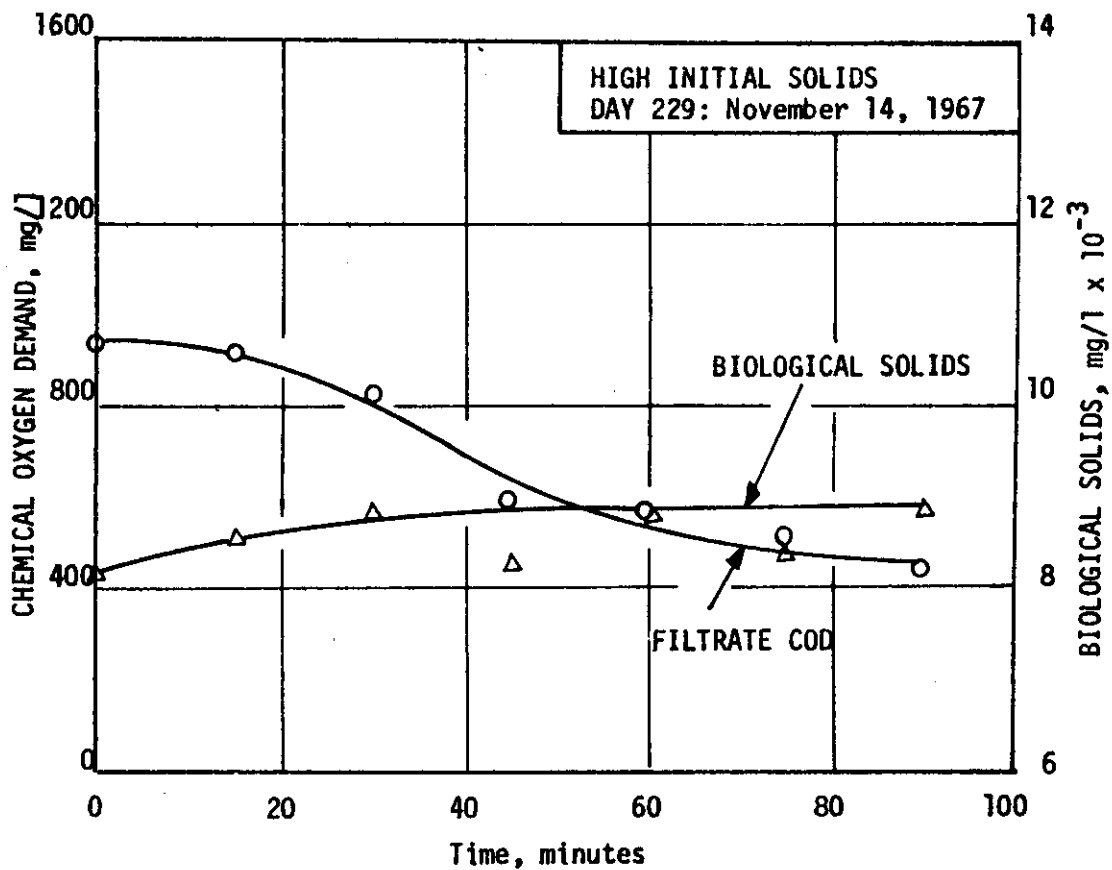


Figure 33 - Response of the extended aeration activated sludge to slug dosage of glucose after 229 days of operation.

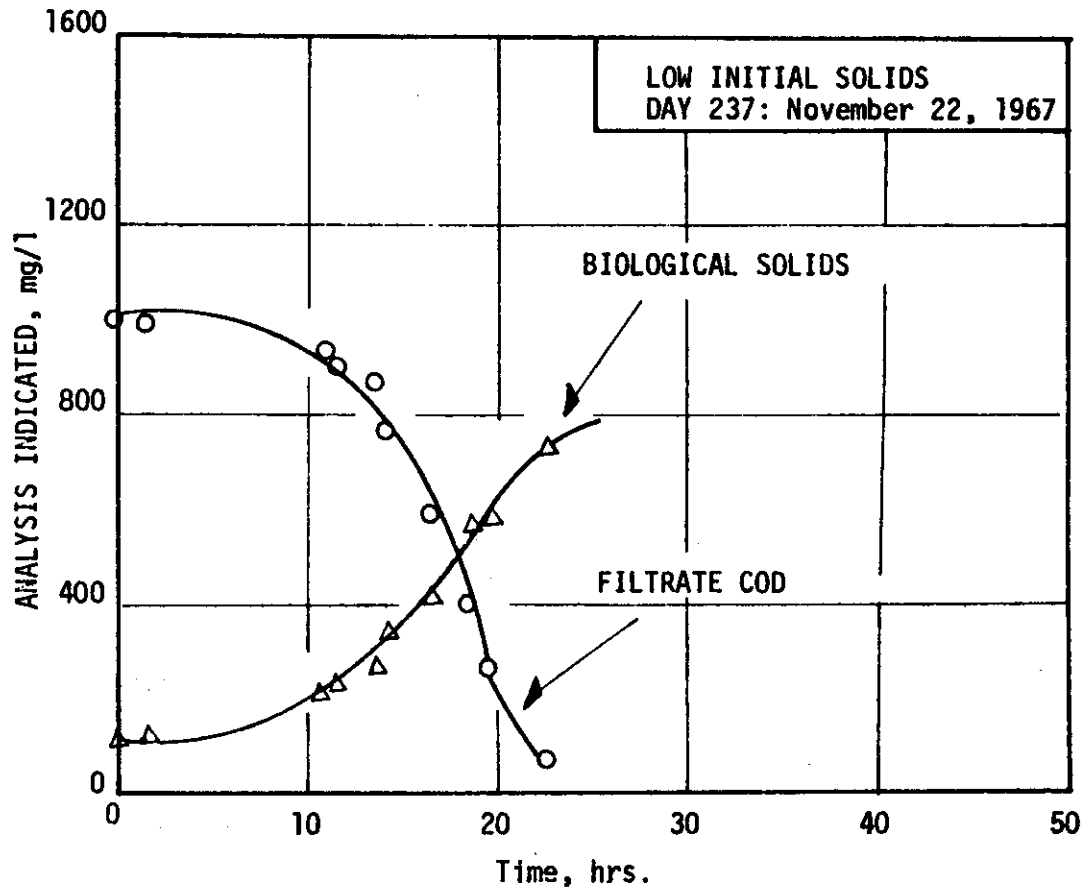


Figure 34 - Response of the extended aeration activated sludge to slug dosage of glucose after 237 days of operation.

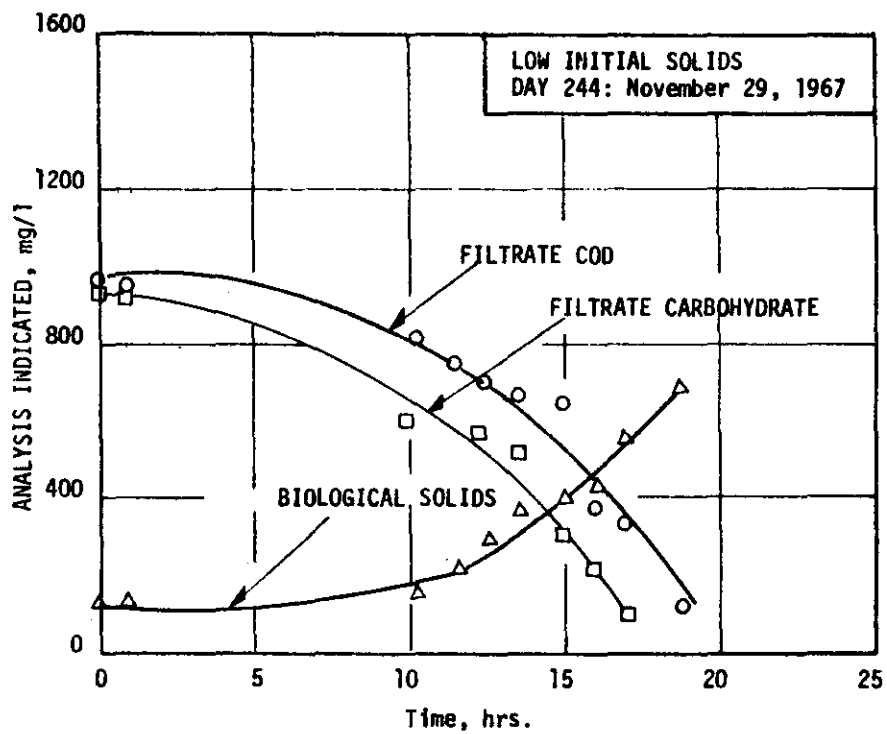
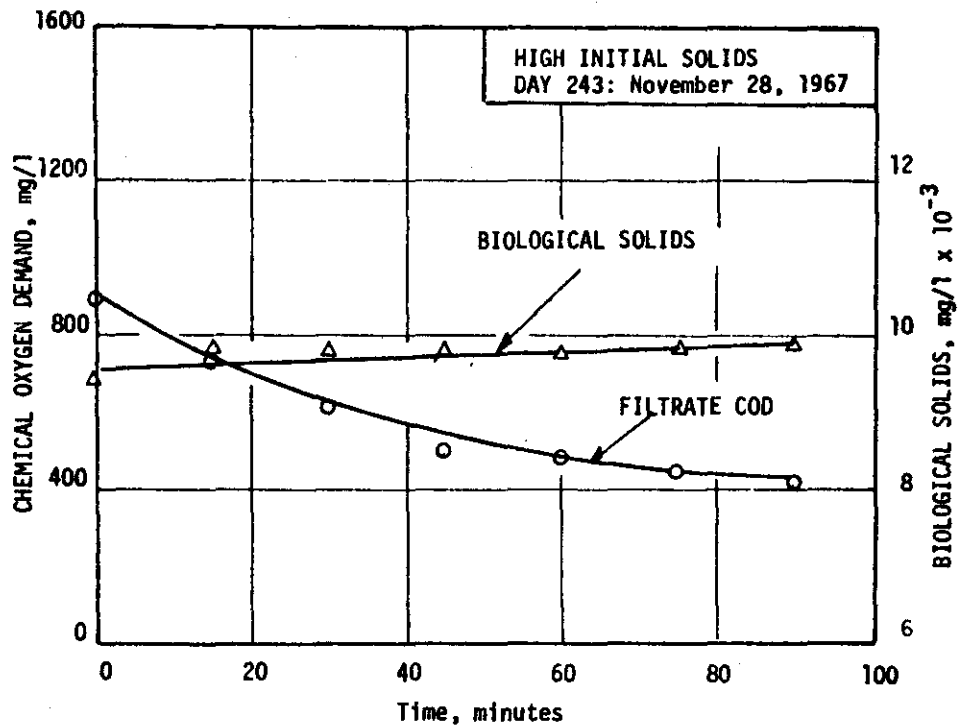


Figure 35 - Response of the extended aeration activated sludge to slug dosage of glucose after 243 days.

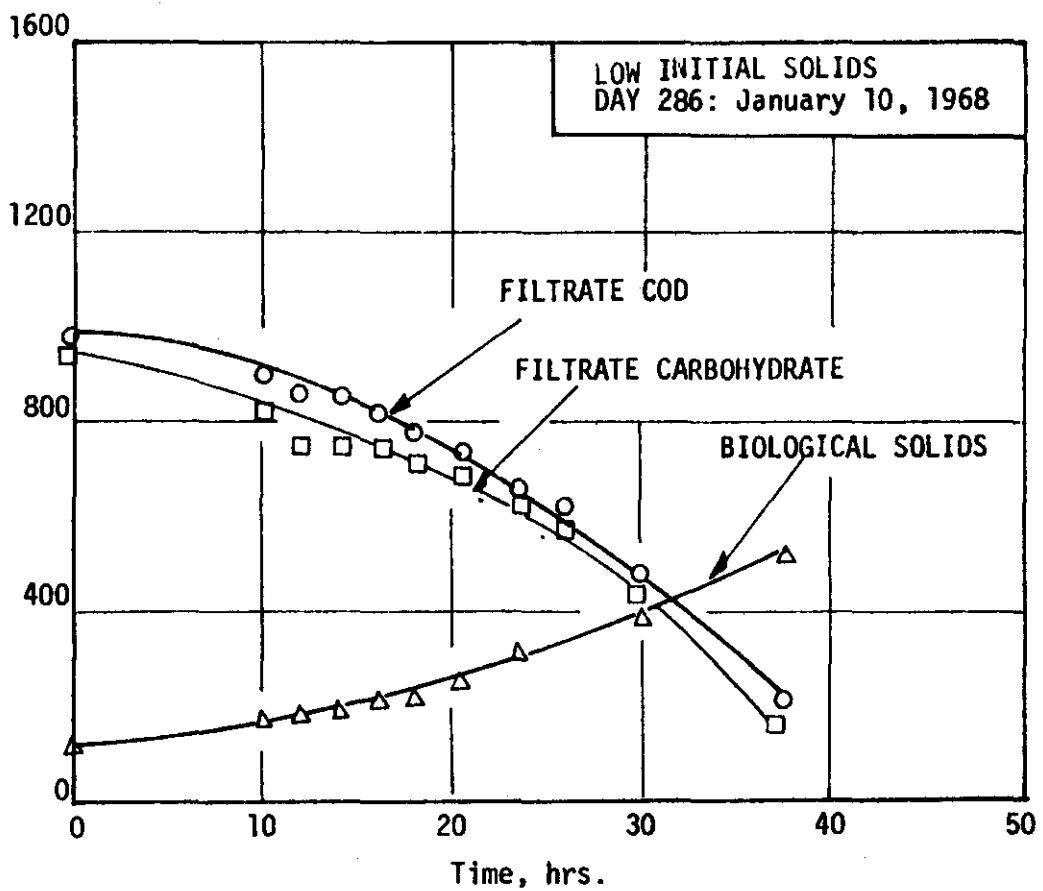


Figure 36 - Response of the extended aeration activated sludge to slug dosage of glucose after 286 days of operation.

