# STUDIES ON THE RESPONSE OF A HYDROLYTICALLY-ASSISTED EXTENDED AERATION ACTIVATED SLUDGE SYSTEM AND A COMPLETELY MIXED ONCE THROUGH SYSTEM TO VARIOUS TYPES OF STEP AND CYCLIC SHOCK LOADS

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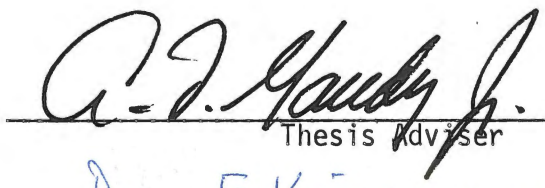

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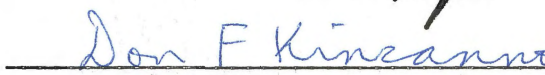

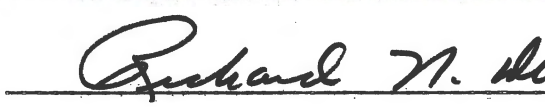

Dean of the Graduate College

Dedicated to my wife, Kathy

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# CHAPTER I

## INTRODUCTION

The new impetus toward pollution control, in this country, has been brought about by the ecological awareness and legislative action of the nation's people. At present there is a "grass roots" estimation of the magnitude of the pollution problem in America as well as an understanding of the long-range ramifications of such a problem. It is for this reason that a push toward further development of practical, efficient, and effective pollution control processes is underway in industry and research institutions throughout the country. To meet this challenge of improved process design, it has been necessary for research engineers to modify operational design of existing unit processes and develop and test new concepts in wastewater treatment. In addition, it is imperative that improvements in the state-of-the-art and in understanding and basic kinetic theory be forthcoming in order to assure a rapid closing of the gap between understanding and practice.

In the past, biological methods of treatment have been used for the elimination of various pollutants from wastewaters. While there are many types of biological processes, they can be generally classified into either fluidized or fixed-bed systems. The fluidized process, more commonly known as the activated sluge process, has been attractive to researchers because of its versatility and opportunity for investigation of operational controls. For research and experimentation purposes,

two modifications of the conventional activated sludge process have been used extensively at Oklahoma State University's bioenvironmental engineering laboratories. The two most applicable to the current research are the extended aeration, activated sludge process, and the once-through, or chemostat, system. Both of these methods of biological stabilization of pollutants have characteristics that make them desirable for use in growth kinetic studies, and both can be operated with relative ease of control.

The extended aeration or total oxidation process has been found to be easily applied to small size plants where nutrient recycle and ease of operation are advantageous. Much research has been conducted on the theory of total oxidation, with a great deal of intellectual discussion finding its way into the literature. To dispel many misconceptions regarding total oxidation, research conducted at Oklahoma State University has been under way over a ten-year period. As a direct result of this research, a process modification has been developed which provides for a high degree of engineering control of biological solids concentration in the extended aeration system. This process modification, termed "hydrolytic assist," has been the object of research by some recent investigators at Oklahoma State University. Since it is an objective of modern bioenvironmental engineers to design control schemes for refinement of operations of existing treatment unit processes, it is important to note that the "hydrolytic assist" offers many new research and practical opportunities to engineers in the field.

Many models for the design of activated sludge processes have been advanced over the years. Some of the most recent kinetic models provide reliable design and operational data for biological systems.

These models, however, depend upon the premise that the system is operating under steady state conditions. That is to say, that the system variables such as dilution rate, influent substrate concentration, cell yield, microorganism concentration, flow rate, and effluent substrate concentration remain constant. There are times, however, in the daily operation of the treatment plant when the activated sludge may be subjected to a change in environmental conditions which tend to disrupt the steady state. These changes or perturbations of the environment can be termed as shock loads. With new effluent guidelines and EPA-sponsored effluent monitoring programs, it is not only sufficient to rely on effluent quality during steady state operation of the treatment plant, but it is also necessary to pursue investigation into the effects of various types of shock loads in order to determine cause-and-effect relationships of shock loads and their subsequent effects upon factors such as changes in predominance of microorganisms and changes in growth characteristics.

The scope of this present study specifically involves the evaluation of the effect of various qualitative, quantitative and hydraulic shock loads upon an hydrolytically-assisted extended aeration pilot plant. Areas of emphasis include purification efficiency, biological solids level, and effluent characteristics. Also considered in this study are the effects of quantitative and hydraulic shock loads upon a once-through chemostat system. These shock loads were administered using a pulsing feed regime or periodic feed schedule. That is to say, the biological solids concentration and purification efficiences are evaluated for shock loads that are administered periodically ar recurring intervals. The shock loads are evaluated on the basis of

variation of environmental conditions at different growth rates or, in the case of hydraulic shock loads, on the basis of variation of the growth rate via variation of dilution rate.

# CHAPTER II

# LITERATURE REVIEW

## A. Development of the Hydrolytically-assisted Extended Aeration Process

One problem that has continually faced wastewater treatment plant designers and operators has been the problem of satisfactory disposal of waste biomass or sludge. A possibly feasible solution to this problem was suggested by the experimental work on dairy wastes done by Porges and his co-workers (1). It was concluded in this study that if biological solids were retained in the aeration chamber, the endogenous respiration or biochemical combustion of the sludge would roughly equal the increase in biomass resulting from assimilation of substrate. Detention times necessary for satisfactory operation of this process were reported to be on the order of twenty-four hours. Porges' findings prompted much intellectual controversy concerning the theoretical possibility of total oxidation of the practical ability of the extended aeration process to accomplish it.

Studies conducted by Symons and McKinney (2) on systems containing low concentrations of nitrogen led them to the conclusion that the total oxidation concept was erroneous. In batch systems, nitrogen concentrations in the feed were varied to gain insight into the biochemistry of nitrogen in activated sludge synthesis. As a result of the

35-day operation of the system, it was concluded that the accumulation of biological material was the result of increases in biologically resistant, extracellular polysaccharide. It was also believed that the increase in polysaccharide would increase the inactive portion of the sludge to a point of system failure. It is interesting to note, however, that one case of significant apparent autodigestion was reported at an intermediate nitrogen level for a period of approximately one-third of the study (see Figure 2, 25.0 mg/l $NH_3$-N). According to Symons and McKinney, there was insufficient data available from which to draw a conclusion concerning the significant decrease in solids shown in Figure 2, page 878.

Busch and Myrick (3) conducted a series of experiments with different influent loadings to determine the limitations of the total oxidation system. From their one-year study, it was reported that a condition of biological solids equilibrium could not be attained, and that continual biological solids increase would lead to a failure of the continuous flow total oxidation system. Also reported by Busch was the cyclic fluctuation of biological solids with respect to concentration and settling characteristics.

A study by Kountz and Forney (4) on the treatment of milk solids by the total oxidation system led to the conclusion that total endogenous oxidation is not possible consistent with reasonable treatment times and treatment plant sizes. Some empirical guidelines based on experimental results indicated that actual endogenous loss is two percent per day of the total weight of sludge in the system. It was also concluded by Koutz and Forney that the accumulation of nonoxidizable sludge is 0.6 percent per day of the total weight of activated sludge

in the unit, and that there can be expected to be a residual material equivalent to 20 to 25 percent by weight of the new activated sludge produced.

To dispel many of the misconceptions of total oxidation, an extensive two-year study was conducted by Gaudy, Ramanathan, Yang, and DeGeare (5). An extended aeration pilot plant was operated at a nominal COD loading of approximately 530 mg/l. The biological solids were carefully maintained so as to prevent any removal or leakage other than by biological combustion. Solids removed from the system in the effluent were subjected to centrifugation and returned to the aeration tank to assure the accountability of biomass. The experimental results indicated conclusively that an extended aeration activated sludge system without sludge wastage could be operated with good biological efficiency and without continual solids accumulation or unacceptable increase in inactive portions of the sludge. It was also observed, however, that the steady state with respect to biomass reported by Porges, failed to materialize. It was concluded by Gaudy that the complex, dynamic ecosystem found in the heterogeneous population was capable of altering predominance ratios in order to allow for specific assimilation of virtually all cellular constituents. Thus, the system could experience periods of solids buildup and periods of de-accumulation due to accelerated autodigestion.

To further examine the practical stability of the extended aeration system, Ramanathan, Gaudy, and Ragthaidee (6) conducted a companion study of the effects of quantitative shock loads on the activated sludge process. An extended aeration pilot plant was developed from sludge used in a previous total oxidation study, and was shock loaded at

increasing sludge ages from 249 to 392 days. The shock loads were administered under both batch and continuous flow conditions at changes varying from 500 to 1000 mg/l glucose step changes to five-fold 500 to 2500 mg/l glucose increases. It was observed that, in general, sludge during continuous flow operation responded more successfully than did sludge under batch operation. In the most severe case, biological solids in the continuous flow pilot plant increased from 7400 mg/l to 8500 mg/l, with the filtrate COD increasing from only 30 mg/l to 50 mg/l. It was concluded that the sludge had exhibited a relatively low specific substrate removal capability and a relatively high degree of stability of operation.

From the research observations, it became apparent that the biological solids in the extended aeration system would not increase indefinitely. It was impossible to predict, however, how long biological solids would increase prior to the de-accumulation of biomass brought about by the predator-prey relationship. It was found, at times, that solids accumulations were very high, making retention difficult with current field procedures. It was necessary, therefore, to explore engineering possibilities for obtaining control of biological solids. One approach for control of the system was reported by Gaudy, Yang, and Obayashi (7). It was postulated that chemical treatment of sludge could enhance the autodigestive process. By chemical hydrolysis (acid, pH = 1) for five hours at 15 psi (1.05 kg/sqm) and 121$^{o}$C, the biological cells were rendered practically soluble. It was then thought by Gaudy that the hydrolyzed, soluble sludge would be readily metabolized by active biological solids when returned to the aeration chamber. To test this hypothesis, sludge was solubilized and fed as a

batch slug dose to an extended aeration pilot plant. The feed hydrolysate was characterized as 50 to 60 percent protein, and 8 to 15 percent carbohydrate, with some light precipitates of inorganic origin present. It was found that the hydrolysate was readily metabolized within a period of approximately one hour with an apparent 90 percent reduction in COD. This COD reduction compared favorably with the reduction of COD observed with a glucose waste to which the sludge had been acclimated for more than two years.

It was concluded by Gaudy, Yang, and Obayashi that an innovative, chemical modification of the extended aeration activated sludge process would enhance the feasibility of the system from an engineering standpoint, while increasing the efficiency and effectiveness of the process. The "hydrolytic assist" enabled the system to do chemically what is difficult to do biologically, and do biologically what is difficult to do chemically. In addition, the total system provided for both digestion of organic material and for disposal of excess sludge to $CO_2$ and water. The system also has the attractive feature of nitrogen recycle for treatment of high strength nutrient-deficient wastes, as well as benefits of simplicity of design. The"hydrolytic assist" modification incorporates sound microbial theory and purification ability with the flexibility of an engineerable unit process.

Additional studies have been conducted by Saidi (8), Murthy (9), and Scott (10) concerning the operational stability of the extended aeration process with hydrolytic pretreatment with additional emphasis on the nitrification characteristics of the sludge. It was found by Saidi that operation at relatively high influent substrate concentrations of 500 mg/l to 1000 mg/l resulted in biological efficiencies

corresponding to those reported earlier by Yang (11). It was also reported by Saidi that it was possible to produce a highly nitrified effluent at organic loadings of 1000 mg/l dextrose and nitrogen loadings of 500 mg/l $(NH_4)_2SO_4$.

A further study conducted by Saidi showed that the hydrolytically-assisted extended aeration pilot plant remained "steady" during shock loads consisting of a three-fold quantitative increase in influent substrate and a halving of detention time. During the shock loads, expected rises in biological solids concentration appeared to accommodate changes in organic loadings, hence, providing good effluent quality.

Murthy (9) further determined the presence of a high degree of nitrification in hydrolytically-assisted systems, and suggested the use of partial unit hydrolysis as an expedient for the removal of filamentous bulking sludge in the extended aeration system. Based on microscopic observation and examination of settling characteristics, it was determined that hydrolysis of filamentous organisms served to "dilute" them from the system, while the hydrolysate, having returned to the aeration basin, served to enhance growth as substrate for more desirable microorganisms. These results were in general accord with earlier work by Yang (11) and Scott (10).

## B. Shock Loads

Shock loads, in general, may be defined as any rapid or abrupt change in the physical or chemical environment in a biological system. Since many waste treatment plants are subject to frequent changes in the environment, research on the effects of shock loads has received

much attention in the last decade.

The major types of shock loads which may disrupt treatment efficiency of biological systems can be described as follows:

1. Quantitative Shock Loads

This shock load involves a relatively rapid change in the amount of organic constituents in the incoming waste stream. This type of shock load can imply a rapid increase or a rapid decrease in the organic content of the water. (Both increases and decreases may cause in-plant operational problems.)

2. Toxic Shock Loads

This type of shock load involves an influx of wastes containing certain materials that tend to disrupt the metabolic patterns of microbial populations (e.g., heavy metals). These toxic materials may result from industrial processes, and may enter the treatment facility as chromates, phenols, cyanates, or other similar compounds.

3. Qualitative Shock Loads

The qualitative shock load refers to a change in the chemical nature of the incoming waste, i.e., a change in the structural configuration of the carbon source to which the sludge has been acclimated. This type of shock load concerns neither the total organic concentration, which may remain the same as before the shock, nor does it imply that the change is toxic to the active biomass.

4. Hydraulic Shock Loads

This type of shock load involves an increase or decrease in the rate of flow reaching a treatment plant. The hydraulic shock load may or may not result in a change in the daily organic loading of the treatment system. Disruption usually occurs when the change in detention time results in a corresponding change in growth rate within the system.

5. Other Shock Loads

As stated before, shock loads involve any change in the environment surrounding a biological system. Some notable types of shock loads, other than those previously mentioned, include pH and temperature shock loads. These and other types of disruptive changes must necessarily be the subject of a voluminous amount of continuing research to assure improvements in the efficiency of operation of wastewater treatment processes.

In general, a response of a system to shock loads is considered successful if the response does not act in any way to the detriment of the normal functioning of the process. The response of a system to a shock load will depend greatly upon the degree of change introduced to the system. If the change is adverse, impairment of biochemical and physiological functions may result. According to George (12), a successful response will depend upon factors such as

a) the severity and/or rapidity of the shock load

b) the detention time in the aerator

c) the physiological characteristics of the sludge

d) biomass concentration

e) the oxygen tension

f) the predominating microbial population and the population diversity.

As stated previously, the scope of this study will involve two of these types of shock loads--quantitative and hydraulic. It will, however, be advantageous to examine some of the other types of shock loads as they pertain to transient models and advances in engineering knowledge.

## C. Effect of Dilution Rate on the Performance of Continuous Cultures

From the theory of continuous culture, it is well established that under steady state conditions in a once-through completely mixed system the logarithmic growth rate of the culture is determined by and is equal to the dilution rate of the system. This theory has been put into practice by microbiologists and bioengineers to establish the effect of growth rate on the metabolic responses of both pure and mixed microbial populations. This research with chemostat or once-through systems also provides the investigator with a conservative estimation of the effects of hydraulic changes on more conventional biological systems involving cell recycle. It is also important to review this work in light of the biochemical adaptations that are made by microorganisms in response to changes, in terms of nutrient requirement and cell composition.

George and Gaudy (13)(14)(15) investigated the effect of hydraulic, pH, and temperature shock loads on a heterogeneous microbial population

in a once-through system. This research led to the conclusion that for systems initially operating at a dilution rate of 0.125 $hr^{-1}$, changes in flow rate leading to dilution rates greater than 0.25 $hr^{-1}$ (detention time less than four hours), or less than 0.062 $hr^{-1}$ (detention time greater than 16 hours) would result in a deleterious transient response as indicated by increase in effluent COD and decrease in biological solids. Guidelines from this study included the observation that a steady state system operating at a dilution rate of 0.125 $hr^{-1}$ (8-hour detention time) can accommodate no greater than a 100 percent increase in flow rate under constant concentration conditions or a 50 percent decrease in flow rate under constant loading conditions. It was also reported by George (12) that cell yield values decreased at decreased dilution rates and increased at increased dilution rates. Protein and carbohydrate content of cells was also decreased at flow rates greater than those present under steady state conditions--the reverse being true at decreased flow rates. It was further reported that shock loads greater than changes from 0.125 $hr^{-1}$ to 0.25 $hr^{-1}$ resulted in significant reduction of COD removal efficiency, while at decreased flow rates, changes in removal efficiency were less significant.