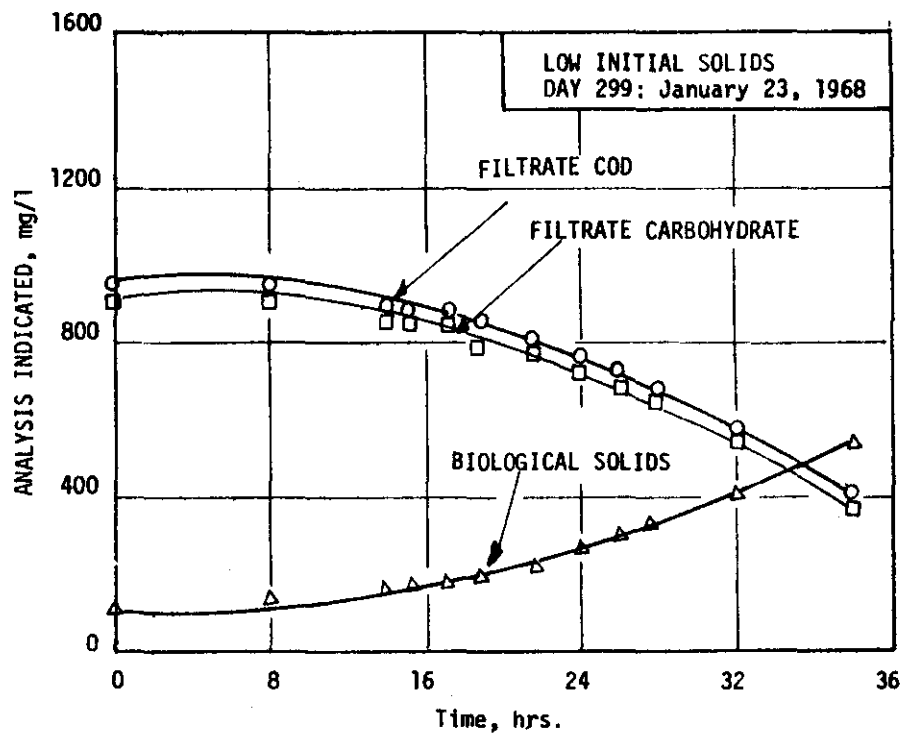
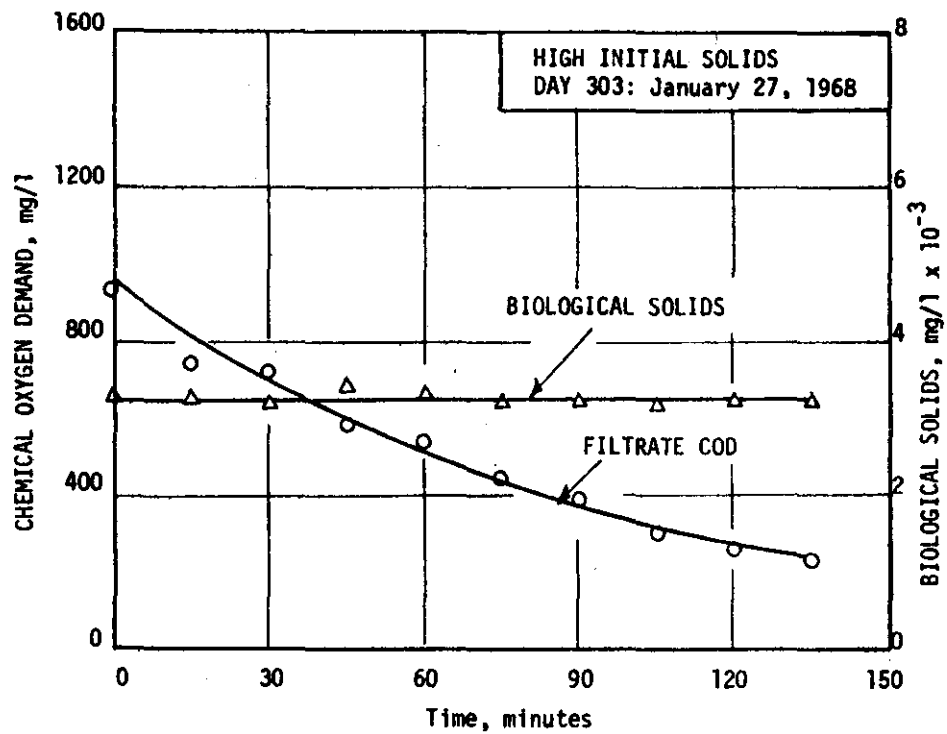


Figure 37 - Response of the extended aeration activated sludge to slug dosage of glucose after 299 days of operation.



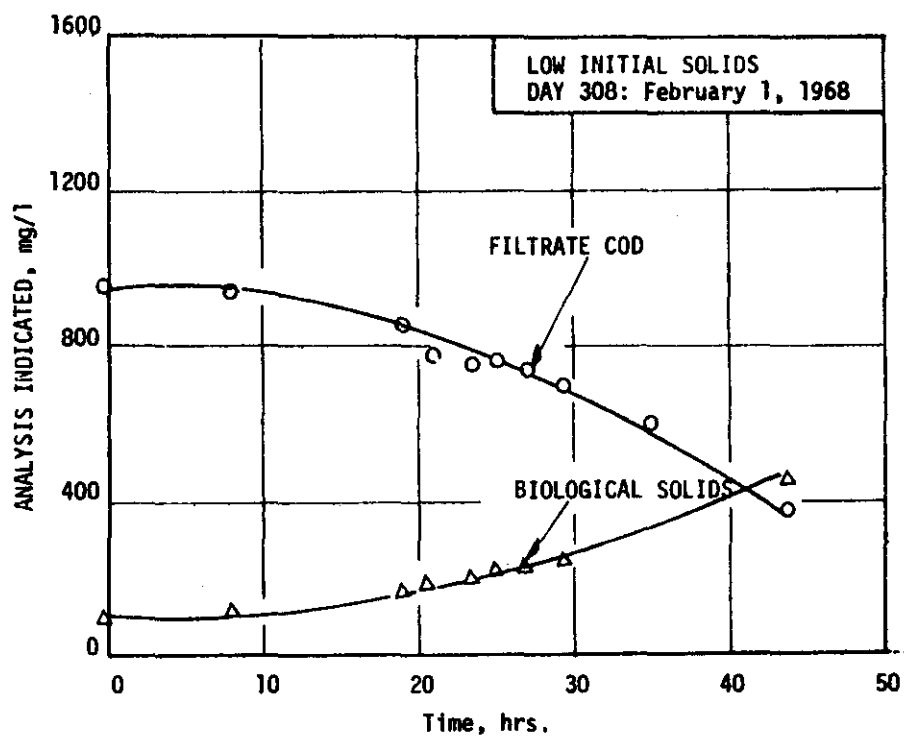
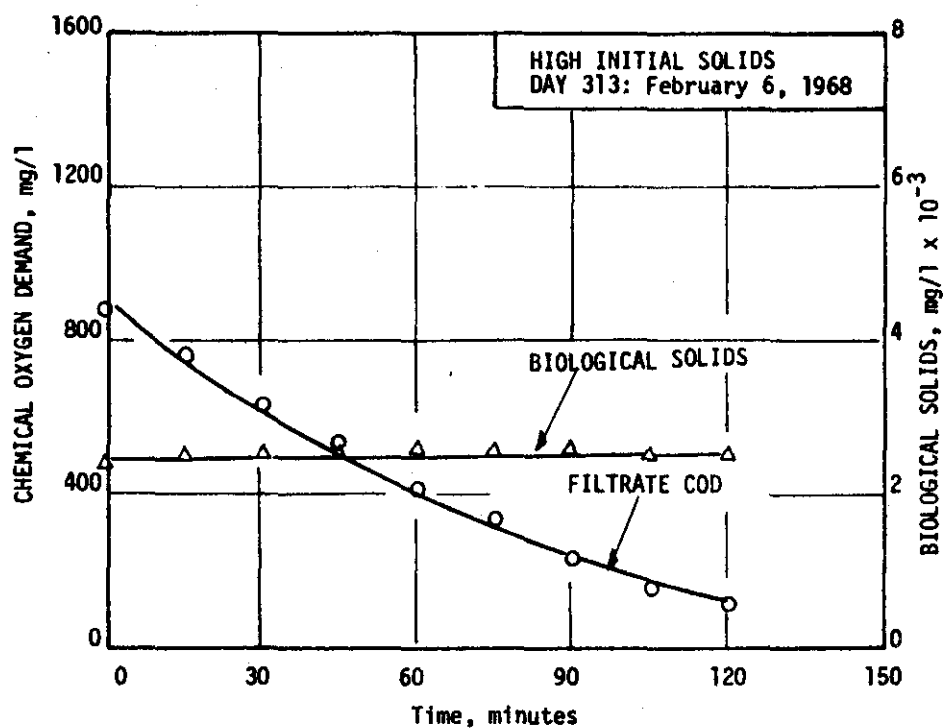


Figure 38 - Response of the extended aeration activated sludge to slug dosage of glucose after 308 days of operation.

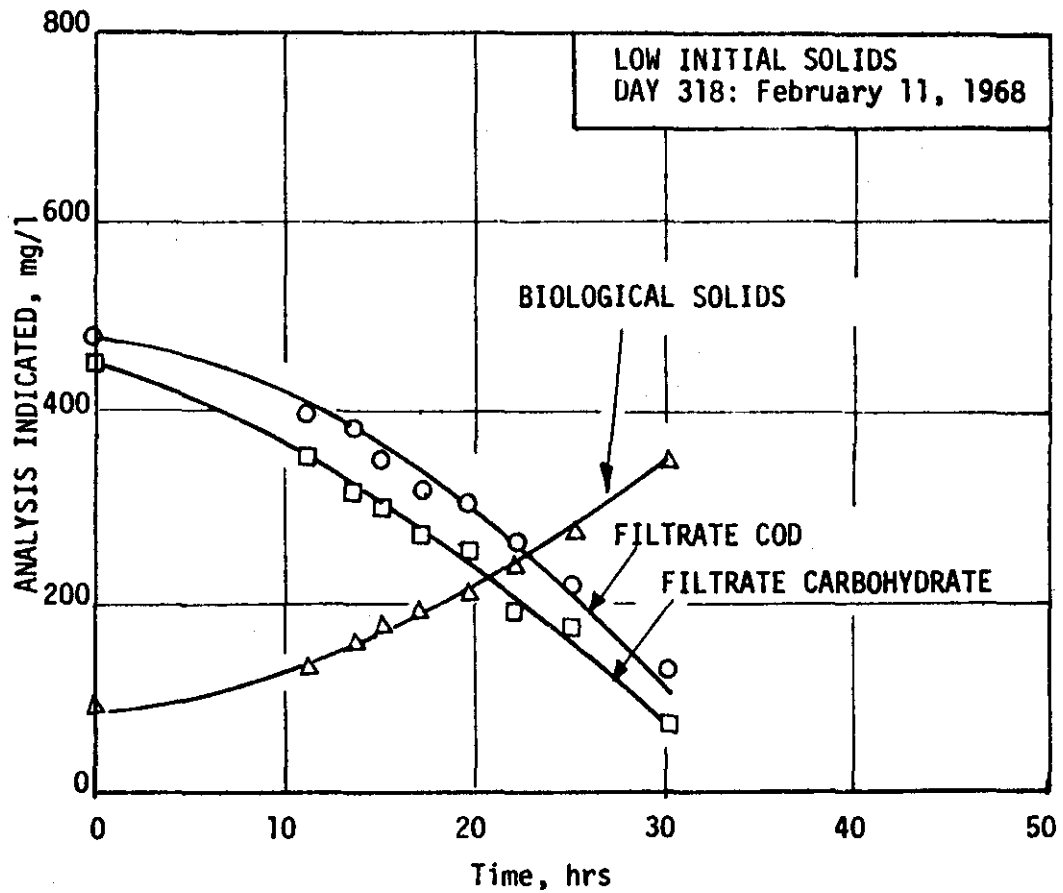


Figure 39 - Response of the extended aeration activated sludge to slug dosage of glucose after 318 days of operation.

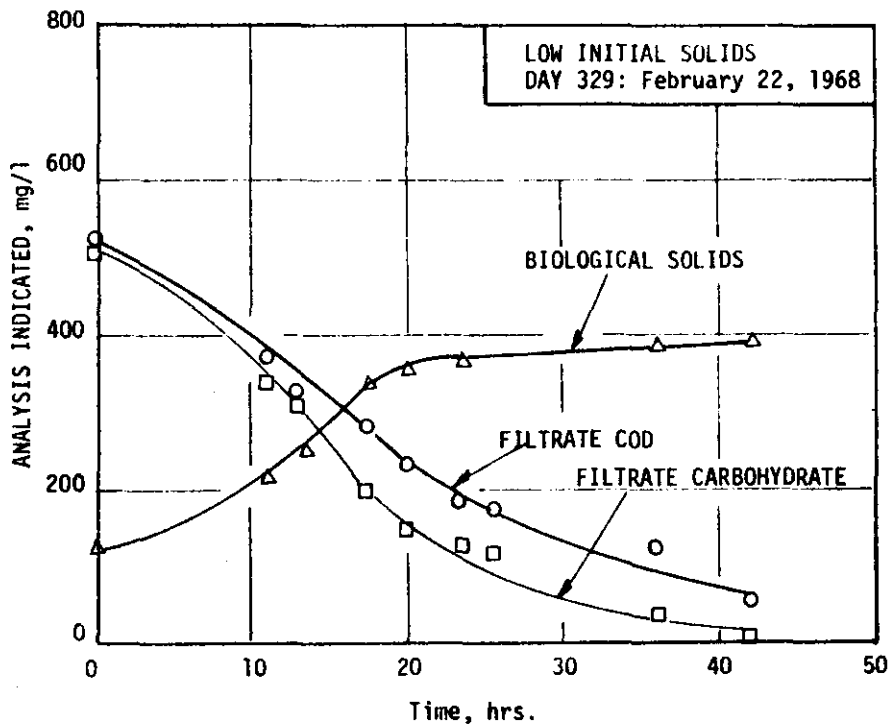
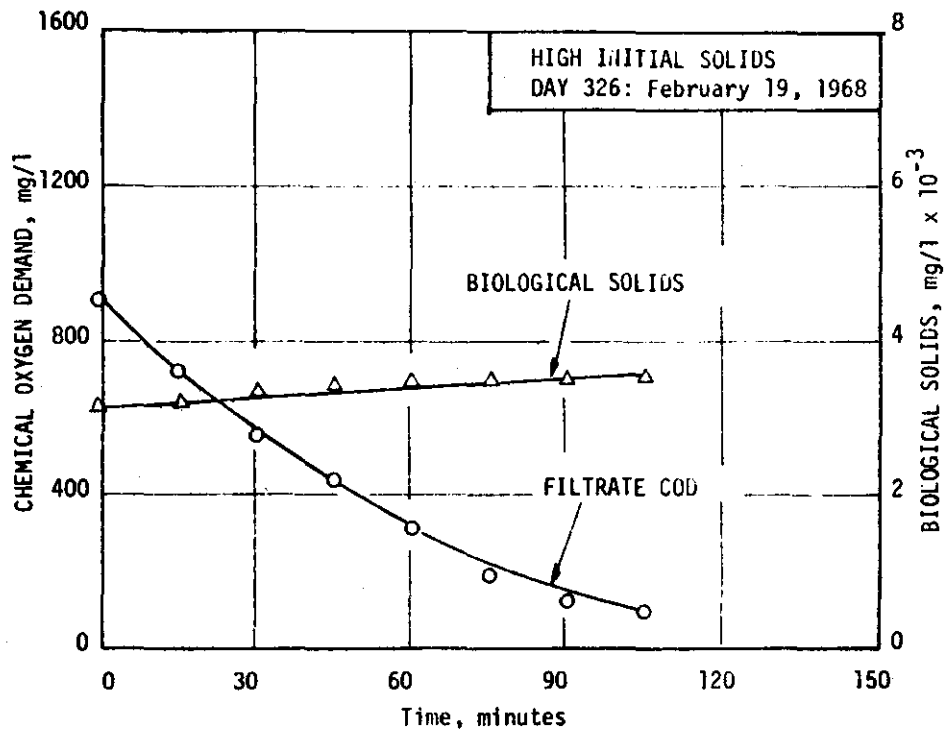


Figure 40 - Response of the extended aeration activated sludge to slug dosage of glucose after 326 days of operation.

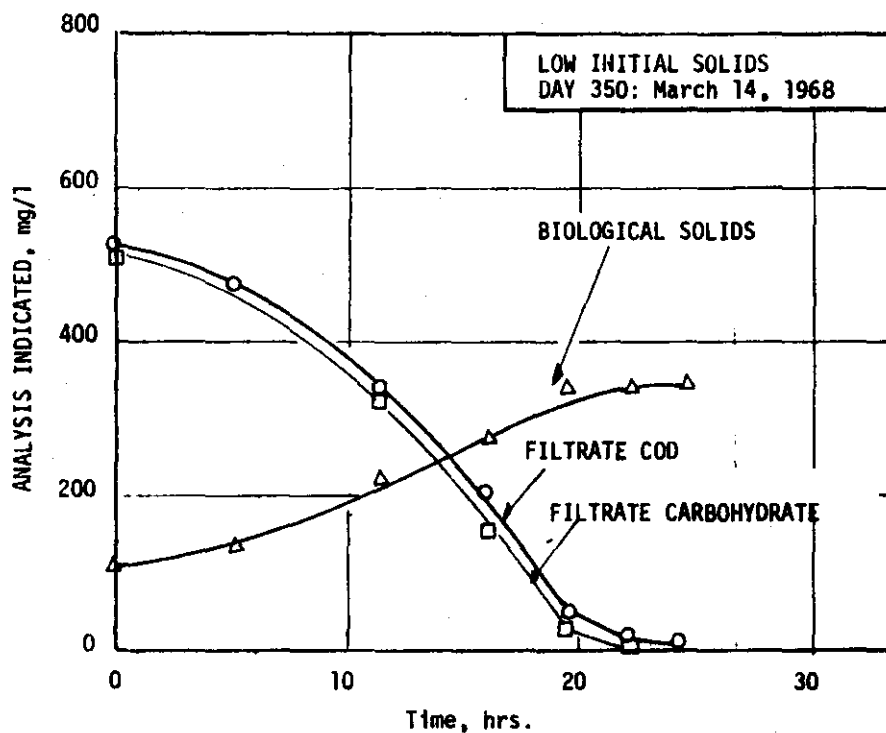
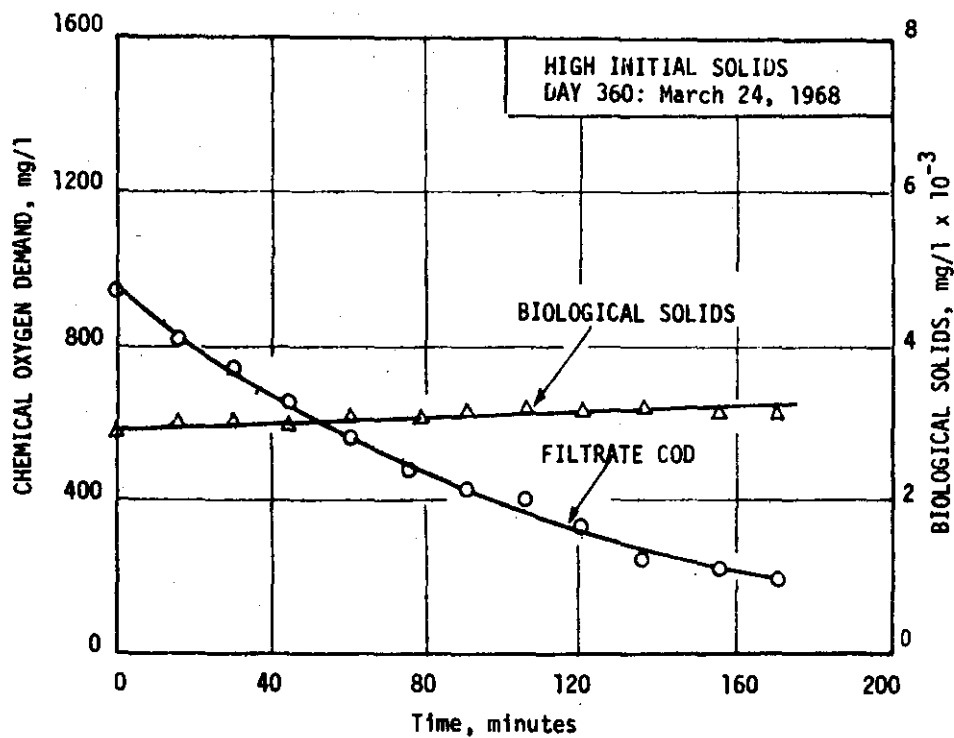


Figure 41 - Response of the extended aeration activated sludge to slug dosage of glucose after 350 days of operation.

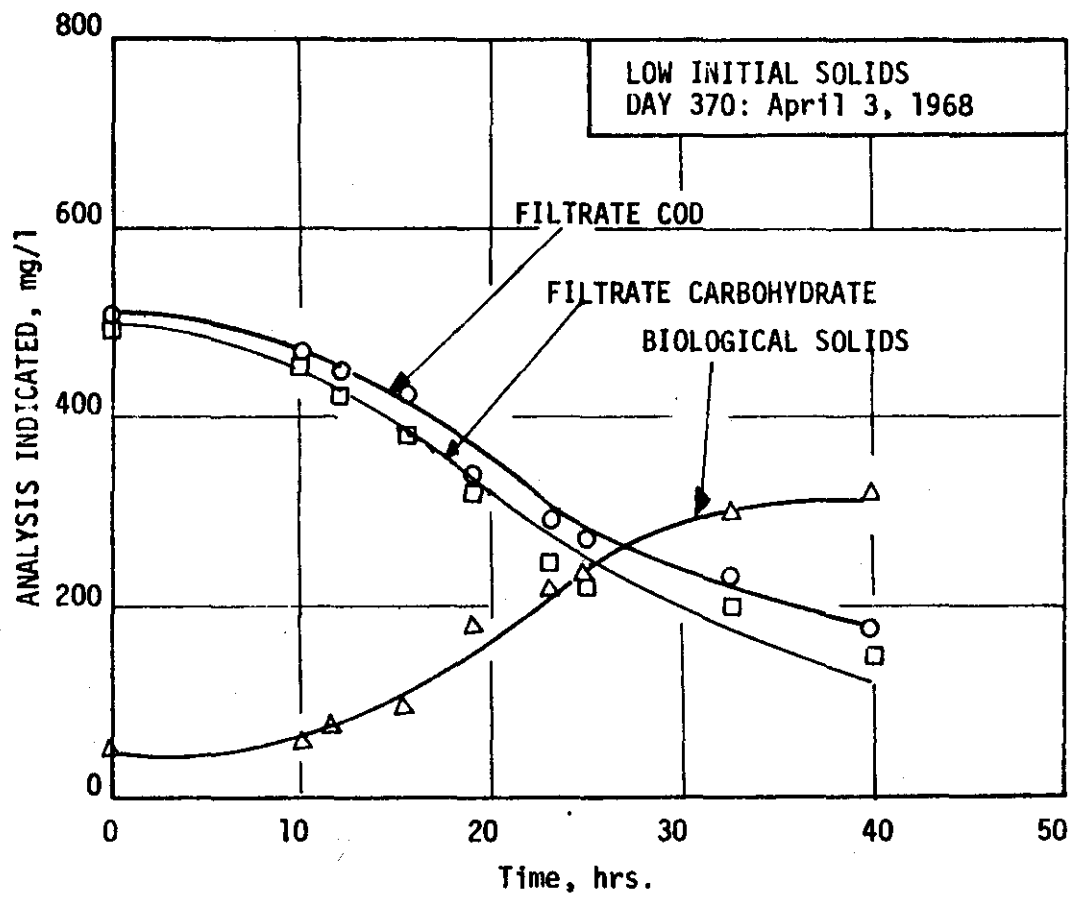


Figure 42. Response of the extended aeration activated sludge to slug dosage of glucose after 370 days of operation.