Regan, Roper, and Moss (16) applied various step changes in dilution rate to continuous cultures of the yeast *Saccharomyces cerevisiae*. Their studies led them to suggest that step changes in glucose feed rates or dilution rates induce a synchrony between individual cells, which resulted in the oscillatory response of the entire culture. In the particular case of *S. cerevisiae*, budding of the individual yeast cells was found to accompany the step increase in

dilution rate and precede a substantial increase in active biological solids.

This experimental work corresponds nicely with earlier continuous culture experiments using *S. cerevaisiae* done by Gilley and Bungay (17). It was reported by Bungay, however, that budding in the yeast culture was experimentally eliminated as an explanation for the oscillatory behavior of the transient state. This appears to indicate that the oscillatory behavior of the continuous culture can be the result of complex biochemical and physiological factors, rather than the result of single physiological processes, such as budding. The oscillatory pattern described by Moss is also discernible in the experimental results of George (12) in mixed microbial populations. It is doubtful that step changes in dilution rate under these conditions would trigger mass division in a heterogeneous microbial population because of the diversity of species. This indicates that the synchrony described by Regan and co-workers is a general transient response mechanism to a shock load.

Mor and Fiechter (18) used continuous cultures of *S. cerevisiae* to study growth characteristics under transient state conditions. The experimental results obtained from dilution rate or hydraulic step changes led the researchers to describe a damping effect on the oscillations of microorganism and effluent substrate concentration in the reactor. It was observed that when a positive step change in dilution rate is made, the concentrations in the reactor increase over the prior steady state value and then damps off to a higher final steady state. When the step change is negative, these trends are inverted. These reactions were termed "overshoot" and "underswing," respectively. Mor

and Fiechter attributed this phenomenon to changes in the yield coefficient, brought about by either energy for maintenance, inhibitor formation, or changes in specific growth rate.

Meers (19) concluded, from his work with *Bacillus subtilis* and *Torula utilis* at various changing dilution rates, that changes in flow rate into a biological waste treatment plant could result in changes in the dominant species of micro flora. Meers found that pulse changes in dilution rate of a mixed culture from 0.05 $hr^{-1}$ to 0.08 $hr^{-1}$ resulted in a culture of *B. subtilis* that appeared pure upon microscopic examination. Reversal of the dilution rate change allowed the undetectably small population of *T. utilis* remaining in the reactor to reverse the predominance trend.

Yu (20) studied the effects of hydraulic shock loads in continuous flow activated sludge processes which were fed component substrate. He found that the biological response to a severe increase in dilution rate was a rapid washout of biological solids and a high degree of leakage of COD in the effluent. The typical response to a severe decrease in dilution rate was similar to the response reported for an increase in dilution rate. These results were similar to the results obtained by George (12) using a different substrate (glucose, as opposed to glycerol used by Yu). Yu also determined that the extent of disruption of system efficiency during hydraulic shock loads is dependent upon the severity of the shock load and the rapidity at which it is applied. Also important in determining the extent of disruption is the "age" of the cells before the shock was applied.

## D. Models of the Transient State

Numerous attempts have been made to describe the performance of activated sludge systems under steady state conditions (21)(22)(23). These analyses generally applied mathematical models similar to those described by Novick and Szilard (24), Monod (25), and Herbert (26) to describe the continuous pure culture systems. These developments rest basically on the Monod (27) expression for growth and yield which was originally developed for a single species or organism with a single growth-limiting substrate. From the kinetics of microbial populations, however, some interest has shifted into the area of transient microbial population kinetics.

The importance of accurate prediction of shock load relationships and effects has been discussed by several researchers in the literature. Approaches to design and prediction of transient response must, however, be considered to be at best only in the early stage of development. The sheer complexity of the material under study, coupled with the effects of biochemical reaction and biological interaction of a perturbed living system, compounds the problem immeasurably. It is by no means sufficient to treat the area of shock loads as a "black box," mass balance exercise. It will be, however, necessary to assemble a voluminous amount of experimental and operational data before it will be possible to gain insight into the biological response of dynamic, transient, microbial populations. Some model studies will be commented on in this chapter; it should be noted again that these do not at this time represent a definitive answer to prediction of shock load response.

Grady (28) developed analog computer models of transient responses of shock loads based on a mass balance written around the reactor. From these studies, several conclusions were drawn tentative to verification by experimental studies. It was determined that the biochemical response to a shock load is strongly dependent upon the steady state specific growth rate prior to the shock, and the lower the growth rate, the better the response. It was also concluded that the response to a shock load is independent of reactor hydraulic retention time prior to the shock. It was further concluded that the biological response is a primary function of the change in the mass rate of substrate input into the reactor and is independent of the manner in which the shock is applied.

Caperon (29) performed a relatively long term chemostat study to determine the time lag response of *Isochrysis galbana* to changes in dilution rate in a nitrogen-limited system. Caperon reported two separate studies of 58 and 79 days in which a pure culture of *I. galbana* was subjected to periodic variations in dilution rate. In the first experiment, the culture was grown at a constant growth rate for several days to ascertain the steady state conditions. The dilution rate was increased for a period of time until a new steady state was approached. This determined when a new increase in dilution rate was to begin. In experiment two, the same protocol was employed; however, after changes in dilution rate (increase) for two to four days, the system dilution rate was held at zero to enhance a transient decrease in system solids. In both experiments, the observed data were plotted against three mathematical models: model one represented specific growth rate as a function of nutrient concentration and its effect on the rate

of change of nutrient concentrations. Model two introduced a time lag constant into the model, and model three represented growth rate as dependent upon an average of past nutrient concentration experience over the preceding twenty-four hours. It was concluded by Caperon that the data fit reasonably well to all models, particularly to the third. It was stated, however, that much more research remained to be done to determine whether to describe the lag phase as a constant or a variable and to increase the descriptive fidelity of the model.

Ierusalimskii, Stepanova, and Chernauskii (30) described inhibitor-induced death and the phenomenon of biological inertia as possible explanation for the oscillatory behavior, frequently observed in continuous cultures during transitional periods. The authors observed that when approaching a new steady state level, the specific growth rate does not change at once, but only after a particular lag phase. This process is apparently associated with the fundamental transformation of the internal apparatus of the catabolic mechanism of the cell. The result is a gradual dampening of oscillation into the new steady state value. It was emphasized by the authors that the oscillatory models do not always appear and do appear only under certain conditions. To describe the oscillatory state, a mathematical model was developed to encompass some specific biochemical and physiological parameters. As an indicator of the lag period, as described by this inertia theory, the synthesis of the large organella, the ribosome, was chosen as a model variable. To determine the relationship between growth rate and the number of ribosomes per unit biomass, the variable of limiting enzyme concentration was introduced via the Michaelis-Menten type relationship. It was disappointing that no experimental

verification of this model was presented. Although the concepts are interesting, it is doubtful that the model will serve as an operational model at any practical level.

Sterkin, Chirkov, and Samoylenko (31) studied the effects of rapid changes of dilution rate, temperature, and growth-limiting substrate upon continuously mixed cultures of *Escherichia coli* and *Pseudomonas fluorescens*. It was concluded, in this study, that the oscillatory characteristics of transitional states could be explained, in part, by the biological inertia theory as previously reported by Ierusalimskii, Stepanova, and Chernauskii (30). To include this phenomenon, an inertia factor was incorporated into a mathematical model to predict growth conditions in a completely mixed system. It was noted by the authors, however, that the phenomenon of biological inertia did not occur in a number of experimental curves. To illustrate this point, a pulsing, quantitative shock load was administered to a biological reactor. Upon recurring pulses of equal amounts of glucose, the reaction of the culture changed to exhibit a substantially more immediate return of the culture to a new steady state with respect to cell and effluent substrate concentration. The authors interpreted the results of this short-term experiment as a change in population age distribution which markedly influenced the effects of biological inertia.

McLellan and Busch (32) determined that it was possible to measure the reaction potential of a mixed microbial population from batch studies. The reaeration potential was defined as the total mass of soluble organic carbon which a unit mass of microroganisms is capable of utilizing in a given time period. This factor in conjunction with steady state loading data led Busch to conclude that microorganisms

have a significant reserve potential to handle shock loads. From observations of hydraulic and quantitative shock loads (four-fold and six-fold increases in influent substrate) it was determined that it was possible to predict whether an increase in loading will cause an increase in effluent organic content by analysis of batch reaction potential data.

Schaezler, McHarg, and Busch (33) proposed a transient design model based on the theory that specific growth rate is independent of substrate concentration. It was reported that flux of substrate described as

$$\frac{\text{dilution rate x influent substrate concentration}}{\text{microorganism concentration (dry wt)}}$$

is the controlling factor of growth rate, rather than the substrate concentration as predicted by Monod-type equations.

The Monod equation (27) has been determined to be a reasonably reliable model of steady state microbial population kinetics. There has been, however, a number of researchers who have proposed the Monod relationship for describing the transient response of microbial populations to environmental change. Northam (34) became a proponent of the Monod equation for use in batch and continuous cultures. In an independent study, Luedeking and Piret (35) similarly extended the use of the Monod equation to shock loaded systems. In activated sludge systems, Eckhoff and Jenkins (36) proposed a modification of the Monod equation which contained an approximation for transient conditions and a variable for absorption of substrate on floc particles. A formal discussion by Gaudy (37) of the Eckhoff model (36) accurately points

out many of the inconsistencies and misconceptions regarding the Monod equation as a transient model.

Mateles and Goldthwaite (38) conducted an investigation into the stability of product-limited continuous cultures of *Saccharomyces carlsbergensis* and *Pseudomonas ovalis*. They reported no oscillations resulting from changes in glucose concentrations or overshoot conditions upon changes from one steady state dilution rate to another. This led them to conclude that the transient prediction model of Luedeking and Piret (35) was experimentally sound. This also led to the investigation by Mateles, Rye, and Yasude (39) of the further functional use of the Monod equation as a model of transient state.

Young, Bruley, and Bungay (40) compared experimental data of Storer and Gaudy (41) with predicted data using the Monod equation (27). It was found that for step increases in influent substrate concentration, the equation was unacceptable, due to the time independent nature of the equation. This information is highly supportive of work done by Mateles, Rye, and Yasuda (39). A model was described by the authors to predict the transient response of chemostats to dilution rate and substrate concentration. The model provides for the variability of factors describing delay of substrate transport across cell membranes, cell maintenance energy and cell yield to predict the transient response of chemostat systems to perturbations in dilution rates and influent substrate concentrations.

Koga and Humphrey (42) developed a model for prediction of transient response in continuous cultures, using as a basis the Monod equation (27). Analysis by phase plane plots was used to show the various possible types of behavior which might theoretically be

expected for transient conditions of the system. It was found that the Monod model represented a highly stable system; that is to say, that cell and substrate concentrations would show a single maximum and/or single minimum in their return to steady state levels when disturbed. To explain the oscillations of cell and substrate concentrations that had been observed experimentally by several investigators (16)(17)(18), Koga and Humphrey introduced the yield coefficient as a variable rather than as a constant. This modification of the initial model indicated the occurrence of a dampened oscillatory behavior at a low dilution rates similar to the responses reported by Ierusalimskii, Stepanova, and Chernavskii (30).

Storer and Gaudy (41) examined the feasibility of using the Monod equation as a predictive model to describe transitional stages of growth. It was determined that during a quantitative increase in influent substrate, growth rate hysteresis was observed to occur. This made it theoretically impossible to use the Monod relationship, since instantaneous changes in growth rate to changes in substrate would be required. It was further indicated that the cell yield may not be constant during the transition phase.

Mateles, Rye, and Yasuda (39) used step changes in dilution rate to ascertain whether the Monod equation was valid under nonsteady state conditions. From experiments with nitrogen-limited *E. coli*, it was concluded that the Monod equation always lacks a maximum and is a smooth transition between steady states. It was, therefore, determined from the data that during transient operation of a continuous culture, Monod's equation may not be used to relate growth rates to concentrations of limiting nutrients.

## E. Quantitative and Qualitative Shock Loads

Some shock load studies have been initiated as the result of advances in the literature on the subject of sequential substrate removal in heterogeneous populations (43)(44)(45)(46). Gaudy, Komolrit, and Gaudy (47) explored the effects of response of activated sludge systems to qualitative shock loads with specific emphasis on sequential substrate removal. Both "young" and "old" cell systems were used in the study to determine the effect of sludge age upon the biological response of a system to shock loads. In the "young" cell systems, which were acclimated on mannitol and sorbitol, suppression of substrate removal (of sorbitol or mannitol) was observed following addition of a new substrate (glucose) into the system. In the "old" cell system, however, the suppression was not in evidence. In the opinion of Gaudy and co-workers, many enzymes necessary for conversion of glucose in the Embden-Meyerhoff-Parnas pathway were absent in the "older" sludge. It was also determined that the enzymes can be repressed under certain physiological conditions, such as those encountered in the 24-hour batch feeding systems used to develop the older sludge (48). Komolrit and Gaudy (49) further studied the effects of substrate interaction in qualitative shock loads of "young" biological systems. The authors found that under severe shock load conditions, the various combinations of substrate compounds exhibited various degrees of suppressive or repressive responses. General patterns of carbohydrate-carbohydrate and carbohydrate-alcohol systems were established and analyzed on the basis of flow patterns in metabolic pathways. A discussion concerning two types of controls of biosynthetic pathways, enzyme repression, and

the "glucose effect" was also included in the presentation.

Krishnan and Gaudy (50) expanded previous work on qualitative shock loading in an attempt to determine the effects of changes in substrates of considerable difference in chemical composition upon biological systems. A three-carbon alcohol glycerol was used to acclimate all heterogeneous cultures used in the study. "Young" and "old" cell cultures were then subjected to additions of shock feed, composed of the six-carbon sugar glucose. It was found that the $C_3$ to $C_6$ substrate change responded in an essentially identical manner, as did $C_6$ to $C_6$ substrate systems in previous studies by Gaudy (45). This work supported the author's conclusion that a previously unstudied type of metabolic control mechanism that acts in catabolic pathways in a manner analogous to feedback inhibition in anabolic pathways, exists in many bacteria. It was further concluded that the mechanism must function at the enzyme-level rather than at the gene-level. Another interesting observation involved the release of intermediates during the metabolism of glucose by the glycerol-acclimated culture.

Gaudy and Engelbrecht (51) studied the effects of quantitative and qualitative shock loads on an activated sludge system. From this study, it was concluded that the time required for successful response to shock loads and the rate of substrate removal was dependent upon the chemical structure of the substrate compound. From studies on quantitative increases in substrate, it was determined that a successful response can occur, even in the absence of a simultaneous balanced increase in nitrogen. In qualitative shock loads, however, balanced nitrogen-enriched medium was required for successful responses to the environmental stress.

Gaudy (52) examined the effects of qualitative shock loads of spent sulfite pulp mill wastes on activated sludge. It was found that the high strength, complex mill waste was relatively quickly acclimated by a previously glucose-acclimated sludge. It appeared that the majority of substrate constituents were metabolized with the same facility as glucose, with the exception of a small portion of substrate that appeared to require acclimation. In addition, Gaudy reported that in glucose-fed, glucose-acclimated systems, the biological solids decreased in the latter portion of the aeration period. However, when the mill waste was fed to glucose-acclimated systems, the biological solids reached a peak where they remained for the duration of the aeration period.

Su (53) studied the response of biological, once-through systems to multicomponent substrate and hydraulic shock load conditions. He concluded that it takes at least ten detention times for a chemostat system to approach a new "steady state" condition after a change in dilution rate. He observed additionally a heavy wall growth and the presence of flocculated cells at high dilution rates. He reported that this occurrence hampered accurate sampling and analysis during these portions of the study.