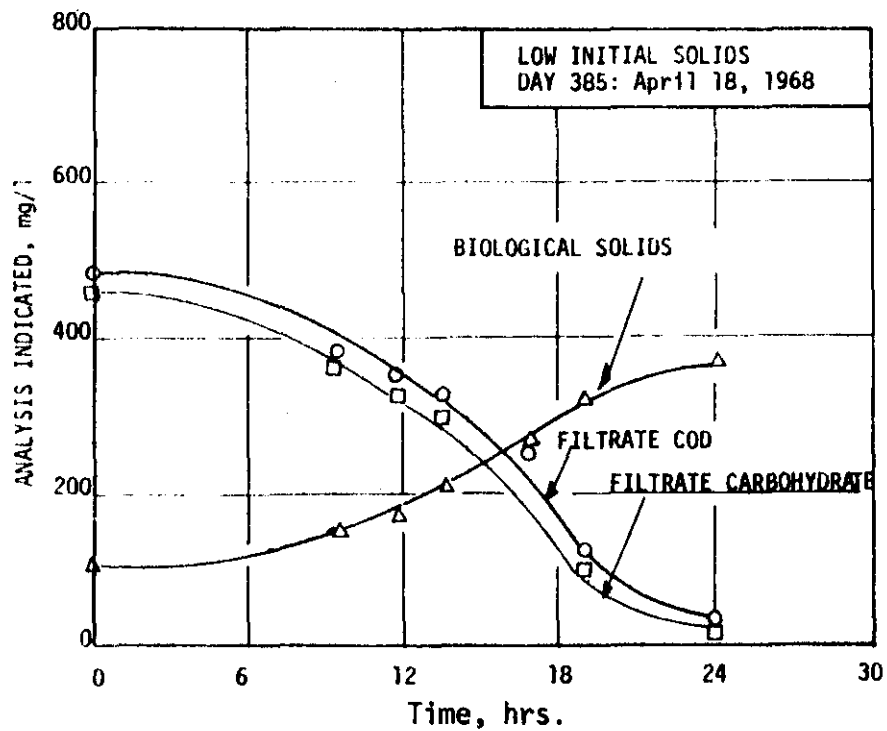
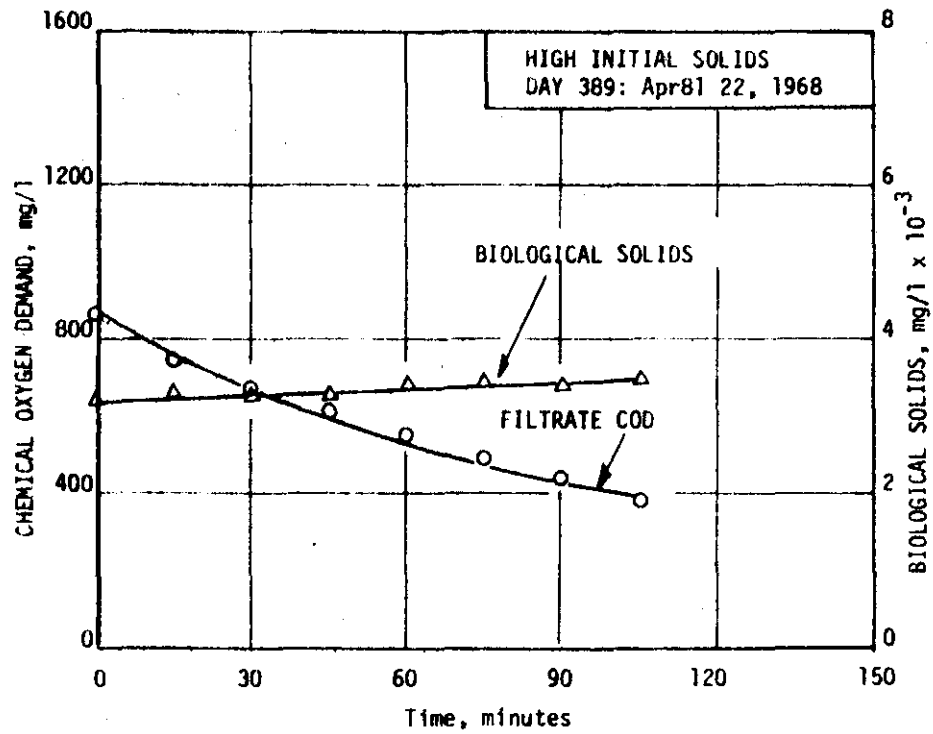


Figure 43 - Response of the extended aeration activated sludge to slug dosage of glucose after 385 days of operation.

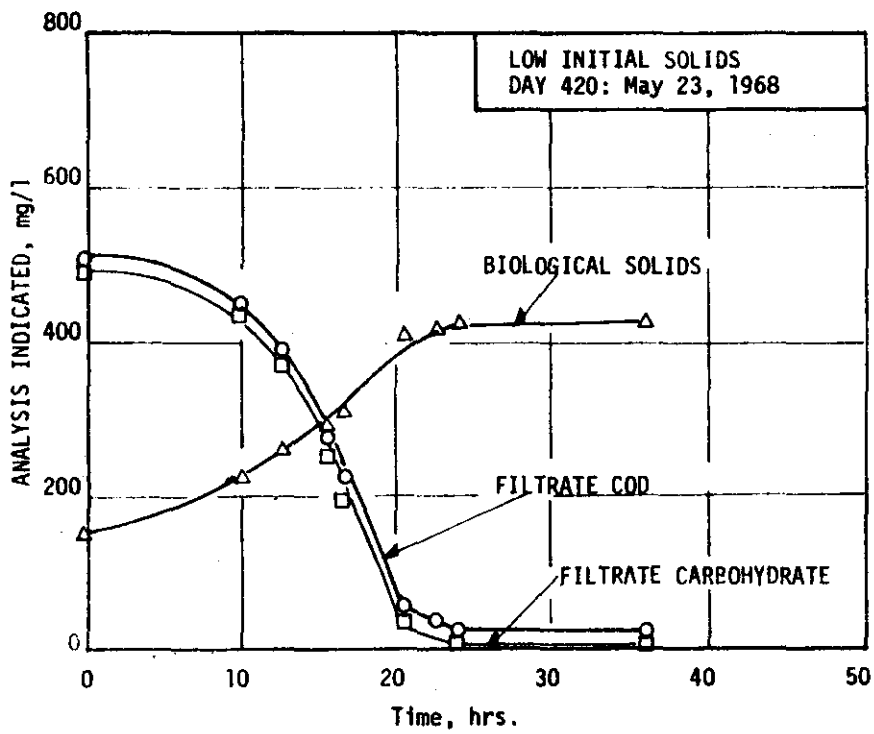
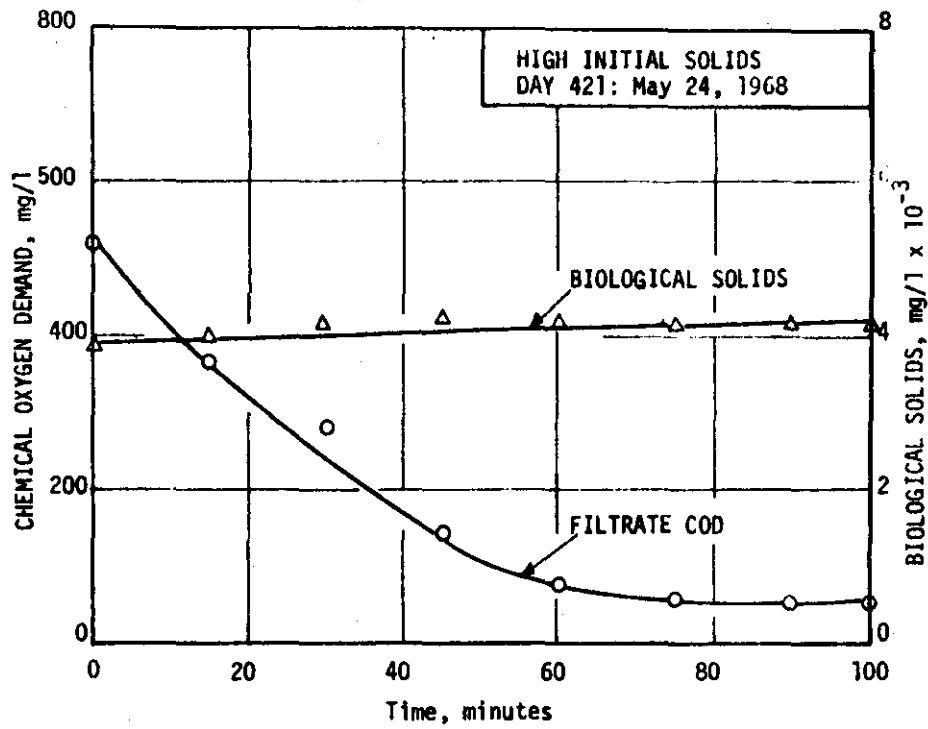


Figure 44 - Response of the extended aeration activated sludge to slug dosage of glucose after 420 days of operation.

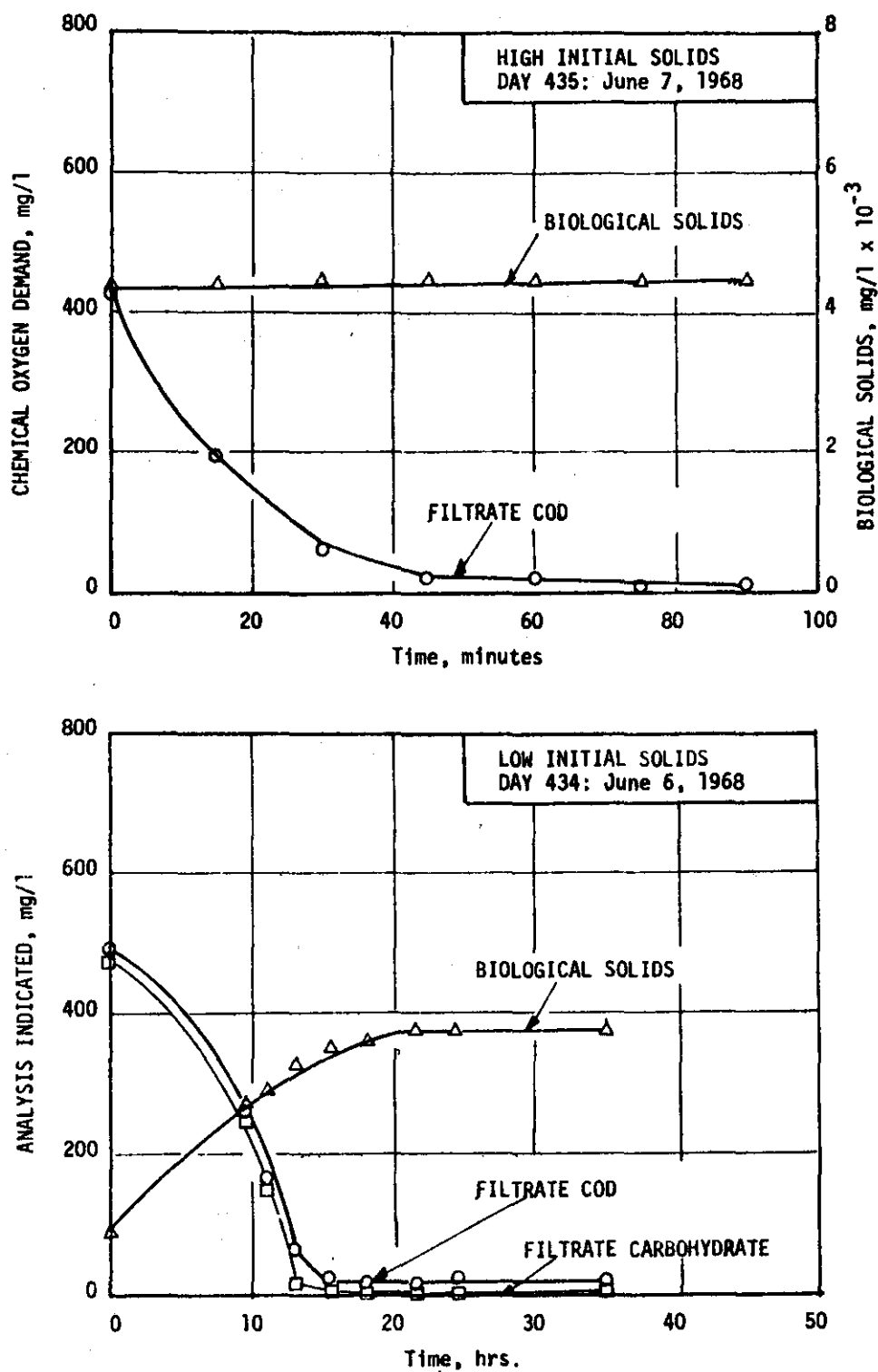


Figure 45 - Response of the extended aeration activated sludge to slug dosage of glucose after 434 days of operation.



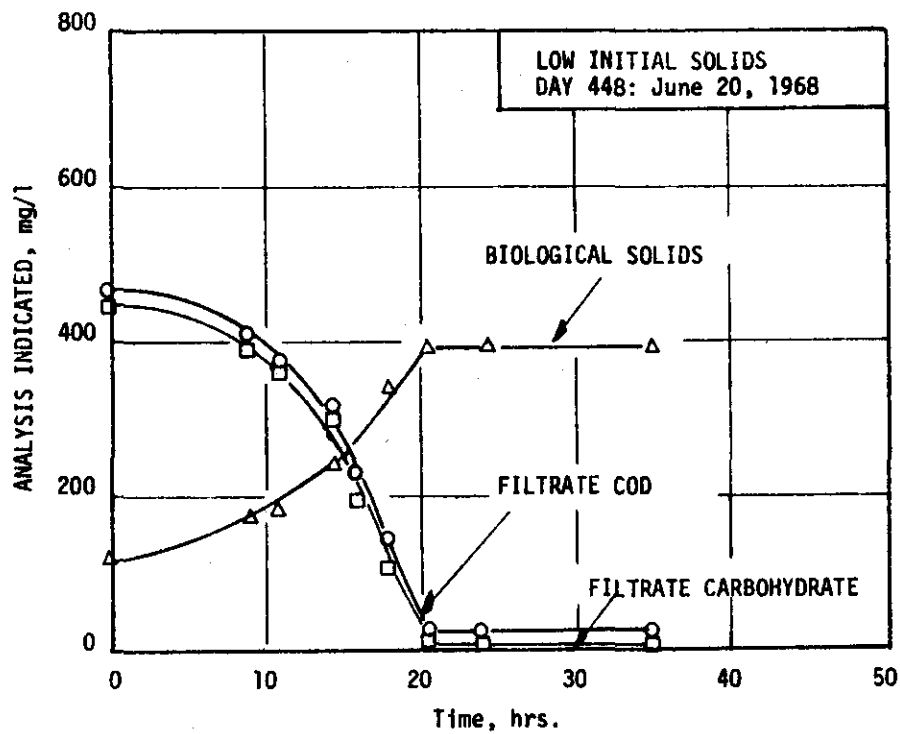
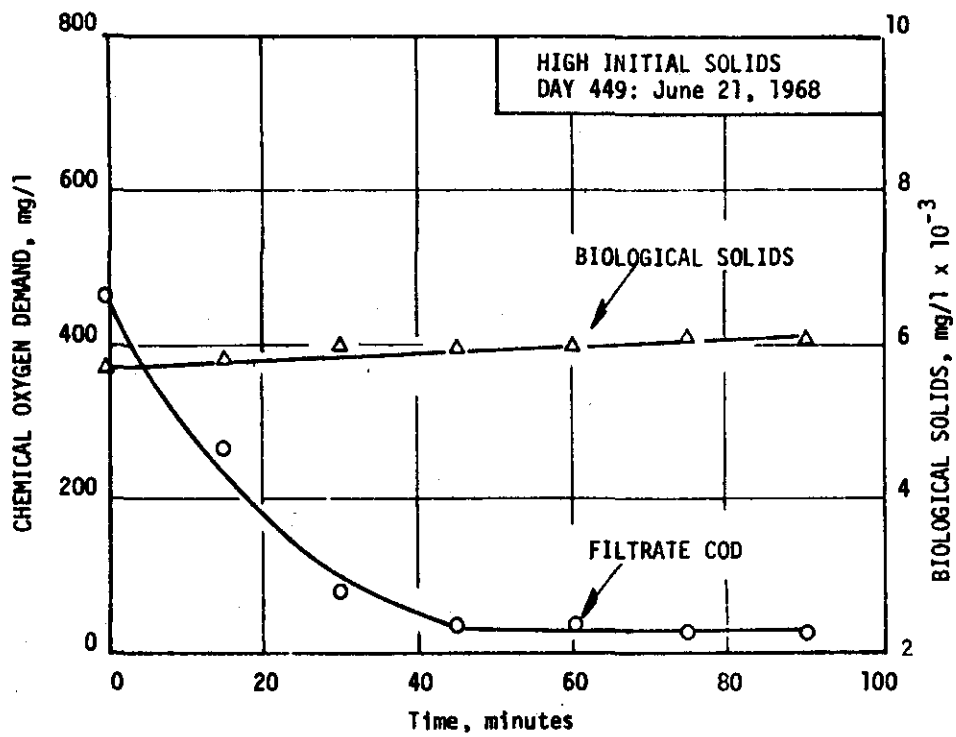


Figure 46 - Response of the extended aeration activated sludge to slug dosage of glucose after 448 days of operation.

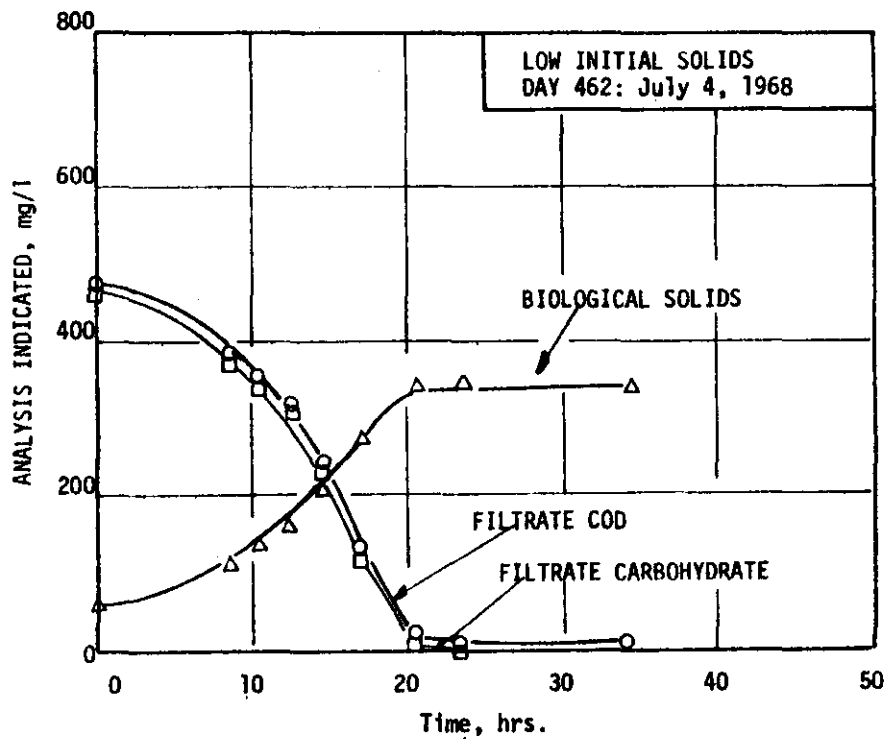
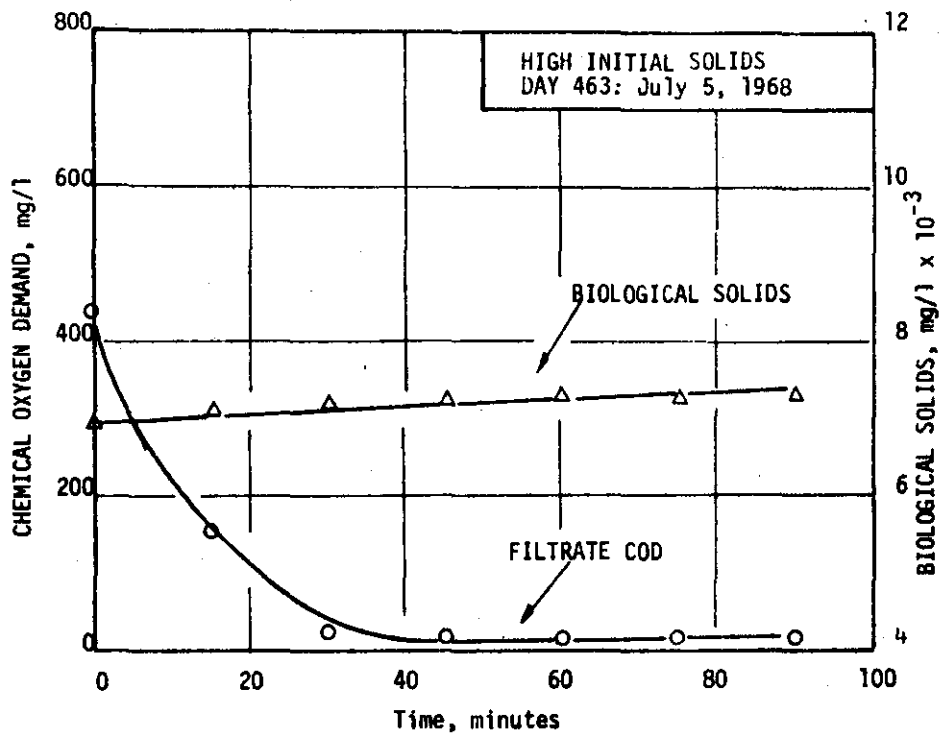


Figure 47 - Response of the extended aeration activated sludge to slug dosage of glucose after 462 days of operation.