Gaudy and Turner (54) conducted experiments to determine the effect of air flow rate on the response of activated sludge to shock loads. Sludge from an activated sludge pilot plant was batch fed and maintained for a prolonged period of time to assure a constant solids balance. On a periodic basis, the normal feed schedule was interrupted and a batch shock load was imposed. Samples from the aeration tank were assessed, as were samples used in concurrent Warburg studies.

The data from this experimental work indicated that a short term shock load, which resulted in essentially zero dissolved oxygen in the reactor, does not severely affect the biochemical efficiency of the system. It was also the opinion of Gaudy that increases in air flow enhanced the rapidity of response to a slight degree. From this discussion of experimental results, the conjecture that the value of oxygen tension which affects metabolic rate is below 0.5 mg/l, was supported.

Ragthaidee (55) operated an extended aeration, activated sludge pilot plant without sludge wastage, to determine the results of quantitative shock loads upon the biological efficiency of the system. He reported excellent 95 percent COD removal throughout the course of the study, which included a five-fold increase in influent COD. The good response of the system, under shock load conditions, was attributed to the relatively high biological solids concentration in the reactor, in conjunction with the relatively long detention time employed in the study. The author further concluded that the successful response of extended aeration was not dependent upon the cell age of the system.

Kiravanich (56) concluded studies exposing batch, once-through, and extended aeration activated sludge, biological systems to various types of shock loads. During this study, a rather extensive attempt was made to ascertain the effects of combined (qualitative and quantitative) shock loads upon biological growth kinetics. Kiravanich reported that the extended aeration pilot plant was relatively free of metabolic disruption under severe four-fold multisubstrate shock load conditions. It was the opinion of the author that the reason such systems can accommodate severe shock loads is due to the high biological solids concentration. These results indicate that the retention of

solids (i.e., no wasting of solids) does not cause severe deterioration of the metabolic activity of the sludge. From further hydraulic shock load studies on the extended aeration process, it was determined that the loss of solids in the clarifier effluent that was found to occur as a result of the shock, was the cause for the most concern in consideration of the shock load effects. It was concluded that the biochemical inability of the system to remove soluble organic carbon was not seriously impaired under the conditions of the study. The dilute-out of cells following a hydraulic shock load, however, was found to warrant serious consideration in future studies.

Shock loads brought about by intermittent industrial operation and peak domestic usage periods were illustrated from conditions at the Peoria, Illinois, treatment plant by Kraus (57). This article points out that shock loads are not necessarily step changes, but may be considered as either uniform or un-uniform surges into the treatment facility.

Anagnostapoulos (58) utilized a semi-continuous culture system of *E. coli* to establish synchronization of cell division. Glucose-limited medium was supplied to the reactor at intervals of one to four hours, and one-half of the diluted culture was discharge. The system was, in effect, an unconventional, cyclic shock load system. The pulsing regime of substrate introduction and biomass removal successfully caused synchronization of cell division in some instances. Other experiments, however, using the same feed regime generated relatively un-uniform, "bi-rectilinear" growth curves and, in some instances, growth patterns characteristic of diauxic growth.

Knopp, Watts, and Rohlich (59) conducted random pulse-type

quantitative shock loads on an activated sludge system. In this study, a biological reactor was subjected to shock loads of substrate which varied as a sequence of step changes of random height and equal duration. The data obtained from this pilot plant study were then subject to scrutiny by frequency response analysis. The agreement of the frequency response analysis of input COD with linear model derived by Rohlich and co-workers led the author to the following conclusions:

1) COD is a poor indicator of system response, due to the fact that the COD measurement is far too gross a measurement to indicate the dynamic changes within the system.

2) The activated sludge system can be described as a low frequency pass system.

3) Frequency response analysis of operational plants in the field is recommended to assist in development of control models for the activated sludge process.

From the foregoing review of literature in the fields of microbiology and pollution control engineering, it can be seen that initial studies concerning shock load systems have been undertaken.

The area of transient responses of biological systems is, however, extremely extensive in scope. At the present time, design conclusions concerning shock load conditions are in the infant stages of development, with enormous amounts of experimental work remaining to be done. It is felt that all areas of shock loading typical to pollution control must be investigated thoroughly to ensure effective biological design, efficient in-plant operation strategies, and improvement in the understanding of microbial kinetics. These improvements in the state-of-the-art may be accomplished relatively easily in the laboratory

using bench-scale reactors and analytical techniques usually employed in the pollution control field. While many of the studies herein reported appear at first glance to be of an unpractical nature, this is, indeed, not the case. All shock load studies have an important, if not immediate, application to the on-line biological systems in the field. With the increased emphasis on EPA guidelines, it has become increasingly important to produce a uniformly high quality product from the treatment process. This adds as an additional requirement, the necessity for intelligent analysis of disruptive influences on biological systems--in short, shock loads. It is to assist in this accumulation of basic knowledge that this research into the effects of hydraulic and quantitative shock loads was conducted.

# CHAPTER III

# MATERIALS AND METHODS

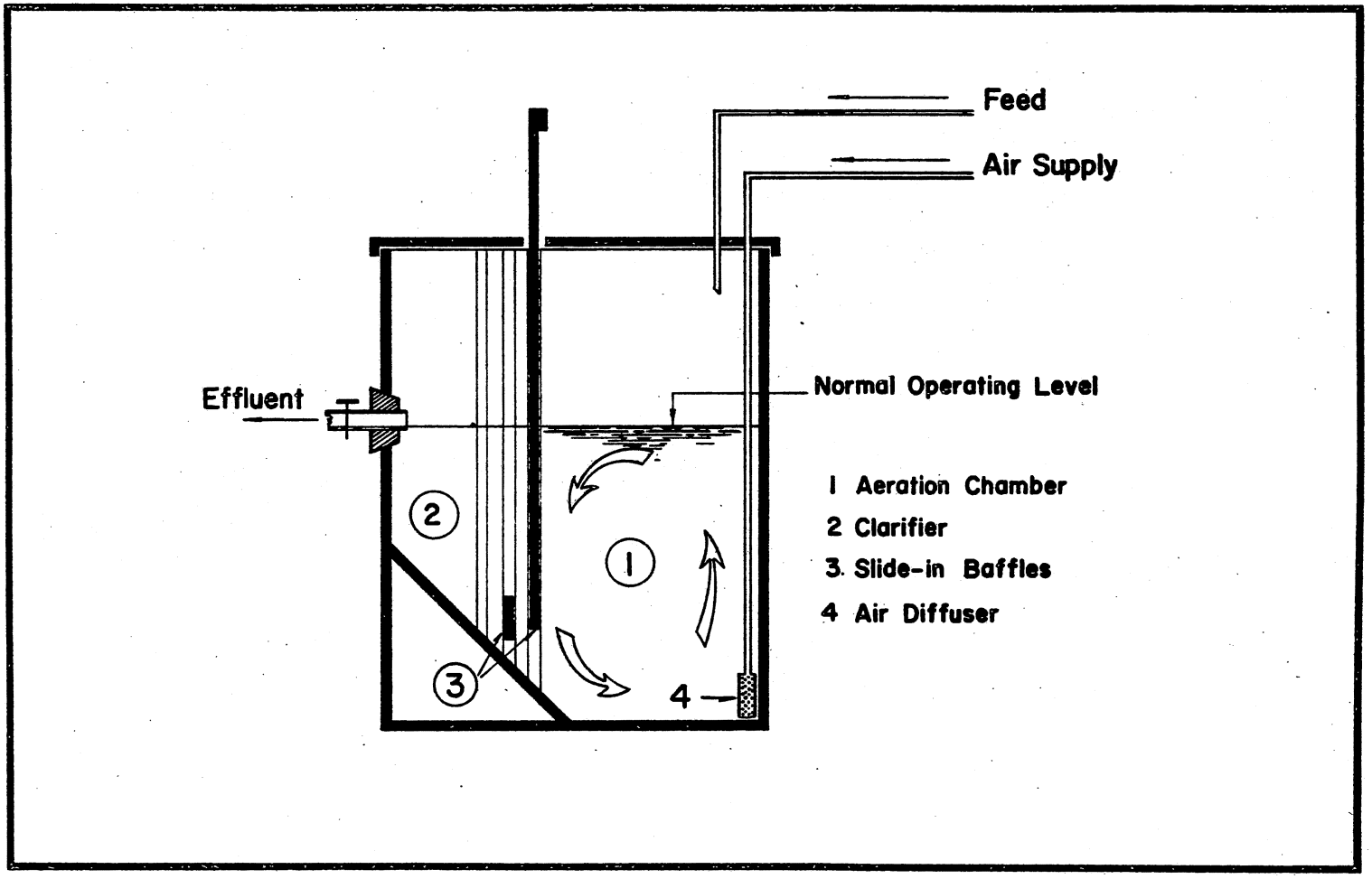
## A. Shock Load Studies on the Extended Aeration Activated Sludge System, With "Hydrolytic-assist"

Before describing the experimental equipment, it will be advantageous to provide a history of the development of the extended aeration activated sludge used in the study. The active biomass was developed by seeding a synthetic waste with sewage obtained from the primary clarifier at the municipal sewage treatment plant at Stillwater, Oklahoma. The pilot plant unit was put into operation as a batch system on 22 October 1973, and converted to continuous operation on 19 November 1973. The sludge was completely retained in the system, with the exception of 25 ml required for daily analysis and the small portion of the sludge leaving the system as effluent suspended solids.

### 1. Experimental Apparatus

Figure 1 shows the cross-section of the plexiglass, total oxidation unit employed in this phase of the study. During the initial startup phase, the baffles were removed from the pilot plant to allow for batch operation of the system. The total volume in this mode of operation was 9.4 liters. For continuous operation, plexiglass baffles were inserted

Figure 1. Cross-section of an Experimental Bench-scale Activated Sludge Unit With Internal Recycle


Feed
Air Supply
Effluent
Normal Operating Level
1 Aeration Chamber
2 Clarifier
3. Slide-in Baffles
4 Air Diffuser
1
2
3
4

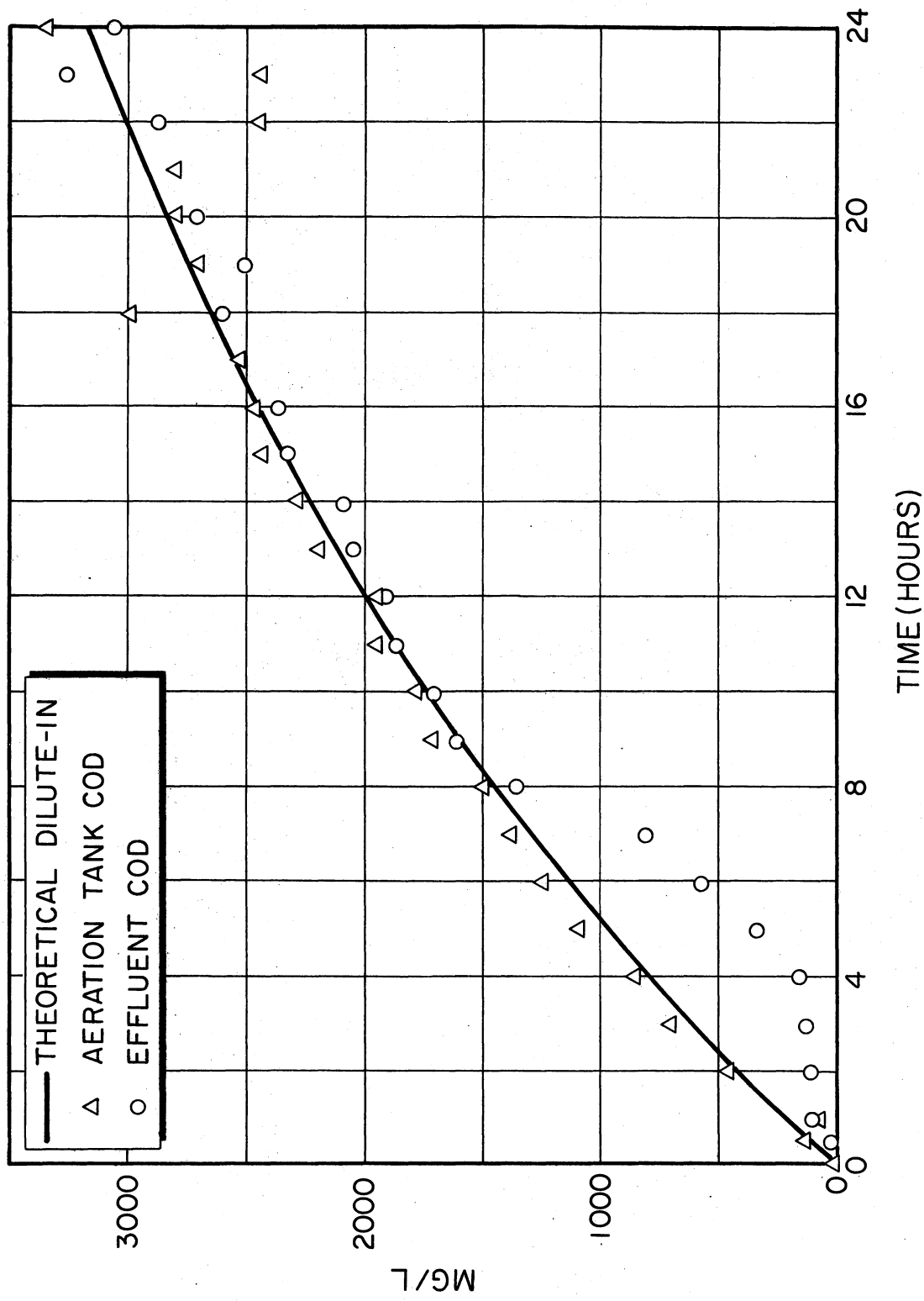
into the slotted portion of the unit sidewall to divide the unit into two parts. The two divisions comprised a 6.2-liter aeration basin and a 3.2-liter settling basin. The detention time in the aeration basin was approximately 16 hours with a corresponding 8-hour detention period in the settling basin. Total system detention time remained at 24 hours throughout the study.

To assess the efficiency of the separation between aeration tank and clarifier detention times, a "dilute-in" experiment was conducted. The unit was filled with a distilled water and one percent commercial bleach solution. At time zero, a pump delivered a 5035-mg/l solution of glucose into the system at a flow rate necessary for a total-system-detention time of 24 hours. COD concentration in the aeration tank and the effluent were determined and plotted as shown in Figure 2. From the data, it was concluded that the system does, indeed, provide for separation of aeration tank (mixed) and settling tank (quiescent) detention times when using soluble substrate. This analysis was also important in determining the possible effects of short-circuiting in the combined system. Sludge was returned to the aeration chamber, due to the bottom angle of the "stilling" basin in conjunction with the "suction" provided by the aeration equipment. Compressed air was provided through four carborundum diffusers attached to the sidewalls of the aeration chamber. Air flow rate was regulated at 9 c ft/hr (4.25 liters/min), using a Gelman air flow regulator.

## 2. Standard Synthetic Wastes

The daily synthetic wastes used in all experiments consisted of glucose and other essential inorganic salts, as shown in Table I. In

Figure 2. Comparison of Theoretical and Experimental Dilute-in Values for Soluble Substrate in the Aeration Tank With Those Found in the Effluent


THEORETICAL DILUTE-IN
AERATION TANK COD
EFFLUENT COD
MG/L
3000
2000
1000
0
0
4
8
12
16
20
24
TIME (HOURS)

quantitative shock load experiments, higher glucose levels were employed with proportional increases in nutrient concentrations. Care was taken to assure that inorganic salt concentrations were in excess, and that the growth-limiting factor was glucose. The addition of tap water provided a source for trace elements. All chemicals used in the study were of ACS grade. Daily additions of hydrolysate were also made since the process was operated in accordance with the concept of the "hydrolytic assist." Operational procedure for this process is discussed below.

TABLE I

COMPOSITION OF FEED FOR 1000 mg/l GLUCOSE SUBSTRATE

| | |
|---|---|
| Glucose | 1000 mg/l |
| $(NH_4)_2SO_4$ | 500 mg/l |
| $MnSO_4 \cdot 7H_2O$ | 100 mg/l |
| $FeCl_3$ | 0.5 mg/l |
| $CaCl_2$ | 7.50 mg/l |
| $MnSO_4 \cdot H_2O$ | 10 mg/l |
| Hydrolysate | variable concentrations |
| Phosphate buffer 1.0 M (pH 7.2) | |
| ($KH_2PO_4$, 38.5 gm; 1 + $K_2HPO$, 124.5 gm/l) | 10 ml/l |
| Tap water | 100 ml/l |
| Distilled water | to volume |

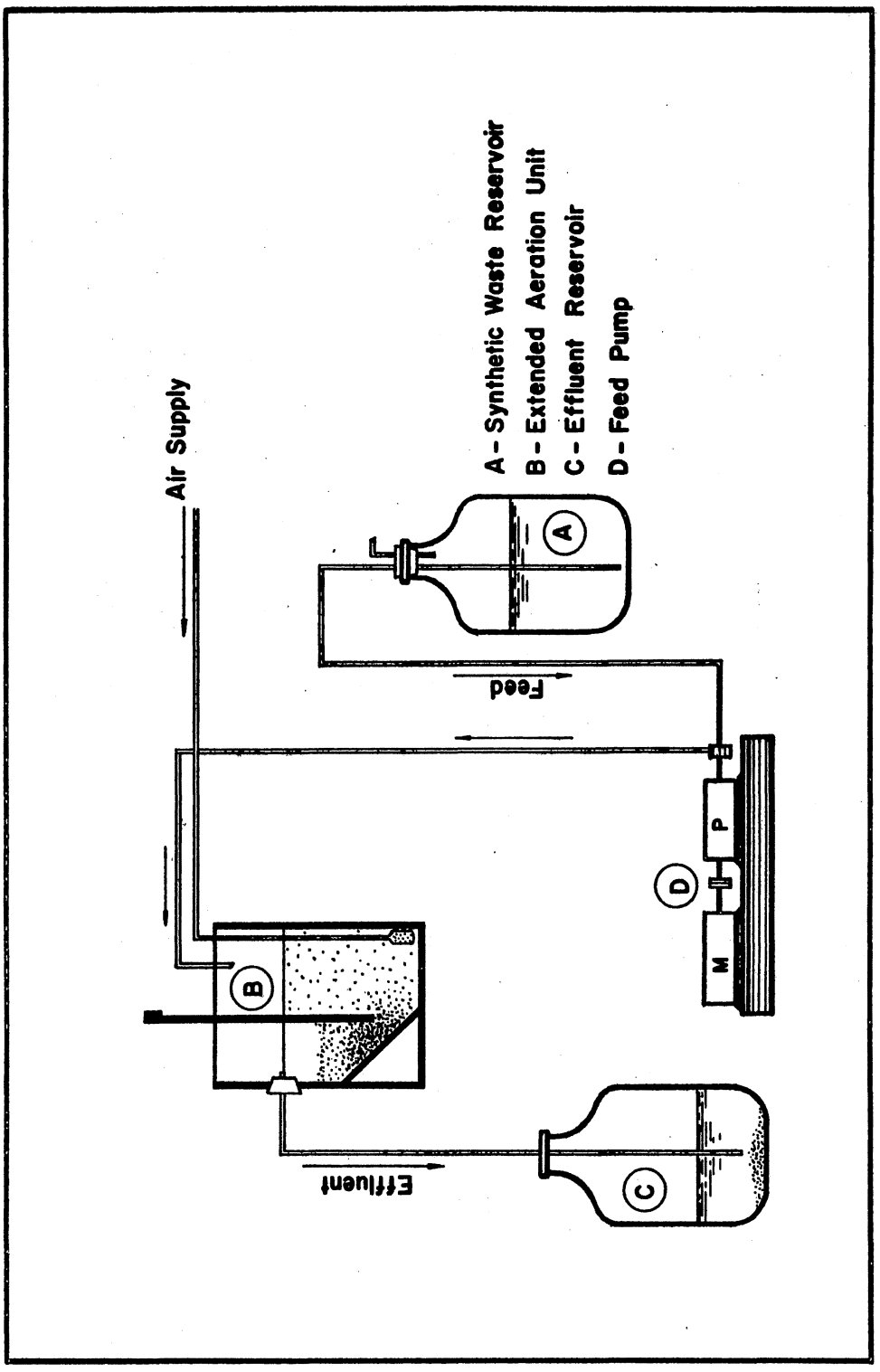
3. Procedure

For continuous flow operation, feeding was regulated by a Milton Roy positive displacement pump (Model 4-C-48R). Feed rate was adjusted to a 24-hour total system detention time, as reported earlier, with a 16-hour aeration tank detention time and an 8-hour clarifier detention time. The feed solution was prepared daily to prevent contamination of the reservoir apparatus. To prevent growth in the feed lines, the feed pumps were alternated on a daily basis. As one pump was in use, the other was used to recycle a chlorine (five percent commercial Clorox bleach) solution. Prior to its return to the feed reservoir, the line was rinsed thoroughly with distilled water. Influent and effluent were stored during the 24-hour period in 5.3 gal (20-liter) Pyrex glass carboys. To secure daily suspended solids samples, the plexiglass baffles were removed and solids in both chambers allowed to mix thoroughly. When sampling was completed (average elapsed time, five minutes), the baffles were replaced and normal operation restored. Effluent samples were collected in a 100-ml graduated cylinder attached to the effluent drawoff line. A schematic representation of the bench-scale system is shown in Figure 3.

The pH of the mixed liquor was determined at frequent intervals and maintained, with phosphate buffer, near neutral pH (6.9 to 7.4). When during the course of a shock load experiment the pH of the mixed liquor showed considerable deviation, it was brought back to the neutral range by additions of alkali (NaOH) solution. The system was maintained at room temperature ($21 \pm 3^{o}C$).

At weekly intervals, a solids determination was made on the mixed

Figure 3. Schematic Flow Diagram of a Laboratory-scale Continuous Flow Extended Aeration System

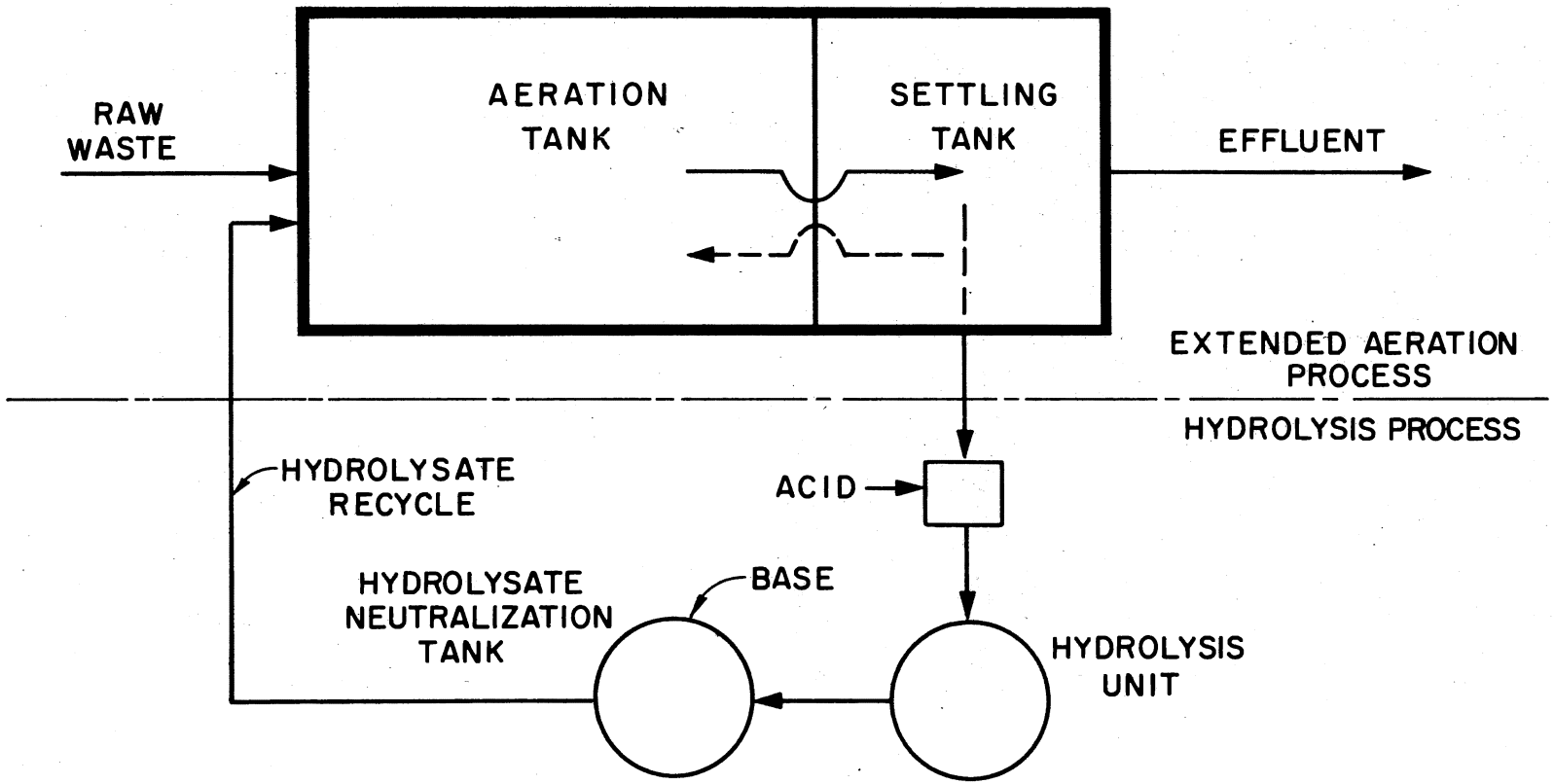

A - Synthetic Waste Reservoir
B - Extended Aeration Unit
C - Effluent Reservoir
D - Feed Pump
Air Supply
A
Feed
P
D
M
B
C
Effluent

liquor for the purpose of determining amounts of sludge to be hydrolyzed. The amount of sludge withdrawn for hydrolysis was calculated to reduce the suspended solids concentration to a pre-determined level, e.g., if suspended solids were at 5000 mg/l in the system, then the volume of mixed liquor suspended solids necessary to restore the solids concentration to 4500 mg/l would be removed. This sludge was then acidified to pH 1 and subjected to 121$^{o}$C and 15 psi for a period of five hours (in a laboratory autoclave). The sludge or hydrolysate was removed, neutralized and fed back into the reactor via the feed reservoir. The hydrolyzed sludge was fed in equal daily portions over the next 7-day period. An example of a flow diagram showing the incorporation of the "hydrolytic assist" process is provided in Figure 4.

4. Experimental Protocol

Growth medium of 1000 mg/l plus hydrolysate was fed during attainment of equilibrium conditions prior to applying shock loads. The "hydrolytic assist" process was set into operation on the extended aeration system on 19 November 1972--47 days before the shock load studies were initiated. To introduce quantitative shock loads into the system, the influent growth medium in the food reservoir was changed to the desired level of glucose or hydrolysate. This "shock feed" was introduced for a specific period of time, e.g., one detention time. For hydraulic shock loads administered in conjunction with quantitative shock loads, the shock feed was introduced at an increased flow rate designed to produce the desired hydraulic flow and detention time. During shock load studies, samples were obtained using the following procedure: the effluent lines were sealed periodically, the baffle

Figure 4. Flow Diagram Showing the Incorporation of the "Hydrolytic Assist" Into the Extended Aeration Activated Sludge Process


RAW
WASTE
AERATION
TANK
SETTLING
TANK
EFFLUENT
EXTENDED AERATION
PROCESS
HYDROLYSIS PROCESS
HYDROLYSATE
RECYCLE
ACID
BASE
HYDROLYSATE
NEUTRALIZATION
TANK
HYDROLYSIS
UNIT

removed, and the unit completely mixed for approximately two minutes. A 25-ml sample of mixed liquor was withdrawn for analysis from the unit and the baffles replaced. As solids subsided in the clarifier below the effluent port, the effluent line was opened and normal operation restored. Between periods of mixed liquor sampling, 50-ml samples of effluent were collected from the effluent drawoff line and analyzed. After shock loading, the unit was returned to normal daily operation for a sufficient period to allow for return of the biomass to semi-equilibrium conditions. The above procedure was then repeated until all of the shock load experiments were completed.

5. Analytical Methods

The chemical oxygen demand test was employed to determine the total organic concentration in solution. The COD test was run in accordance with Standard Methods (60); COD analysis was made of total effluent COD, filtrate COD, and of feed solutions.

The biological solids concentration in both the mixed liquor and effluent was determined by the membrane filter technique (Millipore Filter Co., Bedford, Mass., HA 0.45 μ) as outlined in Standard Methods (60). Aluminum dishes were used to hold the filter papers. Filters were dried for two hours at 103$^{o}$C and equilibrated in a desiccator prior to weighing. The sample was filtered, and the same drying, cooling, and weighing procedures mentioned above were followed.

## B. Shock Load Studies on Once-through Chemostat Systems

### 1. Experimental Apparatus

Figure 5 shows schematic representation of the once-through system used in this study. The aeration vessel was made of Pyrex glass and was 2.3 liters in volume. Aeration and mixing was provided by compressed air supplied through two carbonundrum diffusers. Sufficient air flow was provided to keep the DO in the reactor above 3 mg/l through the course of the study. The temperature in the reactors was maintained at $21^{o} \pm 4^{o}$. The detention time was controlled by varying the rate of inflow of synthetic waste into the system. The feed solution was introduced into the reactor using a Milton Roy four-head pump (Model 4-C-48R). All suction, feed, and effluent lines were made of tygon tubing with glass junctions. Feed and effluent reservoirs were 5.3 gal (20-liter) Pyrex carboys.