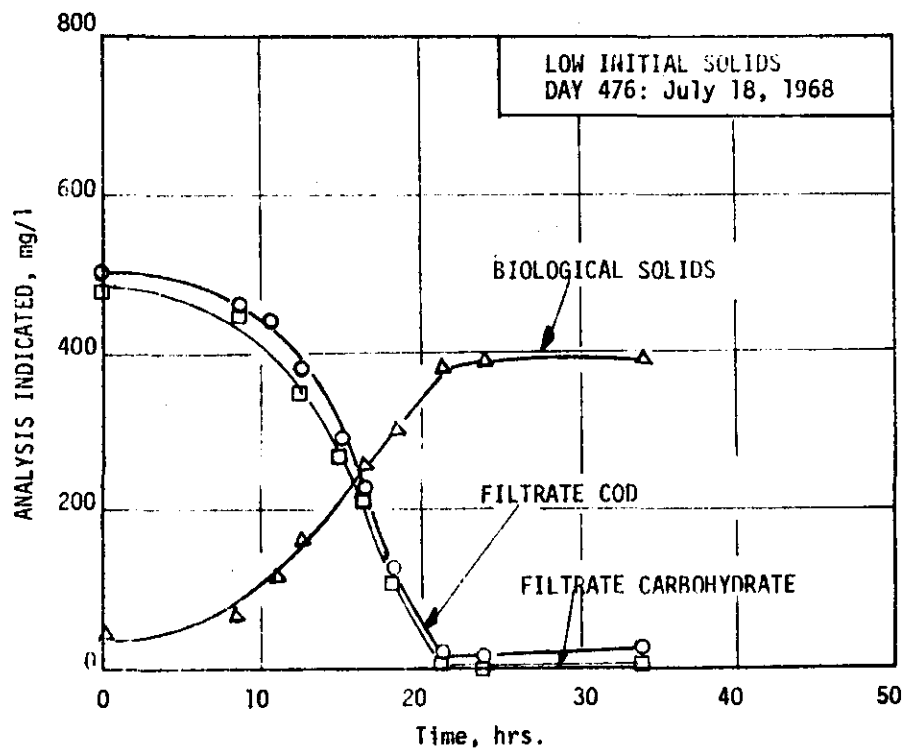
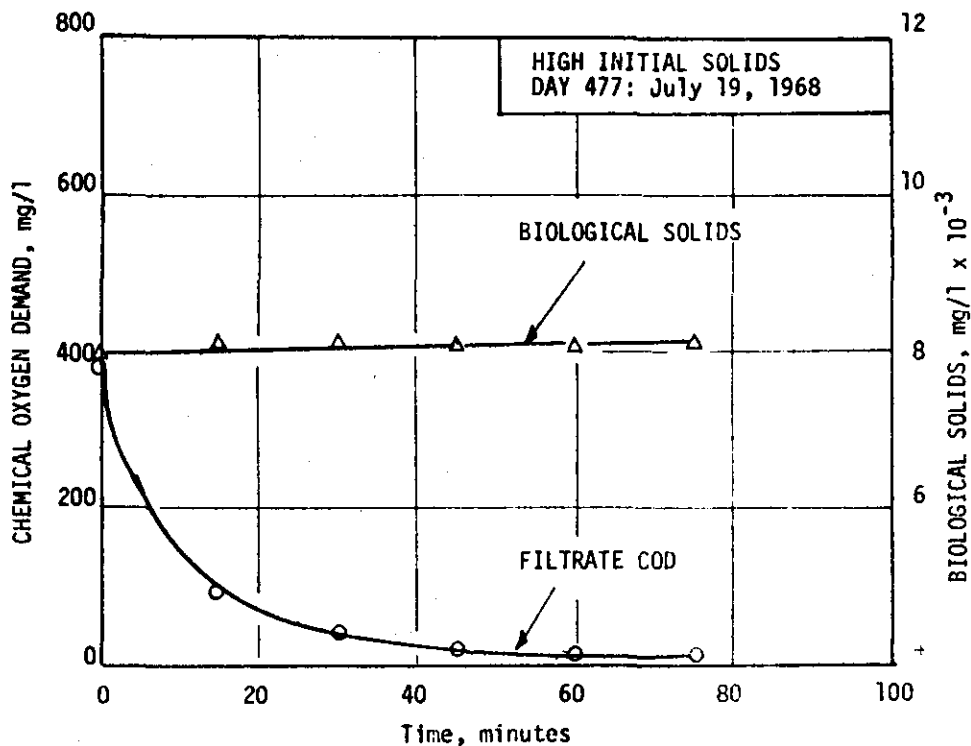


Figure 48 - Response of the extended aeration activated sludge to slug dosage of glucose after 476 days of operation.

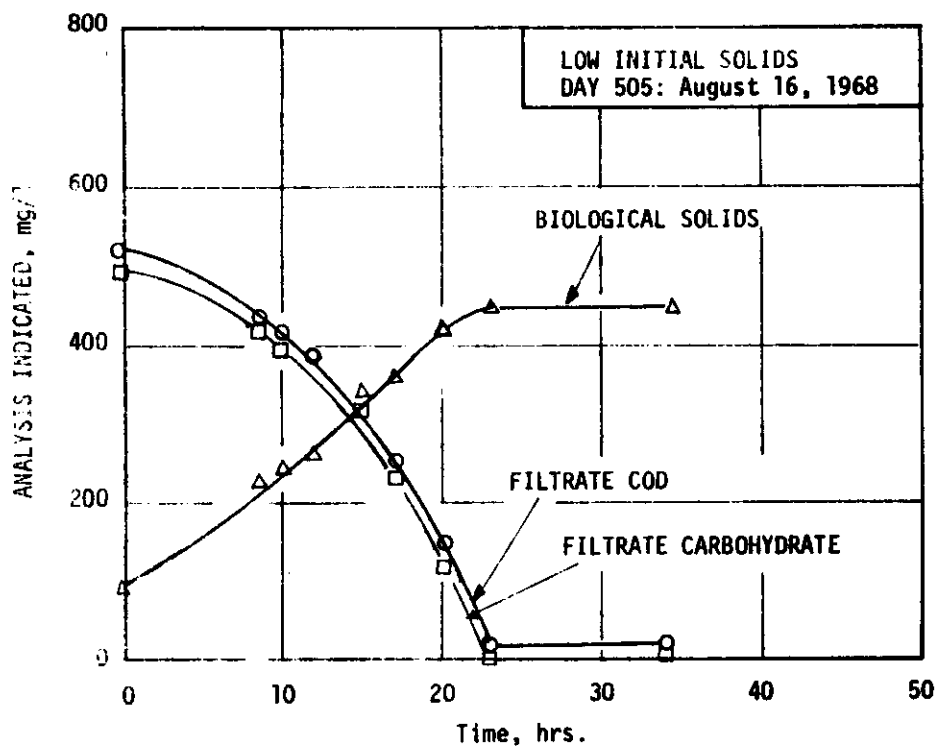
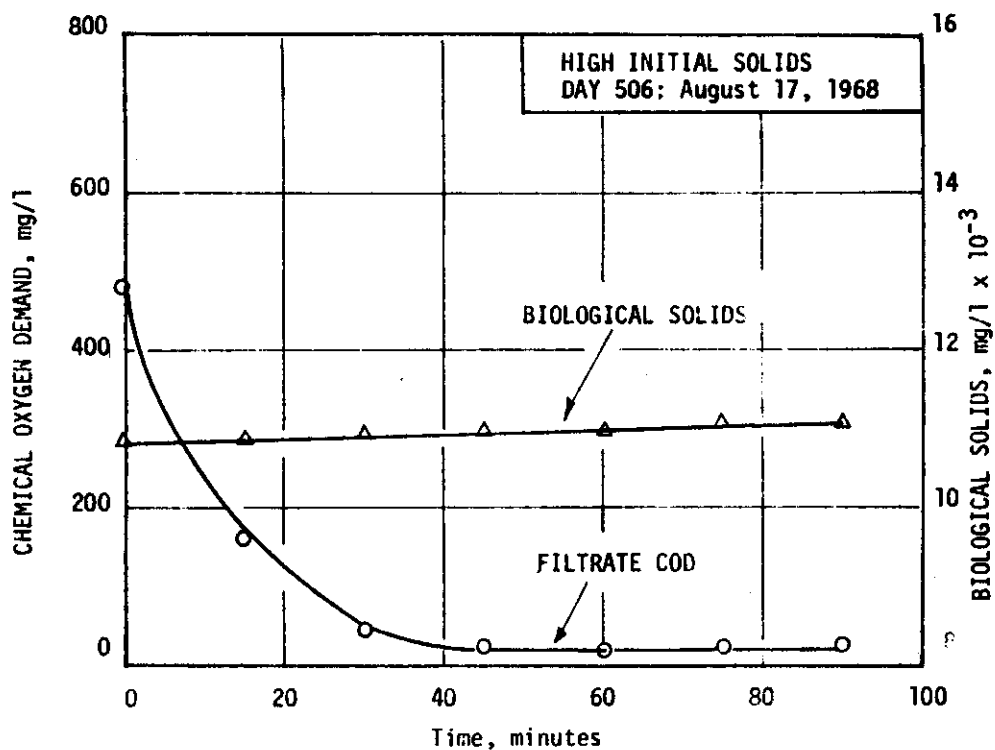


Figure 50 -- Response of the extended aeration activated sludge to slug dosage of glucose after 505 days of operation.

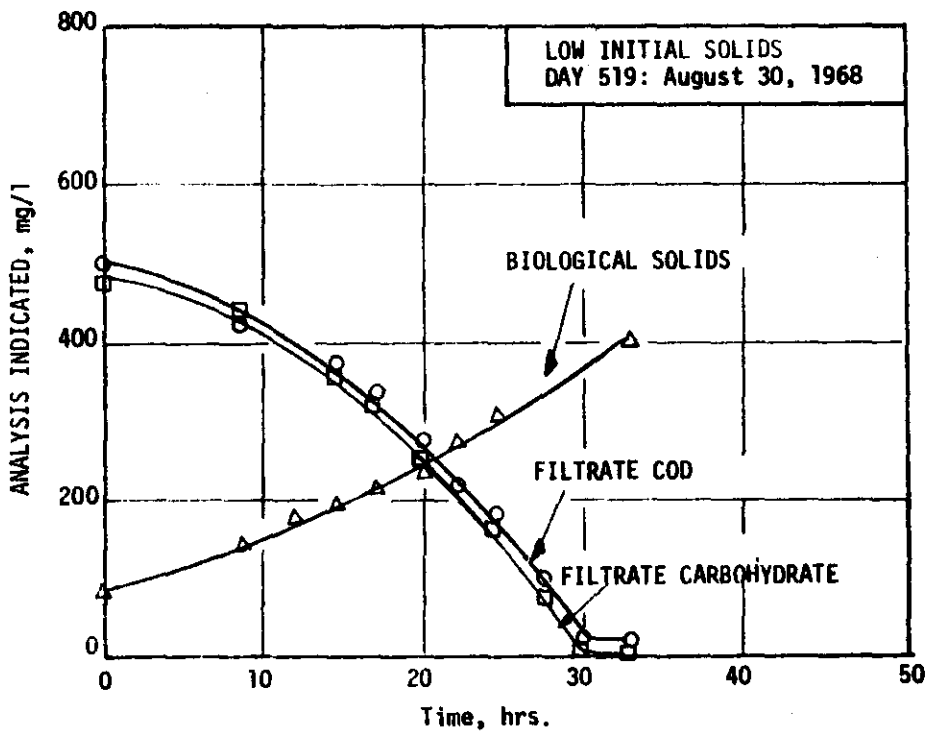
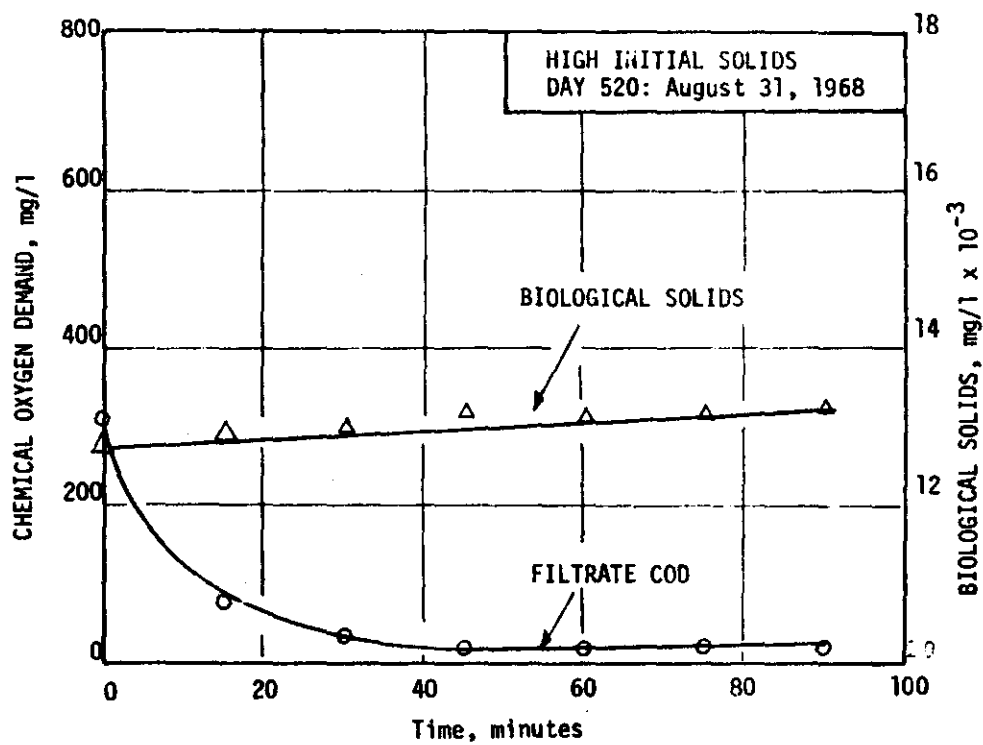


Figure 51 - Response of the extended aeration activated sludge to slug dosage of glucose after 519 days of operation.