The reactor was tested for complete mixing by filling with distilled water, and then pumping in a solution of glucose of known concentration. The COD of the effluent was determined and plotted on arithmetic paper. It was found that the experimental data followed the theoretical expression

$$x = x_o\left(1 - e^{-Dt}\right)$$

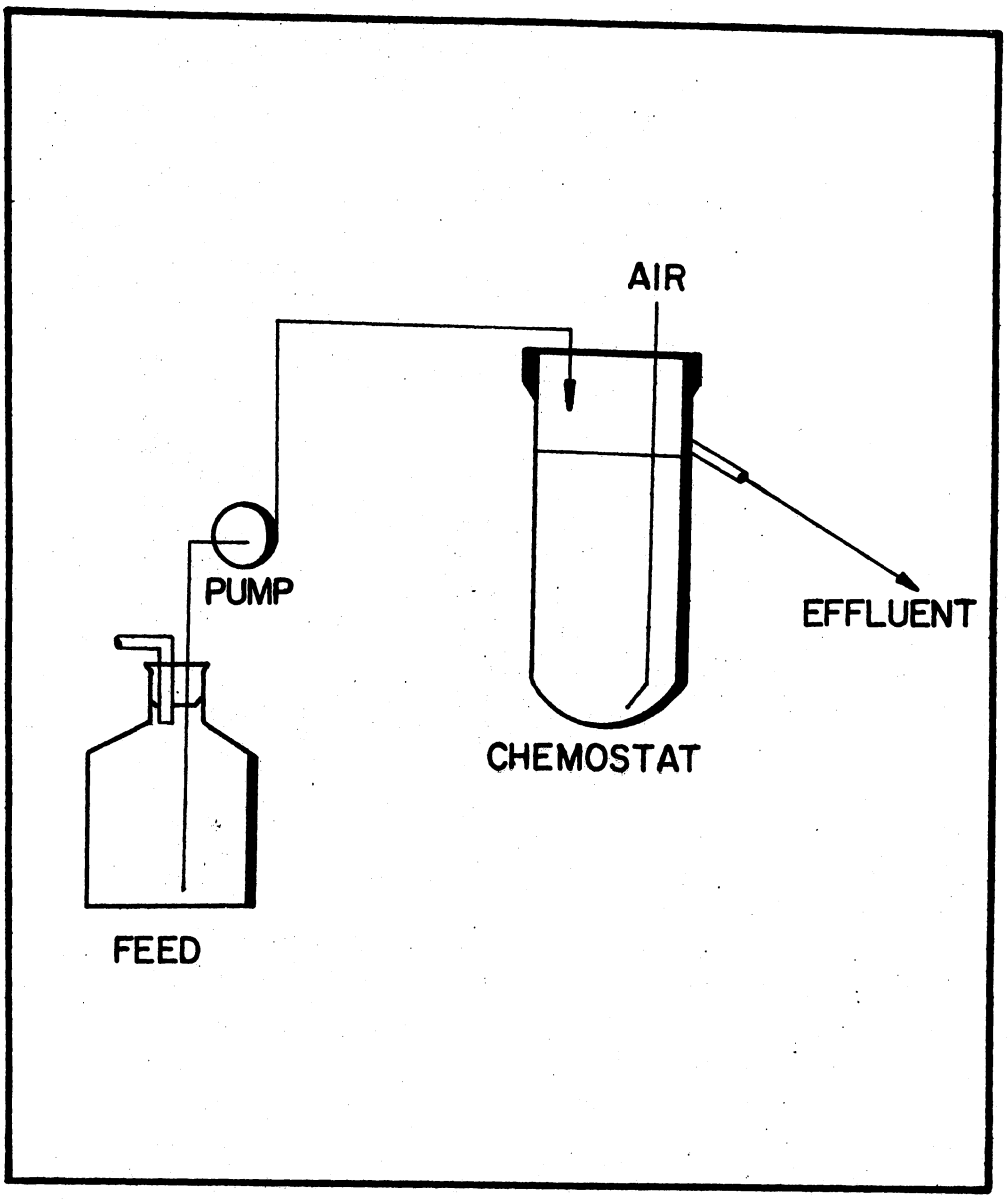
where

$x$ = concentration in the effluent at time, t

$x_o$ = concentration in the influent

$D$ = dilution rate

Figure 5. Schematic Flow Diagram of a Completely Mixed, Once-through System


AIR
PUMP
EFFLUENT
CHEMOSTAT
FEED

thus ensuring that the reactor was completely mixed for soluble substrate.

2. Composition of Synthetic Wastes

Synthetic waste using glucose as the carbon source was used in all experiments. The composition of the waste during non-shock load conditions was as follows:

TABLE II

COMPOSITION OF FEED FOR 500 mg/l GLUCOSE SUBSTRATE

| | |
|---|---|
| Glucose | 500 mg/l |
| $(NH_4)_2SO_4$ | 250 mg/l |
| $MgSO_4 \cdot 7H_2O$ | 50 mg/l |
| $FeCl_3$ | 0.25 mg/l |
| CaCl | 3.75 mg/l |
| $MnSO_4 \cdot H_2O$ | 5 mg/l |
| Phosphate buffer, 1.0 M (pH 7.2) | |
| ($KH_2PO_4$, 38.5 mg/l + $K_2HPO_4$, 125 mg/l) | 5 mg/l |
| Tap water | to volume |

Sufficient nutrients were supplied during the study to assure that glucose was the limiting growth factor.

3. Development of Steady State

Prior to every shock load, the reactor was filled with two liters of tap water and seeded with 100 ml of sewage effluent from the primary clarifier of the Stillwater, Oklahoma, municipal wastewater treatment plant. The solution was aerated and batch fed with 500 mg/l dextrose feed mixture for a periof of 24 to 48 hours. The system was then fed synthetic waste, supplied continuously from a feed reservoir. The feeding was continued for three to five days to assure that an equilibrium, steady state condition was reached. Performance of the system was assessed using COD and suspended solids analyses by periodic measurement of optical density.

4. Hydraulic Shock Load Procedure

The hydraulic shock loads were applied on the basis of changes in growth rate in accordance with hydraulic control of $\mu$ as developed in the following mass balance relationship for once-through completely mixed systems:

Rate of change in biological solids = rate of increase due to growth - rate of decrease due to outflow

$$V\frac{dx}{dt} = V\mu X - FX$$

$\frac{F}{V}$ by definition is equal to the dilution rate, D, so dividing by V and substituting D, yields:

$$\frac{dx}{dt} = \mu X - DX$$

Since $\frac{dx}{dt} = 0$ (approximately at steady state), then

$\mu = D$

where

V = volume of the reactor

F = influent flow rate

$\mu$ = specific growth rate of microorganisms

X = concentration of microorganisms

Since this study was concerned with the effect on biochemical treatment efficiency brought about by gross changes in specific growth (dilution) rate, this approximation was considered to be adequate. Other variables that possibly display a significant but lesser effect on the system (e.g., the decay coefficient) were ignored.

The hydraulic shock loads were administered with constant organic concentration in the influent stream. The shock load consisted of an increase or decrease in flow (dilution) rate from that used in the steady state. In order to generally simulate conditions found in actual treatment facilities, the shock load was administered over a five-to-seven day period, using a pulsing feed regime. The standard 500 mg/l synthetic waste was pumped for an 18-hour period at the rate of flow necessary for attaining the steady state dilution rate. The dilution rate was then changed for a period of six hours to a pre-determined shock load feed rate. The shock load was terminated after six hours and the cycle repeated. Samples (25 ml) of mixed liquor were removed periodically for suspended solids and filtrate COD analyses.

5. Quantitative Shock Load Procedure

The quantitative shock loads in this study were also of a cyclic nature. The normal 500 mg/l standard feed solution was pumped for 18

hours and an increased substrate concentration introduced into the reactor for a 6-hour period. In all quantitative shock loading experiments, the inorganic salt concentration was maintained proportional to the increase in glucose concentration. Special care was also taken to remove wall growth at frequent intervals during the course of the experiment. As in hydraulic shock load studies, the mixed liquor suspended solids and COD were analyzed on a regular basis. Complete mixing was checked frequently by comparing the optical density of the reactor and effluent solutions. Analytical determinations were made in accordance with Standard Methods (6), as outlined in Section A. 5 of this chapter.

# CHAPTER IV

# RESULTS AND DISCUSSION

The results of this investigation will be presented in two parts. The first part deals with the operation of a hydrolytically-assisted extended aeration activated sludge pilot plant under steady and shock load conditions. The second phase deals with the presentation of results on the operation of a once-through chemostat under various types of cyclic, hydraulic, and quantitative shock load conditions.

## A. Part I

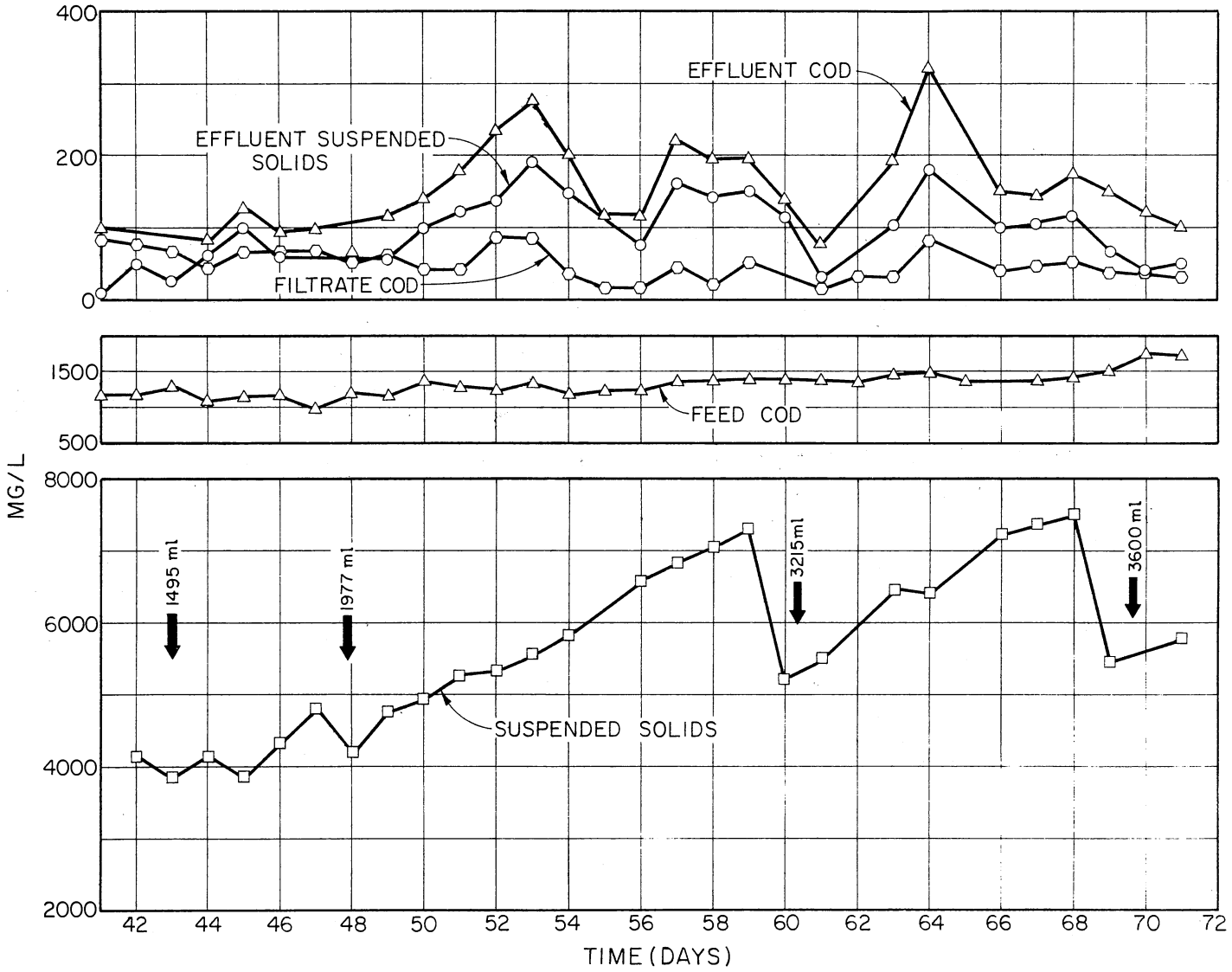
### 1. Performance of Hydrolytically-assisted Extended Aeration System Prior to Shock Loading

The daily performance of the system fed with 1000 mg/l glucose under continuous flow operation is shown on Figure 6. The system was in operation for a period of 40 days prior to compilation of data shown on Figure 6, and 78 days prior to shock loading. The "hydrolytic assist" process modification was begun on 2 December 1973, 42 days prior to the initial shock load experiments. The operation of the "hydrolytic assist" portion of the study was similar to that reported by Murthy (9), Saidi (8), and Yang (11); however, the method of wastage of solids to subsequent hydrolysis was modified. In previous studies

Figure 6. Performance of an Extended Aeration Activated Sludge System With "Hydrolytic Assist" From the 41st Day to the 71st Day of Operation

Arrows indicate the times at which mixed liquor suspended solids were removed from the reactor for hydrolysis. The details of hydrolysis are given below:

| Day of Withdrawal | Volume of MLSS Withdrawn |
|---|---|
| 43 | 1495 ml |
| 48 | 1977 ml |
| 60 | 3215 ml |
| 69 | 3600 ml |


400
200
0
EFFLUENT COD
EFFLUENT SUSPENDED SOLIDS
FILTRATE COD
1500
500
FEED COD
MG/L
8000
6000
4000
2000
1495 ml
1977 ml
3215ml
3600ml
SUSPENDED SOLIDS
42
44
46
48
50
52
54
56
58
60
62
64
66
68
70
72
TIME (DAYS)

(8)(9)(11), a pre-determined amount of sludge from the clarifier underflow (usually 900 ml/week) was "wasted" to the hydrolysis vessel. In the current study, a lower boundary level of mixed liquor suspended solids concentration was selected (approximately 4500 mg/l). A sufficient amount of mixed liquor was then removed on a weekly basis so as to maintain the base line MLSS concentration. This method required the handling of a greater volume of sludge at a more dilute concentration than the method described in previous works. As indicated in Figure 6 (see day 60), the biological solids decreased after wastage to the hydrolysis vessel. The solids then increased through the week to approximately 7000 mg/l. Upon subsequent hydrolysis, the cycle was repeated (see Figure 6). It appeared that the system reached peak solids concentration near day 75 at approximately 7500 mg/l, and that the MLSS could be expected to cycle between 5000-7500 mg/l (under the hydrolytic procedure). Pre-shock load data indicated that the filtrate COD remained, for the most part, below 50 mg/l, which further indicated an excellent biological purification efficiency of 95+ percent. Samples for determination of biological solids concentration, effluent solids concentration, effluent COD, and effluent filtrate COD were taken daily. The pilot plant was started on 22 October 1973 and was run until 28 April 1974. The results during each month of operation (except the first month) are shown in Figures 6, 7, 8, 9, and 10. Shock loads were administered during this time and are located on the graphs. The shock load responses are also presented in detail in Figures 11, 12, 13, 14, and 15. All data in this section are presented in chronological order to ensure the coordination of pre- and post-shock load data with the actual transient period.

Figure 7. Performance of an Extended Aeration Activated Sludge System With "Hydrolytic Assist" From the 72nd Day to the 102nd Day of Operation

Arrows indicate times at which mixed liquor suspended solids were removed from the reactor for hydrolysis. The details of hydrolysis are given below:

| Day of Withdrawal | Volume of MLSS Withdrawn |
|---|---|
| 76 | 3710 ml |
| 82 | 3000 ml |
| 93 | 1000 ml |
| 98 | 1300 ml |

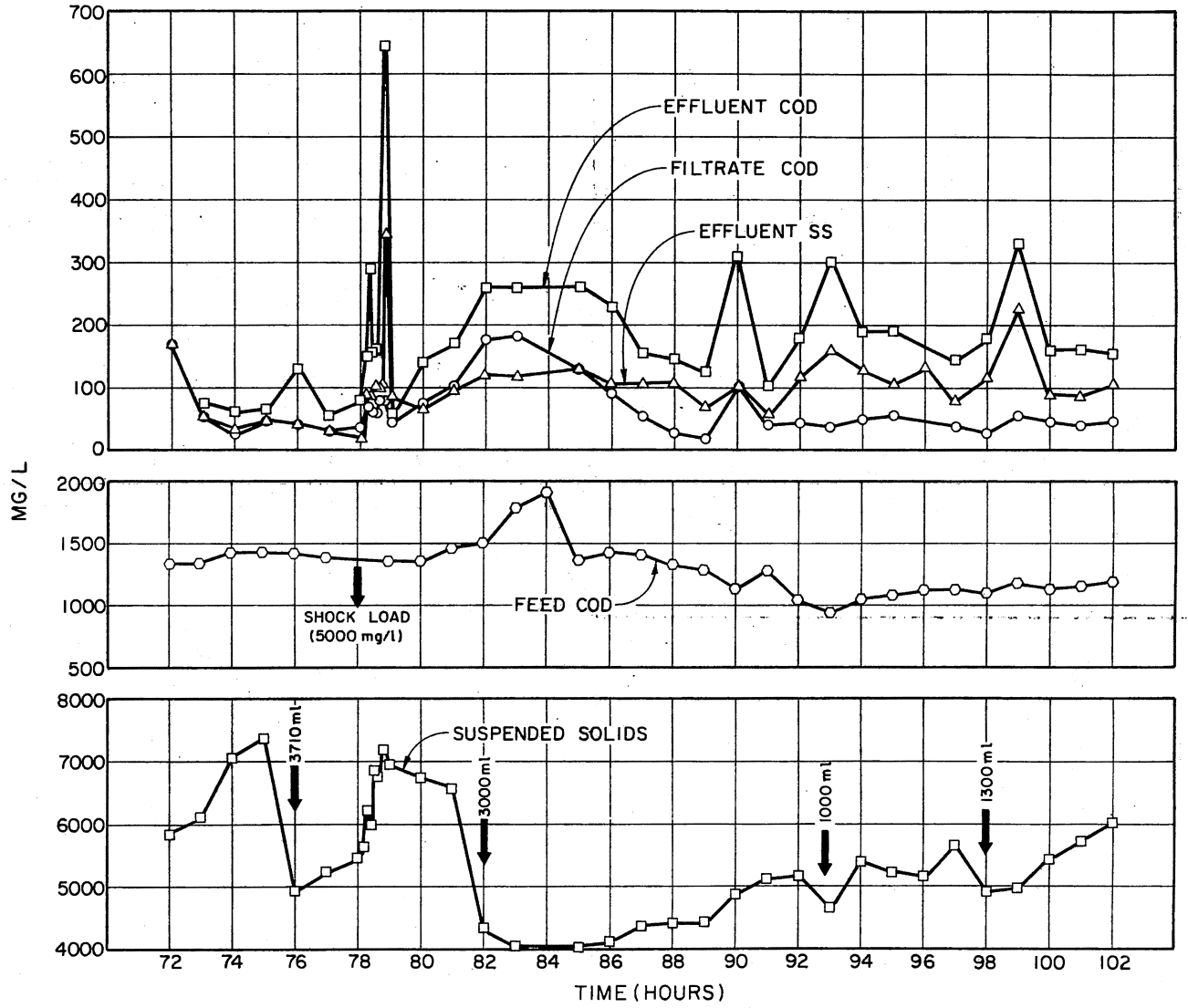

MG/L
700
600
500
400
300
200
100
0
EFFLUENT COD
FILTRATE COD
EFFLUENT SS
2000
1500
1000
500
SHOCK LOAD
(5000 mg/l)
FEED COD
8000
7000
6000
5000
4000
3710 ml
SUSPENDED SOLIDS
3000 ml
1000 ml
1300 ml
72
74
76
78
80
82
84
86
88
90
92
94
96
98
100
102
TIME (HOURS)

Figure 8. Performance of an Extended Aeration Activated Sludge System With "Hydrolytic Assist" From the 103rd Day to the 130th Day of Operation

Arrows indicate times at which mixed liquor suspended solids were removed from the reactor for hydrolysis. The details of hydrolysis are given below:

| Day of Withdrawal | Volume of MLSS Withdrawn |
|---|---|
| 112 | 3500 ml |
| 120 | 3500 ml |
| 130 | 5435 ml |

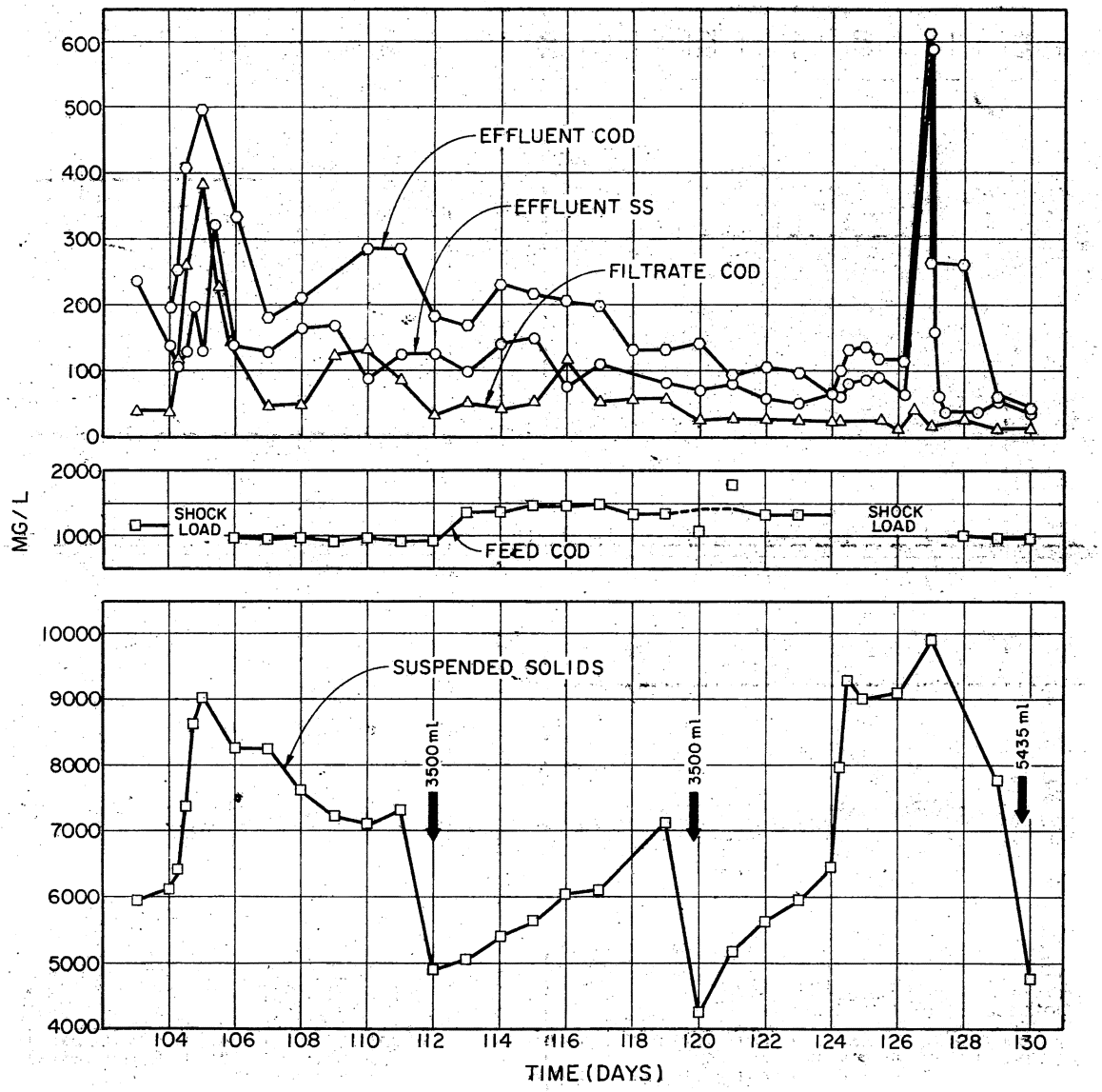

EFFLUENT COD
EFFLUENT SS
FILTRATE COD
SHOCK LOAD
FEED COD
SHOCK LOAD
SUSPENDED SOLIDS
3500 ml
3500 ml
5435 ml
MG/L
TIME (DAYS)

Figure 9. Performance of an Extended Aeration Activated Sludge System With "Hydrolytic Assist" From the 131st Day to the 159th Day of Operation

Arrows indicate times at which mixed liquor suspended solids were removed from the reactor for hydrolysis. The details of hydrolysis are given below:

| Day of Withdrawal | Volume of MLSS Withdrawn |
|---|---|
| 141 | 3700 ml |
| 149 | 4100 ml |
| 157 | 4500 ml |

From day 140 to day 148, the values for effluent suspended solids were approximately the same as those observed for filtrate COD. In the interest of clarity, the suspended solids values were omitted for this period. The majority of these values may be found on graphs showing the results of shock loads administered on days 141 and 142 (see Figures 14 and 15)

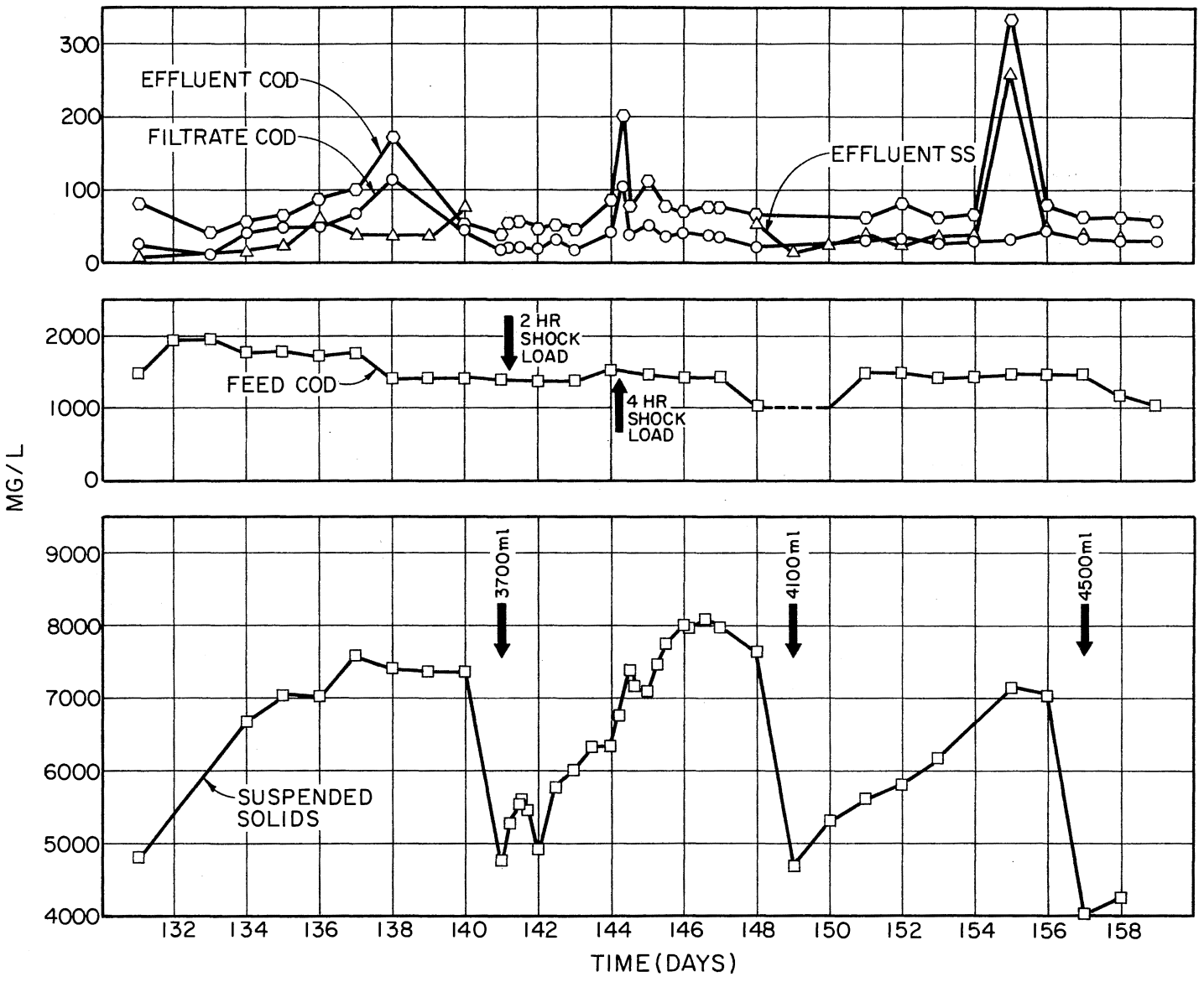
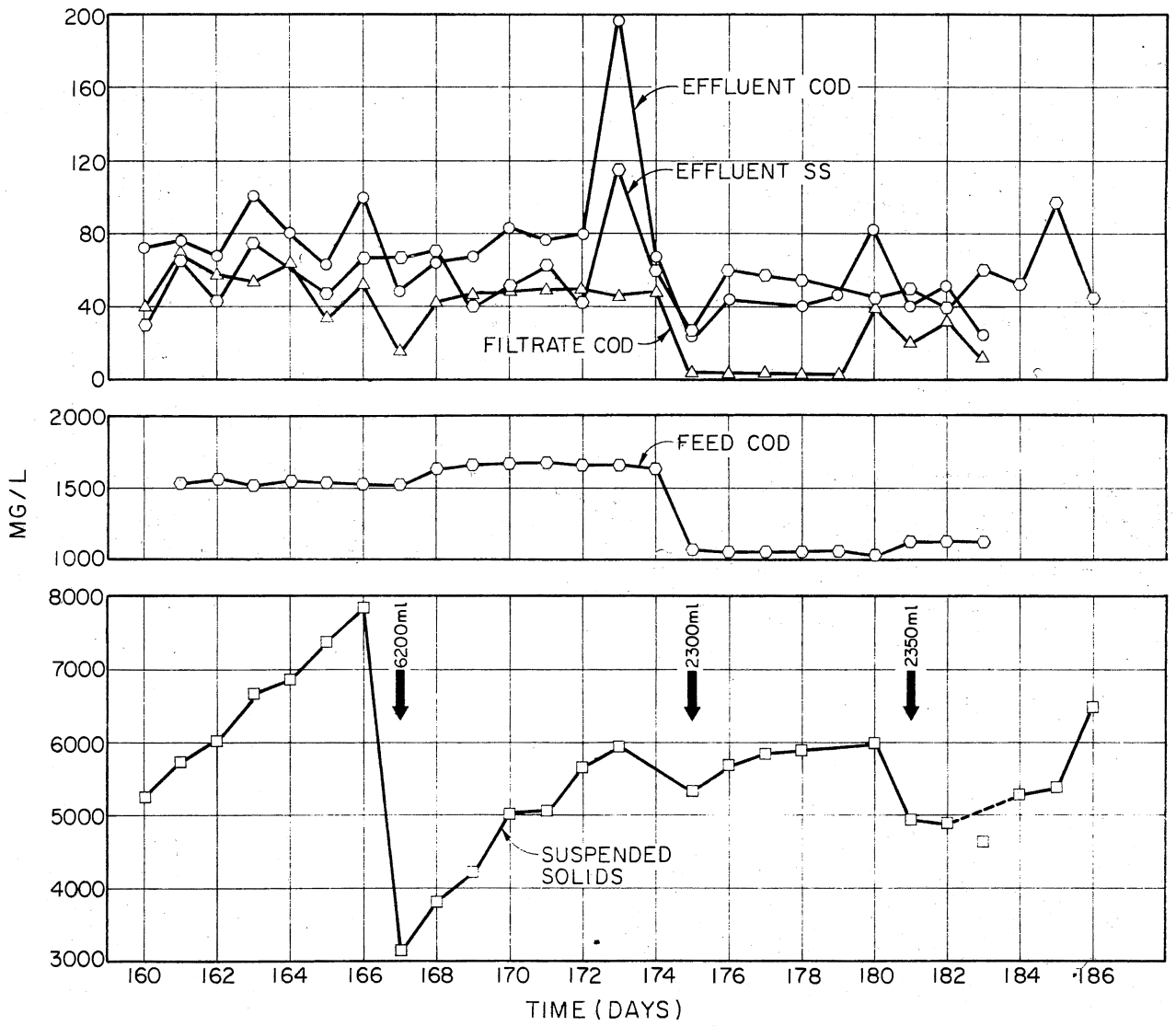

EFFLUENT COD
FILTRATE COD
EFFLUENT SS
300
200
100
0
2 HR SHOCK LOAD
FEED COD
4 HR SHOCK LOAD
2000
1000
0
MG/L
3700ml
4100ml
4500ml
SUSPENDED SOLIDS
9000
8000
7000
6000
5000
4000
132
134
136
138
140
142
144
146
148
150
152
154
156
158
TIME (DAYS)

Figure 10. Performance of an Extended Aeration Activated Sludge System With "Hydrolyic Assist" From the 160th to the 186th Day of Operation

Arrows indicate times at which mixed liquor suspended solids were removed from the reactor for hydrolysis. The details of the hydrolysis are given below:

| Day of Withdrawal | Volume of MLSS Withdrawn |
|---|---|
| 167 | 6224 ml |
| 175 | 2331 ml |
| 181 | 2280 ml |