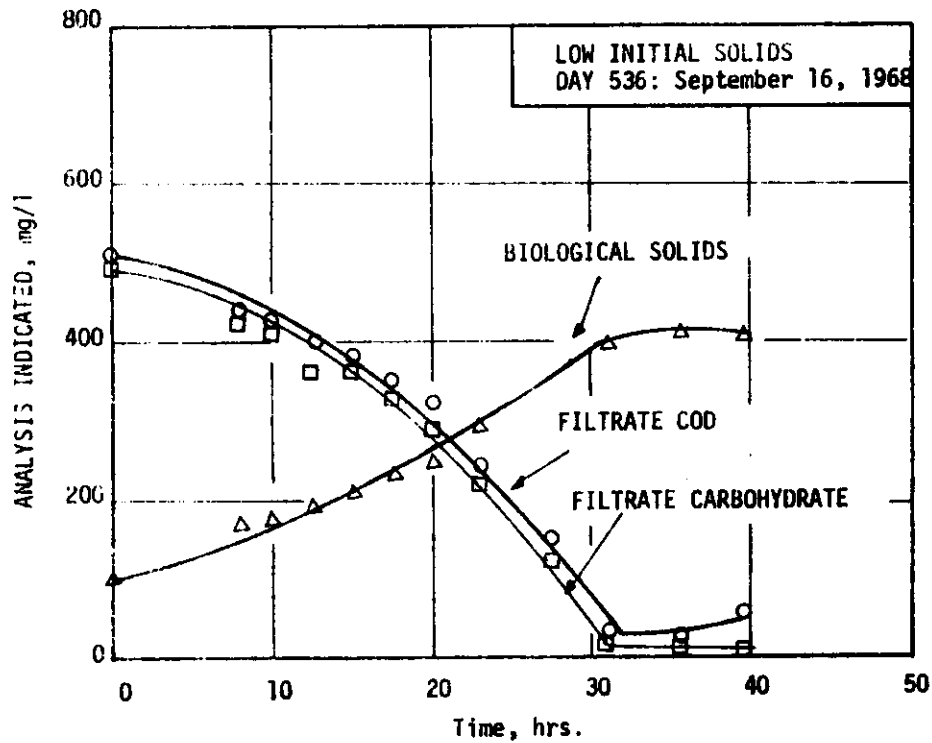
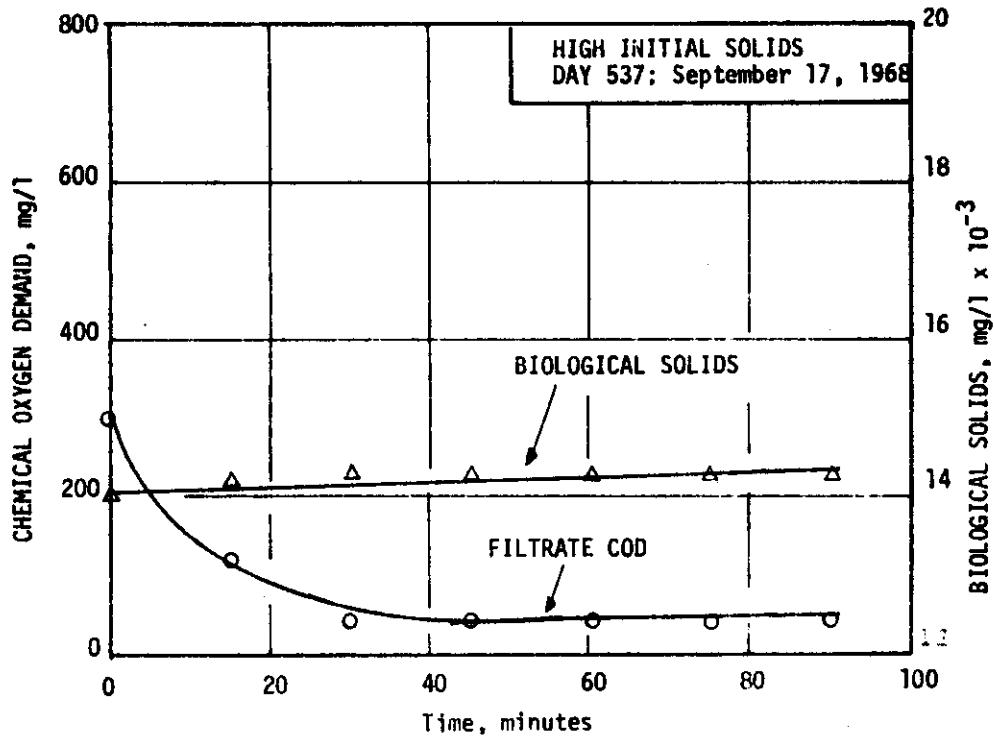


Figure 52 - Response of the extended aeration activated sludge to slug dosage of glucose after 536 days of operation.

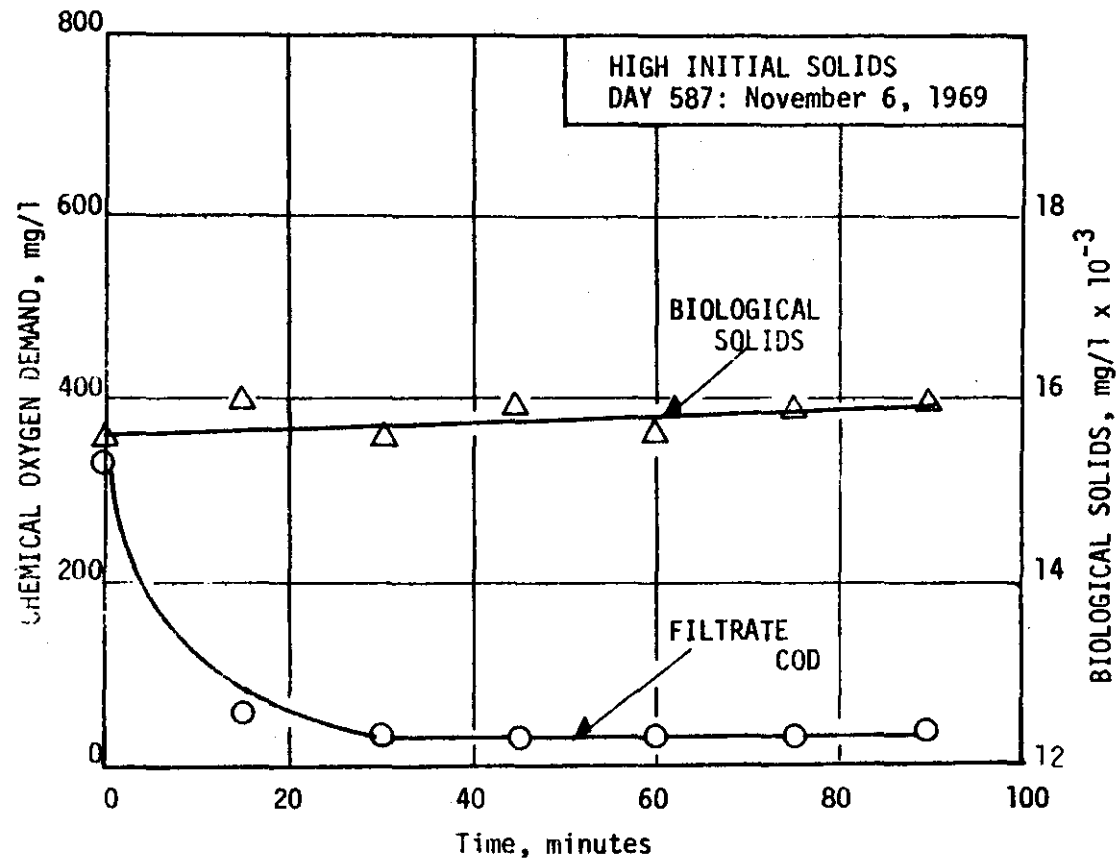


Figure 53 - Response of the extended aeration activated sludge to slug dosage of glucose after 587 days of operation.

rising. As mentioned previously, the mode of batch operation was such that the normally low residual COD observed during continuous flow operation could not escape from the unit during batch periods of operation. However, the results shown in Figure 32 would appear to indicate that at the time the run was made, some of the buildup in residual COD was caused by decreased ability of the cells to metabolize the substrate. Unfortunately, anthrone analyses were not made at this time and there was no way of estimating the nature of the residual COD. In other work (9) the residual COD was also high during batch operation of an extended aeration unit, and carbohydrate analyses run in conjunction with COD's showed that the residual COD was not due to the original substrate. Two additional high initial solids runs were made during the period of batch operation, one on day 229 (Figure 33), and one on day 243 (Figure 35). At the time these runs were made, the residual COD had levelled off at approximately 500 mg/l. It is seen in both Figures 33 and 35 that the amount of COD (500 mg/l glucose) which was fed was removed in less than 1.5 hours. On this basis it was concluded that the high residual COD in the unit was not an indication of loss of COD removal capability of the sludge, but was a result of accumulation of the small residual COD which normally would have been in the effluent during continuous flow operation but was prevented from escaping during batch operation. The same effect was noted in other studies (9).

In all runs made directly in the extended aeration unit (high initial solids) with the exception of the run on day 218, the COD was removed very rapidly, usually in less than two hours, and often in approximately thirty minutes. During periods of continuous flow



operation, the detention time in the aerator was 16 hours, and it was 23 hours during periods of batch operation. Thus the detention time was much in excess of that absolutely required. The low initial solids experiments indicate that the cells do not go into a log phase rapidly, and that the population is a rather slow-growing one. For many of the experiments at low initial solids concentration the course of substrate removal was determined by the anthrone test as well as COD analyses (see Figures 37 through 39, and Figures 39 through 52). It is seen that in general, both analyses give similar values, thus for these slower-growing cells there was no evidence for the accumulation of metabolic intermediates and/or endproducts.

### 3. $PO_4^{=}$ , $NH_3$ -N, $NO_2$ -N, and $NO_3$ -N in the Effluent (Filtrate)

The removal of phosphate and nitrogen by extended aeration plants or, on the other hand, the emission of these materials by such plants, is of increasing interest. It was therefore appropriate to gain some insights into this aspect. Such analyses were not run on a routine basis, but between days 351 and 664, thirty-one samples were examined. The results are shown in Table II. In the present study the  $PO_4$ -P and  $NH_3$ -N concentrations in the feed were 169 mg/l and 53 mg/l, respectively. It is interesting to note that at times the effluent  $PO_4$ -P concentration was higher than the influent concentration. This result may be attributed to release of phosphate by the cells. In general, small quantities of  $NO_3$ -N were noted. It would appear that the lengthy aeration period (16 hours) did not enhance excessive nitrification. Organic nitrogen was not run on the filtrate.

TABLE II  
 INORGANIC PHOSPHATE, PHOSPHORUS, AND NITROGEN CONTENTS IN THE  
 EFFLUENT (FILTRATE), mg/l

	Date	Age of Sludge* Days	PO <sub>4</sub> <sup>-3</sup>	P	NH <sub>3</sub> -N	NO <sub>3</sub> -N	NO <sub>2</sub> -N	Total Inorg. N
1968	3-15	351	384	125	25	10	< 0.1	35.0
	22	358	399	130	31.5	7.5	"	39.0
	4- 4	371	395	129	26	9.5	"	35.5
	10	377	425	138	25.5	6.3	"	31.8
	20	387	390	127	23	6.3	"	29.3
	29	396	565	184	26	4.3	"	30.3
	5- 8	405	555	180	26	2.8	"	28.8
	14	411	445	145	22	3.2	"	25.2
	23	420	445	145	26	3.2	"	29.2
	6- 3	431	445	145	30.5	3.2	"	33.7
	13	441	465	153	27.5	3.2	"	30.7
	21	449	-	-	25	3.2	"	28.2
	29	457	380	124	25	3.2	"	28.2
	7-12	470	540	176	25	3.2	"	28.2
	19	477	510	166	49	2.1	"	51.1
	26	484	420	137	25.5	3.2	"	28.7
	8- 4	493	395	129	25.5	3.1	"	28.6
	13	502	435	142	25.5	3.2	"	28.7
	24	513	460	150	25.6	2.2	"	27.8
	9- 1	521	465	153	25	3.2	"	28.2
	11	531	465	153	26.2	3.7	"	29.9
	24	544	800	260	37	8	"	45
	10- 9	559	400	130	28.8	1.9	"	51.2
	12	562	465	153	28.2	2.3	"	51.2
11- 3	584	800	260	33	3.4	"	36.4	
12	593	680	222	41	1.4	"	42.4	
23	604	600	195	40	2.5	"	42.5	
12- 7	618	620	202	36	3.4	"	39.4	
24	635	520	170	42.8	8.3	"	51.1	
1969	1-10	652	655	214	40	7.2	"	47.2
	22	664	265	90	16.5	4	"	20.5

\* Time in days since startup of the extended aeration unit.