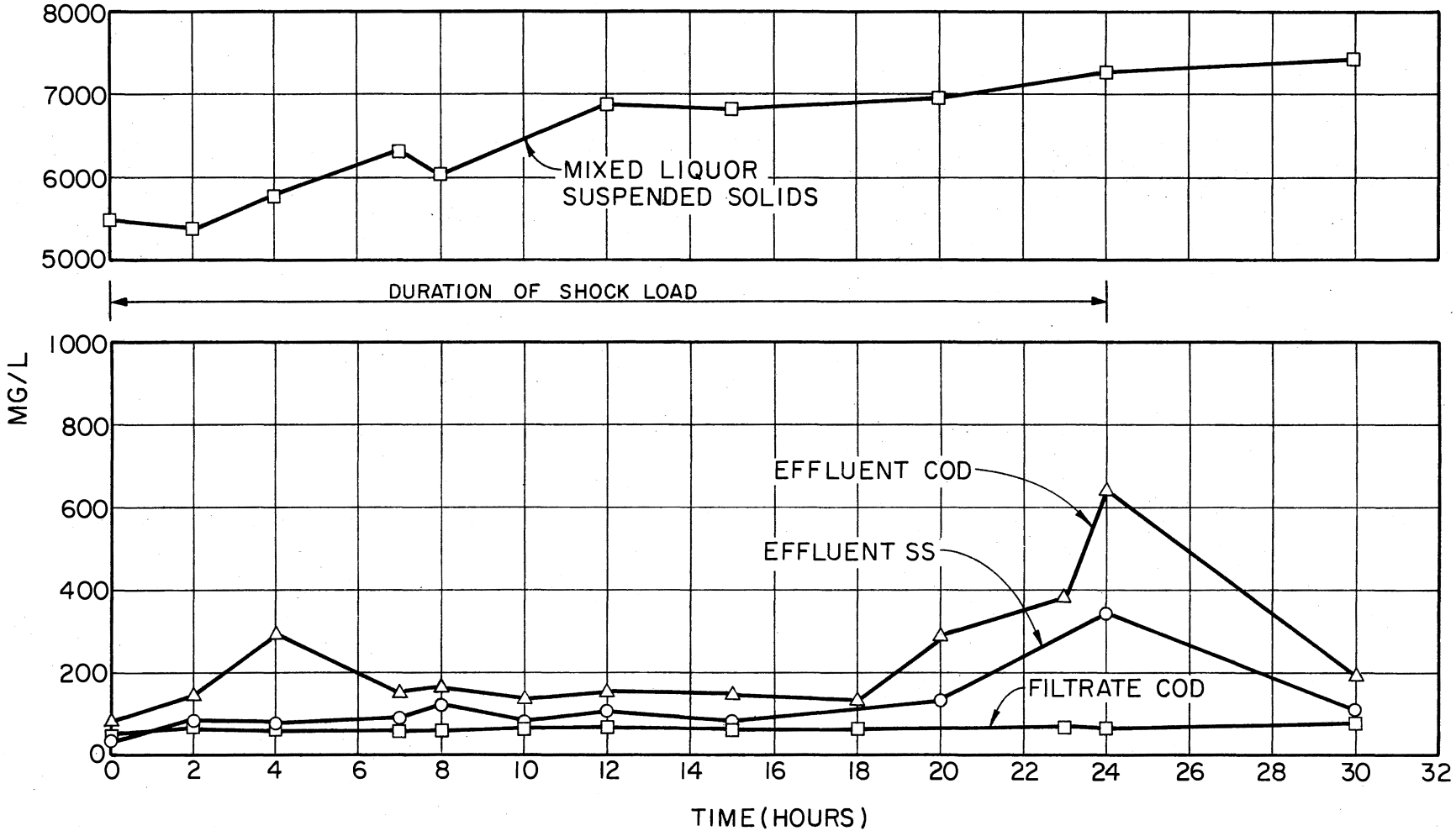

EFFLUENT COD
EFFLUENT SS
FILTRATE COD
FEED COD
SUSPENDED SOLIDS
6200ml
2300ml
2350ml
MG/L
TIME (DAYS)
200
160
120
80
40
0
2000
1500
1000
8000
7000
6000
5000
4000
3000
160
162
164
166
168
170
172
174
176
178
180
182
184
186

## 2. Response of Hydrolytically-assisted Extended Aeration Activated Sludge Pilot Plant to Various Shock Loads

Figure 11 shows the biochemical response of the system after being subjected to a five-fold increase in influent substrate concentration. The shock load occurred on the 78th day of operation (also see Figure 7) and was maintained for a period of 24 hours (one detention time). On the whole, the system responded very well. The filtrate COD of the effluent varied from a low of 48 mg/l to a high of 80 mg/l; the overall purification efficiency remained near 98.7 percent. The shock load resulted in an increase of 1750 mg/l of biological solids over a 24-hour period. After termination of the shock load (see Figure 7), the solids concentration decreased steadily and four days after the shock load was imposed, cells were taken for hydrolysis. After hydrolysis on day 82, the biological solids concentration remained depressed until day 89 (see Figure 7); then a more normal increase in solids began to occur. During this period, the normal hydrolysis schedule was suspended in order to allow the solids to reach the baseline level. The data suggest that fairly active autodigestion continued for a five- to eight-day period following reduction of biomass through hydrolysis, i.e., note the relatively steady level of solids from day 82 to day 89. In conjunction with this post-shock load behavior, a substantial (60 mg/l) increase in filtrate COD was observed. A portion of this increase substrate leakage may be attributable to the inadvertent increase in influent COD which occurred on days 83 and 84. On the other hand, if the depression in the level of biological solids

Figure 11. Response of a "Hydrolytically-assisted" Extended Aeration Activated Sludge System to an Increase in Influent Substrate Concentration From 1000 mg/l Glucose Plus Hydrolysate to 5000 mg/l Glucose Plus Hydrolysate

Total system detention time = 24 hours
Duration of shock load = 24 hours


8000
7000
6000
5000
MIXED LIQUOR
SUSPENDED SOLIDS
DURATION OF SHOCK LOAD
MG/L
1000
800
600
400
200
0
EFFLUENT COD
EFFLUENT SS
FILTRATE COD
0
2
4
6
8
10
12
14
16
18
20
22
24
26
28
30
32
TIME (HOURS)

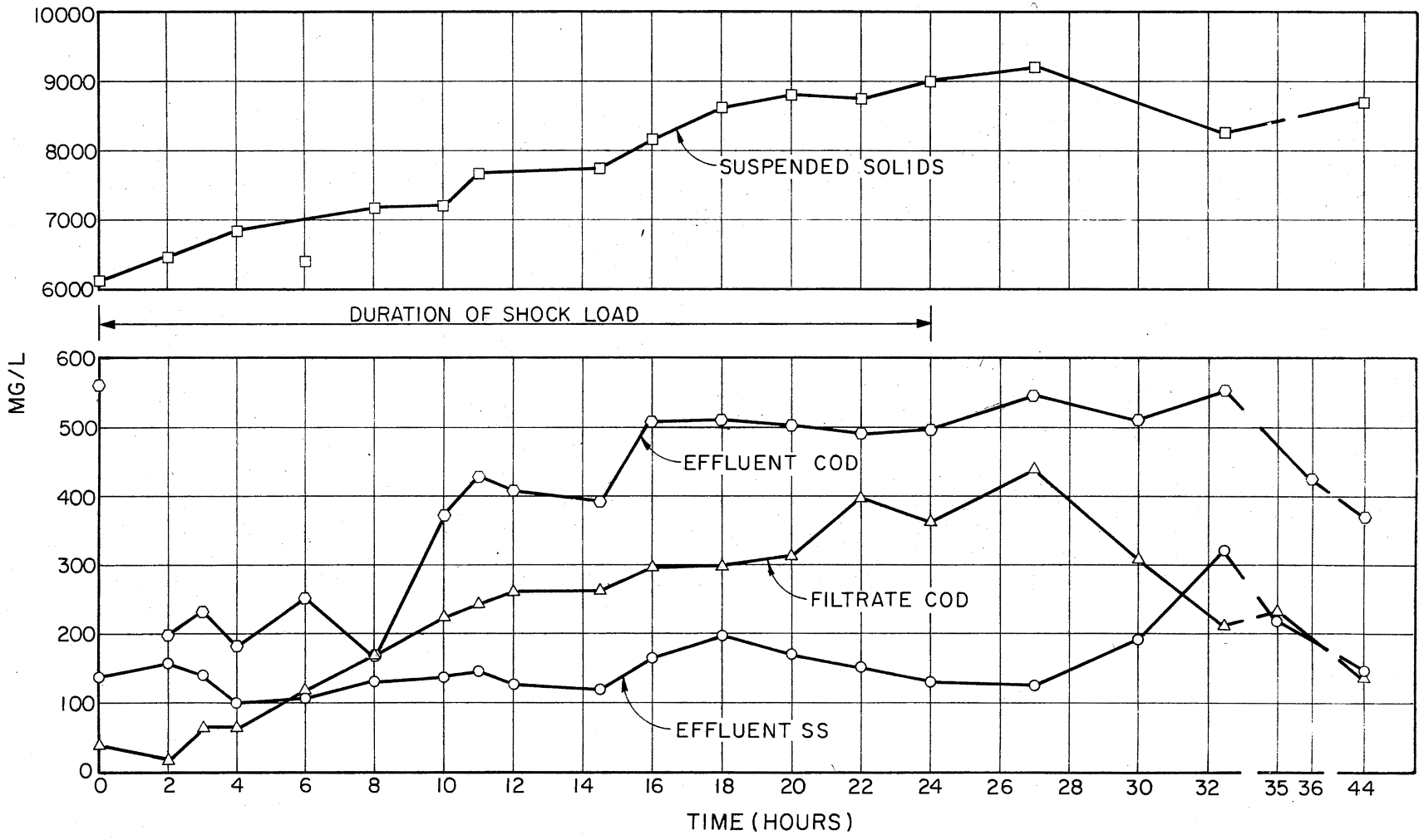
to the 4000 mg/l range was a manifestation of some decrease in metabolic activity for growth, one might expect some increased leakage of substrate. It was observed that during the final stages of this shock load experiment, the sludge, rapidly grown in response to an increase in influent substrate concentration, became lighter in color and was less dense than it was in the pre-shock condition. Microscopic examination revealed a moderate amount of protozoan activity and an almost total absence of filamentous organisms. The propagation of the "new" sludge during the shock load did tend to increase the effluent suspended solids concentration and effluent turbidity. It appeared that the rapid biological growth associated with the shock load resulted in a more dispersed sludge which was difficult to settle and there was a greater amount of unflocculated cells.

Figure 12 shows a similar quantitative shock load. At time zero, the normal feed concentration was interrupted and a new feed reservoir containing 1000 mg/l of glucose and 18.7 ml per liter of hydrolysate was placed in the system. The hydrolysate COD was equal to 2800 mg/l, giving a total concentration of shock feed of 3865 mg/l COD.

As indicated in Figure 12, the biological solids increased gradually throughout the course of the shock (24 hours). The effluent quality, as shown by the filtrate COD, exhibited a marked decrease in purification efficiency. The effluent filtrate COD increased from a low of 20 mg/l at time two hours to a high of approximately 435 mg/l, 27 hours after the shock load was begun. The highest point of substrate leakage occurred three hours after termination of the shock load when the influent substrate consisted of glucose (1000 mg/l) and the normal hydrolysate feed constituents. The apparent leakage occurring

Figure 12. Response of a "Hydrolytically-assisted" Extended Aeration Activated Sludge System to an Increase in Influent COD Resulting From an Increase in Feed Hydrolysate Concentration From 5.1 ml/l to 18.7 ml/l

| | |
|---|---|
| Duration of shock load | 24 hours |
| Shock feed concentration | 2800 mg/l COD (hydrolysate) plus 1066 mg/l COD (glucose) |


10000
9000
8000
7000
6000
SUSPENDED SOLIDS
DURATION OF SHOCK LOAD
MG/L
600
500
400
300
200
100
0
EFFLUENT COD
FILTRATE COD
EFFLUENT SS
0
2
4
6
8
10
12
14
16
18
20
22
24
26
28
30
32
35
36
44
TIME (HOURS)

at this time could be the result of a decrease in metabolic activity brought about by the shock load or by the blockage of hydrolysate usage by the now relatively high concentration of glucose in the system. The glucose blockage of multiple substrates reported by Komolrit, Gaudy, et al. (45)(46)(47)(49) could account for the continued rise in filtrate COD subsequent to the shock load. The high glucose, low hydrolysate concentration in the post-shock feed stream could block the ability of the microorganisms to utilize the hydrolysate entering the system and the hydrolysate remaining as a residual from the shock load. This blockage would not be readily apparent during normal operation, due to the high glucose to hydrolysate ratio. During normal operation, the glucose is expected to be quickly incorporated by the cells while the hydrolysate is probably utilized by a lesser fraction of species in the biomass. It may be that during the shock load shown in Figure 12 the glucose is still readily utilized, but the complex protein, lipid, and carbohydrate contained in the hydrolysate is in excess of that which can be used by the active biomass. As operation continues, three things may occur which reduce the concentration of hydrolysate in the system. First, it will naturally be diluted from the unit. Secondly, the microorganisms that are capable of metabolism of the complex substrate will increase in numbers, resulting in a decrease in filtrate COD. Third, many of the microorganisms may conceivably develop or activate the enzymes and metabolic pathways necessary for utilization of the excess substrate. The main objective in these particular studies was to determine the severity of substrate leakage due to the various shocks. Thus, only the most critical parameters were assayed. However, in view of the results shown in Figure 11, it can be seen

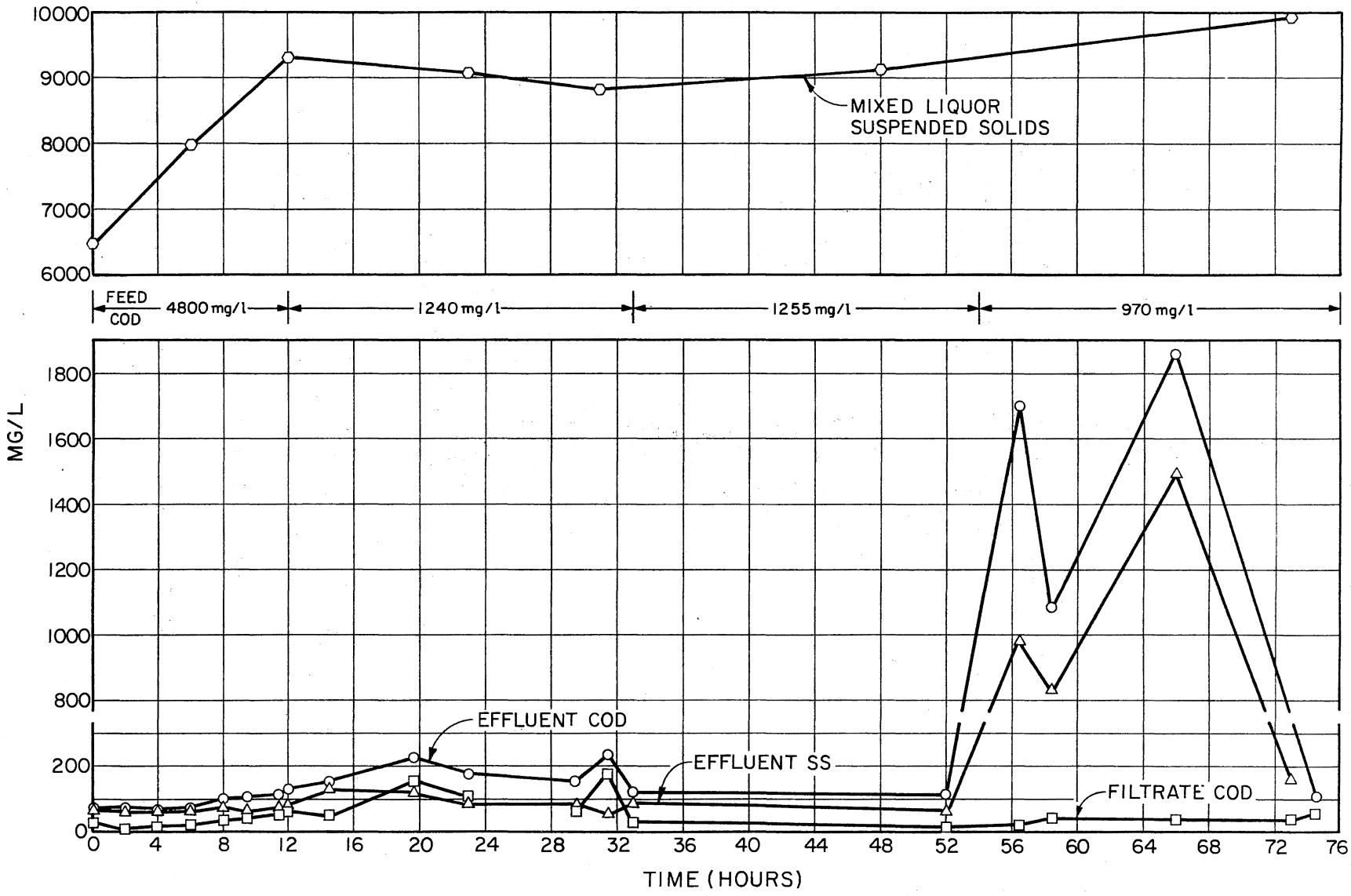
that much of the speculative analysis could have been obtained by running anthrone and/or glucostat as well as COD.