Note: Influent PO<sub>4</sub><sup>-3</sup> concentration = 518 mg/l  
 Influent PO<sub>4</sub> - P concentration = 169 mg/l  
 Influent NH<sub>3</sub>-N concentration = 53 mg/l.

## CHAPTER V

### DISCUSSION

The extended aeration process for the treatment of organic waste has received considerable attention by researchers during the past two decades. The majority of workers have concluded that biological solids concentration will continue to build up and cause the ultimate failure of such systems. Therefore, it has been generally concluded that sludge wasting is necessary to avoid the final failure (i.e., prevent loss of ability to metabolize substrate). The present study was originally designed to determine the time required to produce such failure in a system in which no sludge was wasted. The results after nearly two years of operation of an extended aeration system with no sludge wasting indicate that the system has not yet lost its biochemical efficiency. The effluent quality compares favorably with that of the more conventional (sludge wasting) activated sludge plants. The results also show that sludge does not accumulate steadily, but there are periodic cycles of decreasing solids concentration followed by succeeding periods of solids accumulation. Furthermore, during the periods of decreasing solids concentration, no gross leakage of COD in the effluent has been observed. Since biological solids were neither intentionally nor accidentally wasted and there were no external stresses, such as changes in pH, temperature, etc., to which the

decreasing cycles can be ascribed, it seems reasonable to conclude that the periodic relief of solids accumulation is brought about due to natural phenomena, and for causation of this effect it is necessary to examine the nature of the ecosystem in an extended aeration activated sludge.

Activated sludge consists of bacteria, protozoa, rotifers and, sometimes, nematodes. The bacteria are generally considered to be the most important group of microorganisms, for they are the ones which metabolize most of the soluble organic matter in the waste water. The use of an extended aeration was first suggested on the surmise that during endogenous metabolism of the bacteria they would autodigest their biomass. It is well known that endogenous respiration can cause the reduction of cell mass, but this does not indicate that any species of microorganism can totally oxidize itself. However, it is known that species of microorganisms undergo complete or nearly complete autolysis after attaining a maximum growth level (15)(16), thus the cellular materials can become available as food for the other species. If no other species were present, the system would fail. Fortunately, activated sludge represents a mixed population system, and other species are present. Also, the various microorganisms can produce enzymes which can cause the partial lysis or complete dissolution of other cells; thus, it is not necessary to rely on autolysis for relief of solids accumulation (16)(17)(18)(19). Another way a decrease in biological solids concentration can be brought about is through bacteriophage-induced lysis. Fortunately, this kind of infection, if it occurred in the activated sludge system, would not affect all species in the ecosystem, since specific bacteriophage infect specific species,

thus relief of sludge accumulation but not total killing can be expected. All of the above mechanisms can convert cell material into exogenous substrate. In order for it to be used, it is necessary that species which possess the metabolic capability to utilize it as a substrate be present, or else the quality of the effluent will deteriorate. In the present study such species were apparently present, since the effluent COD did not rise appreciably during cyclic decrease in biological solids concentration. It cannot be expected that cell disruption or dissolution will stay in balance with cell synthesis thus allowing the system to operate at some equilibrium solids concentration. Attainment of constant biological solids concentration would require a precise balance between the specific natural lytic agents and species which could metabolize the various cell components which are made available as substrate. Such a situation might exist at times, but it cannot be expected to be the normal state of the system. One must expect an ever-changing population to exist in an activated sludge. In addition, an extended aeration system is a low organic loading system, and the cells exist under starvation conditions. Ability to compete for the limited supply of substrate is a principal factor in determining the prominence of microbial species in environments in which the demand for the substrate greatly exceeds the supply. Since the nature of the cell constituents can be expected to change, so, too, can the predominant bacterial species. In addition to bacterial interactions, predatory and parasitic relationships are operative in natural ecosystems. The action of protozoa feeding upon bacteria, the action of myxobacteria and myxomycetes upon bacteria, and the parasitism of one fungal species by another can be expected to be operative in activated sludge systems

(especially in the highly competitive environment of the extended aeration activated sludge system). All of these factors militate against development of stable or equilibric conditions with respect to biological solids concentration in an extended aeration system. The time required to complete a cycle of net accumulation and net decrease of sludge cannot be predicted, nor can the periodicity of such cycles, but the results of this study to date attest to the fact that such cycles do exist. It is worthy of note that the data of Washington, et al. (20) also suggests that in the total oxidation system cyclic reduction in biological solids accumulation can occur.

Results of recent experiments with heterogeneous populations have indicated that the soluble cell fraction, released after mechanical breakage of cell walls, provides an excellent substrate for microbial growth (21). In other recent work employing heterogeneous populations, systems have been observed wherein essentially total oxidation of sludge synthesized in the log and declining growth phase has occurred in a subsequent prolonged endogenous phase (22)(23).

During the present study there were times when the sludge did not settle very well. However, the settling problem was no greater than that often observed in normal activated sludge processes (sludge wasting systems). When solids built up to very high levels, settling problems can be anticipated. There are various engineering expedients which might be employed to alleviate the sludge settleability problem during periods of extremely high solids concentration (10). Thus there is reason to believe that the settling problem is not an insurmountable one, and can eventually be solved.

The results of this experimentation do not contribute much to

remove the stigma of theoretical or mechanistic "unsoundness" which has surrounded the process. However, the results should not be construed as a guarantee that the process will not or cannot fail. The results may be interpreted as a definite indication that the process is not theoretically unsound. Its successful operation depends upon developing in the population (either naturally or possibly through bioengineering procedures) the unique combination of agents to disrupt or dissolve an excess portion of the population, and those agents (microbes) which can metabolize the substrates which have been made available. There may be long periods when the required combination of organisms is not present. During such times, sludge can be expected to accumulate. If the period is long enough and cells which can in essence feed on other members of the population do not develop, the various cell components may indeed be considered "biologically inert." They are inert until cells develop which can use them. The results to date indicate that such species will develop. Even if the systems failed now, after two years of successful operation, it would still have to be adjudged a successful one. In the field, if extended aeration systems had to be restarted after two years, it would still provide a satisfactory and economical engineering expedient in which secondary treatment was accomplished without the expense of separate sludge digestion.

## CHAPTER VI

### CONCLUSIONS

On the basis of this work, the following conclusions seem warranted:

1. In an extended aeration activated sludge process, with all solids returned to the aeration tank, the biological solids concentrations will not necessarily keep building up. The biological solids concentration decreased periodically, and causation for this effect can be attributed only to natural phenomena characteristic of the heterogeneous population comprising the activated sludge.

2. After nearly two years of operation, the system has not lost its ability to remove a reasonably high organic loading (500 mg/l glucose, retention time 16 hours). Furthermore the substrate removal studies indicate the sludge capability for removing the substrate exceeds the loading which is being applied.

3. In accordance with conclusions one and two, it seems apparent that it can no longer be considered that total oxidation is theoretically impossible. On the basis of the present results, it seems reasonable to make a tentative recommendation for wider application of the extended aeration process. It should prove a useful method for treatment of soluble organic industrial wastes.



## CHAPTER VII

### FUTURE WORK

The present results warrant a re-examination of the extended aeration process, since they have done much to remove the stigma of theoretical unsoundness which has been associated with this process since it was first proposed. The major work which should be undertaken next is the investigation of the natural causes by which periodic relief of solids accumulation is brought about. Work designed to assess the operation of these ecological mechanisms may not lead to engineering predictions of the extent and periodicity of the decreasing solids cycles, but may provide further proof of the theoretical soundness of the process. Work should also be undertaken to determine if various bioengineering expedients, i.e., external controls designed to enhance lysis, changes in predominance, etc., can be employed to control the solids level in the system. Work on ways and means to enhance settling, or work on other possible means of separating the solids from the effluent is also warranted.

## BIBLIOGRAPHY

1. Dawes, E. A., and Ribbons, D. W., "The Endogenous Metabolism of Microorganisms." Annual Review of Microbiology, 16, 241-264 (1962).
2. Porges, N., Jasewicz, L., and Hoover, S. R., "Aerobic Treatment of Dairy Wastes." Jour. Applied Microbiology, 1, 262-270 (1953).
3. Symons, J. M., and McKinney, R. E., "The Biochemistry of Nitrogen in the Synthesis of Activated Sludge." Sewage and Industrial Wastes, 30, 874-890 (1958).
4. Washington, D. R., and Symons, J. M., "Volatile Sludge Accumulation in Activated Sludge Systems." Journal Water Pollution Control Federation, 34, 767-789 (1962).
5. Busch, A. W., and Myrick, N., "Food Population Equilibria in Bench Scale Biooxidation Units." Sewage and Industrial Wastes, 32, 949-959 (1960).
6. Kountz, R. R., and Fourney, C., Jr., "Metabolic Energy Balance in a Total Oxidation Activated Sludge System." Sewage and Industrial Wastes, 31, 819-826 (1959).
7. McCarty, P. L., and Brodersen, C. F., "Theory of Extended Aeration Activated Sludge." Journal Water Pollution Control Federation, 34, 1095-1102 (1962).
8. Westrick, J. J., and Morand, J. M., and Eye, D. J., "A Field Study of the Extended Aeration Process." Proceedings 21st Industrial Waste Conference, Purdue University, Ext. Ser. 121, 424-439 (1966).
9. Ramanathan, M. Gaudy, A. F., Jr., and Ragthaidee, W., "Responses of Extended Aeration Activated Sludge to Quantitative Shock Loads." Proceedings 19th Oklahoma Industrial Wastes and Pollution Control Conference, Stillwater, Oklahoma (1968). (Submitted for publication in Journal Water Pollution Control Federation.)
10. Gaudy, A. F., Jr., Ramanathan, M., Yang, P. Y., and DeGeare, T. V., "Studies on the Operational Stability of the Extended Aeration Process." Proceedings 20th Oklahoma Industrial Wastes and Pollution Control Conference, Stillwater, Oklahoma (1969).
11. Standard Methods for the Examination of Water and Waste Water. 12th Ed., American Public Health Association, New York (1965).

12. Gaudy, A. F., Jr., "Colorimetric Determination of Protein and Carbohydrate." Industrial Water and Wastes, 7, 17-22 (1962).
13. Ramanathan, M., Gaudy, A. F., Jr., and Cook, E. E., "Selected Analytical Methods for Research in Water Pollution Control." Publication M-1, Center for Water Research in Engineering, Oklahoma State University.
14. "Precision Galvanic Cell Oxygen Analyzer." Publication TS-68850, Precision Scientific Company, Chicago, Ill. (1961).
15. Moustafa, H. H., and Collins, E. B., "Molar Growth Yields of Certain Lactic Acid Bacteria as Influenced by Autolysis." Journal Bacteriol., 96, 117-125 (1968).
16. Stolp, H., "Bacteriolysis." Annual Review of Microbiology, 19, 79-104 (1965).
17. Ensign, J. C., and Wolfe, T. S., "Lysis of Bacterial Cell Walls by Enzyme Isolated from Myxobacter." Journal Bacteriol., 90, 395-402 (1965).
18. Burke, M. E., and Pattee, P. A., "Purification and Characterization of a Staphylolytic Enzyme from Pseudomonas aeruginosa." Journal Bacteriol., 93, 860-865 (1967).
19. Whitside, T. L., and Corpe, W. A., "Extracellular Enzymes Produced by a Pseudomonas sp. and Their Effect on Cell Envelopes of Chromobacterium violaceum." Canadian Journal of Microbiol., 15, 81-92 (1969).
20. Washington, D. R., Hetling, L. J., and Rao, S. S., "Long-term Adaptation of Activated Sludge Organisms to Accumulated Sludge Mass." Proceedings 19th Industrial Waste Conference, Purdue University, Ext. Ser. 117, 655-666 (1965).
21. Obayashi, A. W., and Gaudy, A. F., Jr., Unpublished Data (1969).
22. Thabaraj, G. J., and Gaudy, A. F., Jr., "Effect of Initial Biological Solids Concentration and Nitrogen Supply on Metabolic Patterns During Substrate Removal and Endogenous Metabolism." (In preparation, 1969).
23. Thabaraj, G. J., and Gaudy, A. F., Jr., Unpublished Data (1969).

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