Effluent suspended solids during this experiment remained relatively constant throughout the course of the study. The effluent was turbid and found to be lacking in significant protozoan activity. The dissolved oxygen was analyzed during the experiment and was found to be maintained at approximately 5.3 mg/l in the aeration chamber and 2.2 mg/l in the clarifier. Under microscopic examination it was found that at 20 hours after the start of shock loading, a small number of filamentous organisms were observed in sludge samples. Settling characteristics were good prior to shock loading and remained unaffected during the experiment. From Figure 8, the long range effect of the shock load upon the operational efficiency of the system can be seen. After termination of the shock load, the biological solids decreased steadily in response to the decrease in influent substrate from that used in the shock feed. On day 112, the normal hydrolysis cycle was resumed. The increase in filtrate COD between 108 to 112 could be a delayed enmasse metabolic response to the shock load--or more probably--was an ecological response involving changes in predominance due to the shock load similar to the after-effect reported by Thabaraj and Gaudy (61).

Figure 13 shows the response to a rather severe shock load consisting of a four-fold increase in influent COD and a corresponding halving of detention time. The feed COD (4000 mg/l glucose plus hydrolysate) was determined to be approximately 4800 mg/l. (The shock load was continued for 12 hours or one detention time.). This shock load essentially increased the organic loading to the unit from 11.75 grams per day to approximately 50 grams per day of COD. Prior to

Figure 13. Response of a "Hydrolytically-assisted" Extended Aeration Activated Sludge System to an Increase in Influent Concentration From 1000 mg/l Glucose Plus Hydrolysate to 5000 mg/l Glucose Plus Hydrolysate at a 100 percent Increase in Flow Rate

| | |
|---|---|
| Duration of shock load | 12 hr |
| Shock feed concentration | 4800 mg/l COD |
| Total system detention time | 12 hr |

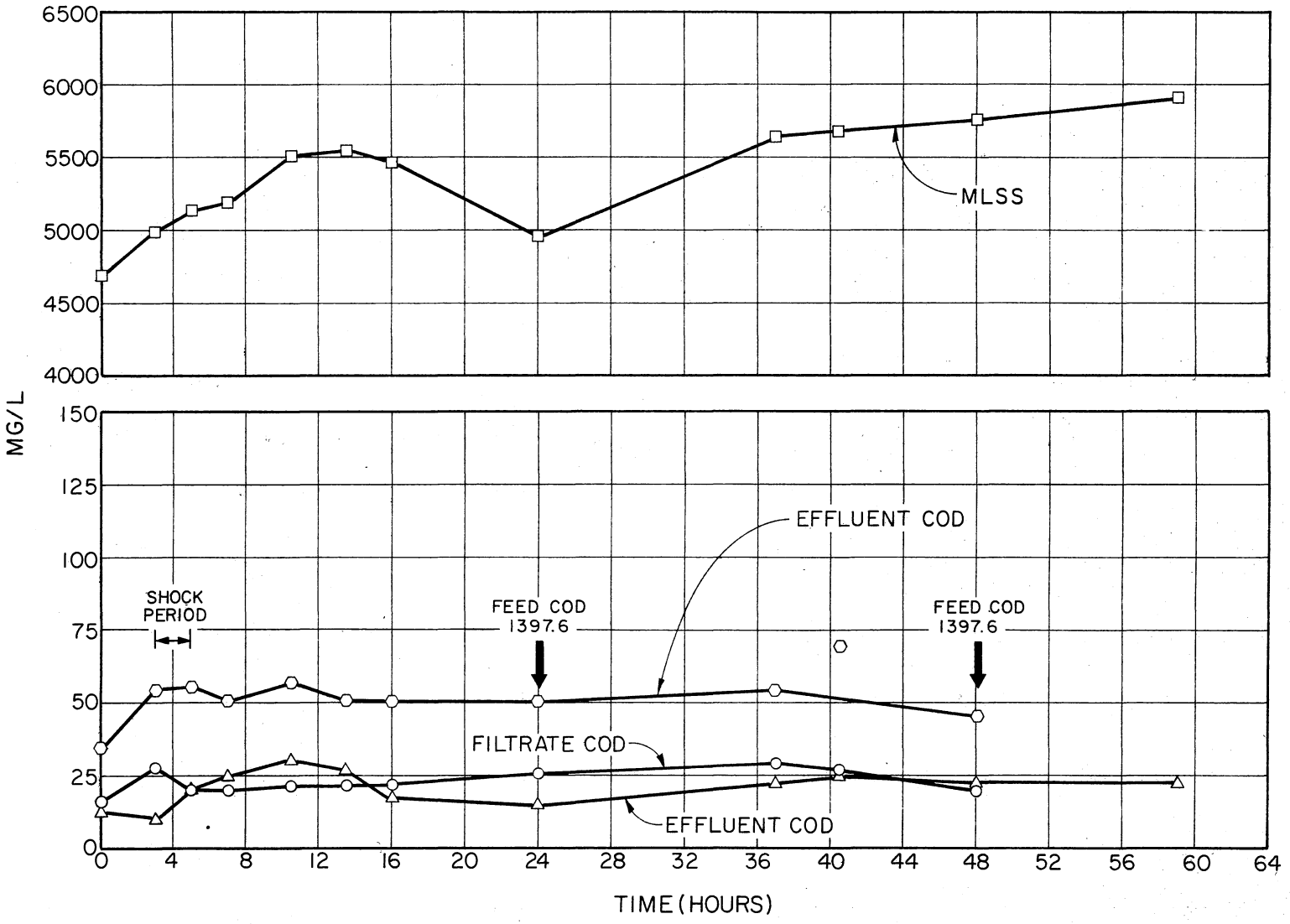

10000
9000
8000
7000
6000
MIXED LIQUOR
SUSPENDED SOLIDS
FEED
COD
4800 mg/l
1240 mg/l
1255 mg/l
970 mg/l
MG/L
1800
1600
1400
1200
1000
800
200
0
EFFLUENT COD
EFFLUENT SS
FILTRATE COD
0
4
8
12
16
20
24
28
32
36
40
44
48
52
56
60
64
68
72
76
TIME (HOURS)

shock loading, the sludge exhibited a moderate number of protozoa and a noticeable lack of filamentous growth. Initial solids were high at approximately 6500 mg/l. As can be seen in Figure 13, an immediate rise in biological solids maintained the filtrate COD at excellent levels, hence, successfully accommodating the shock load. Purification efficiency remained near 97 percent during the shock load and was maintained at 94 percent after shock loading.

At 52 hours after applying the shock load, the effluent suspended solids increased tremendously. The sludge, at approximately 9000 mg/l, exceeded the capacity of the system to provide effective clarification. During the shock load, the rapid proliferation of new biomass resulted in a light brown bulky growth, similar to that described in the discussion of Figure 11. It was concluded from the experiment that the activated sludge responded extremely well to the shock load. In addition, the loss of effluent suspended solids over the weir would be easily preventable with separate clarification or chemical assist. All biological solids passing in the effluent were collected, hydrolyzed, and fed back to the system along with the hydrolysate obtained on day 130 (see Figure 8).

Figure 14 shows the response of an extended aeration activated sludge pilot plant to a severe shock load of short duration. The normal daily feed (glucose plus hydrolysate, COD = 1410 mg/l) was supplemented with 9750 mg/l (COD) of glucose at an increased feed rate of twice normal. All nutrients were provided to assure that carbon was the limiting growth factor. The shock load was introduced for a period of two hours, at which time 8570 mg of COD entered the system. This is in direct comparison with the 1083 mg that were entering the system

Figure 14. Response of a "Hydrolytically-assisted" Extended Aeration Activated Sludge System to a 2-hour Shock Load Consisting of an Influent Substrate Increase From 1000 mg/l Glucose Plus Hydrolysate to 11,000 mg/l Glucose Plus Hydrolysate and a Doubling Flow Rate

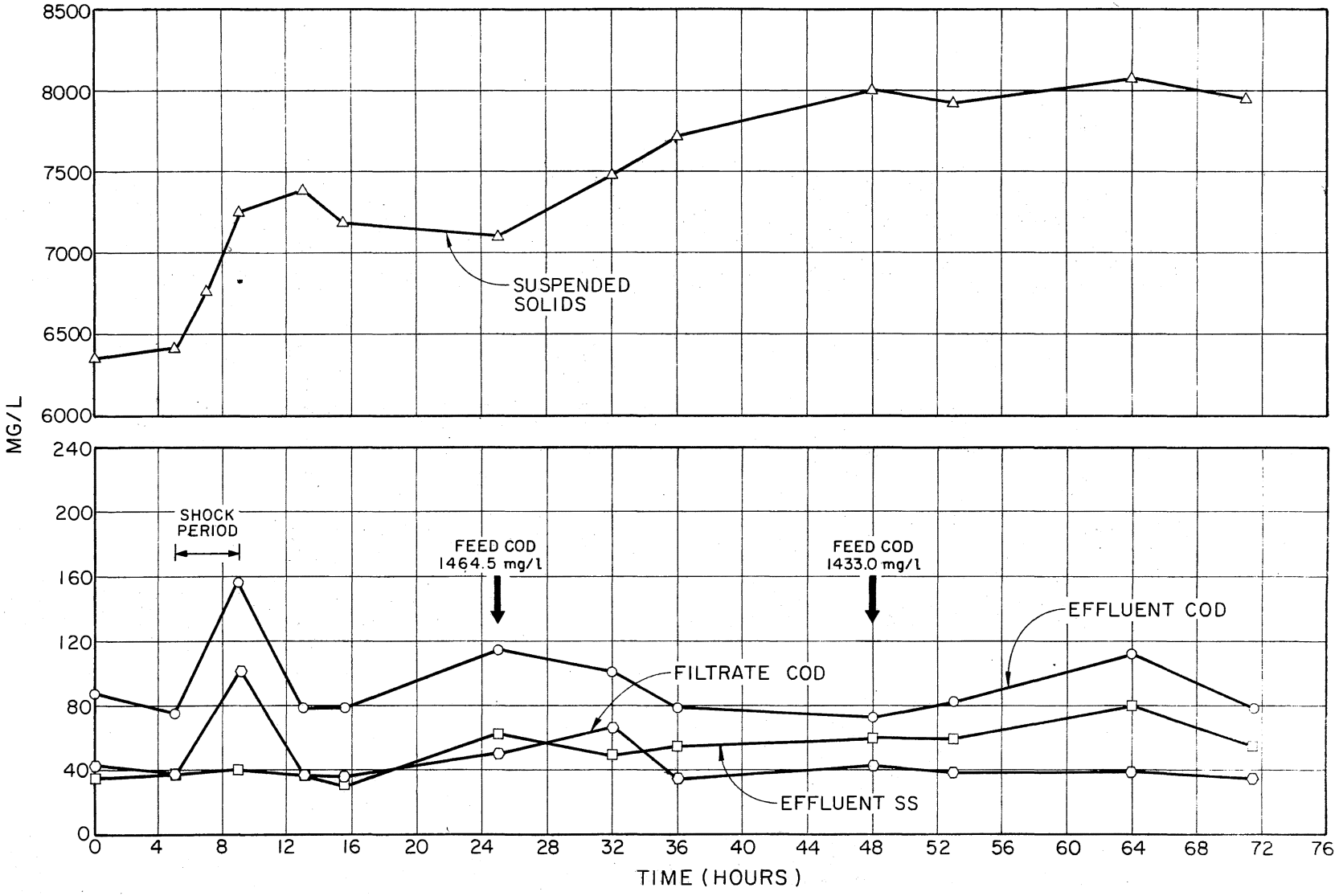

6500
6000
5500
5000
4500
4000
MLSS
150
125
100
75
50
25
0
MG/L
SHOCK PERIOD
FEED COD 1397.6
FEED COD 1397.6
EFFLUENT COD
FILTRATE COD
EFFLUENT COD
0
4
8
12
16
20
24
28
32
36
40
44
48
52
56
60
64
TIME (HOURS)

prior to the shock load. During the shock load, the biological solids increased significantly from approximately 5000 to 5500 mg/l. As can be seen by the filtrate COD, the effluent quality remained good and the shock load was accommodated successfully by the system. The rise in effluent suspended solids from hour three to hour 16 was the result of increased turbidity brought about by motile and dispersed growth that was found to be characteristic of a rapid transient rise in biological solids.

Figure 15 shows a similar shock load of slightly longer duration. As before, the detention time during the shock load is halved and the normal feed supplemented with approximately 10,000 mg/l of dextrose. This experiment and the previous one represent slug dose-types of shock loads that may enter on-line treatment facilities as the result of accidental spills, industrial malfunctions or dumping of batch processes. The total amount of COD that entered the unit during the shock load was 18,057 mg over a 4-hour period. The shock load caused an immediate rise in biological solids from 6400 mg/l to 7400 mg/l. The solids increase was not, however, sufficient to prevent a substantial leakage of filtrate COD from 40 to approximately 100 mg/l. The leakage was of short duration, and purification efficiency returned to nearly 97 percent. Effluent suspended solids and effluent COD remained relatively constant after the shock load period. The biological solids were decreased slightly after termination of the shock load, and then resumed a gradual accumulation pattern.

Figure 10 shows the daily operational performance of the system from a period after the last shock load until the termination of the study on 27 April 1974. In all, the unit was in operation for 187 days.

Figure 15. Response of a "Hydrolytically-assisted" Extended Aeration Activated Sludge System to a 4-hour Shock Load Consisting of an Increase in Influent Substrate Concentration From 1000 mg/l Glucose Plus Hydrolysate to 11,000 mg/l Glucose Plus Hydrolysate and a Doubling of Flow Rate


8500
8000
7500
7000
6500
6000
SUSPENDED SOLIDS
MG/L
240
200
160
120
80
40
0
SHOCK PERIOD
FEED COD 1464.5 mg/l
FEED COD 1433.0 mg/l
EFFLUENT COD
FILTRATE COD
EFFLUENT SS
0 4 8 12 16 20 24 28 32 36 40 44 48 52 56 60 64 68 72 76
TIME (HOURS)

During this period, the extended aeration activated sludge system operated extremely well under normal daily operation and several types of severe external environmental changes. It was found that the pilot plant was easily operable and that the "hydrolytic assist" was a reliable and efficient engineering tool for the control of biological solids concentrations without use of conventional sludge disposal methods. The system was found to be able to accommodate a five-fold increase in influent substrate concentration with little effect on purification efficiency.

## B. Part II

### 1. Response of a Once-through System to Various Types of Cyclic Shock Loads

This section deals with the effect of hydraulic and quantitative shock loads upon a heterogeneous microbial population in a completely mixed once-through reactor. As stated in Chapter III, the microorganisms used in this phase of the study were obtained from seed from the primary effluent of the Stillwater, Oklahoma, wastewater treatment plant. Care was taken throughout the study to ensure that a heterogeneous population of microorganisms growing at "steady state" was developed prior to shock loading. Frequent spot checks of reactor and effluent solids were made, using optical density, to assure complete mixing of the reactor. The shock load was administered in a cyclic manner, consisting of an 18-hour period under the normal pre-shock condition and six hours of shock loading per day. In all figures, at least 18 hours of steady state data are shown prior to administration

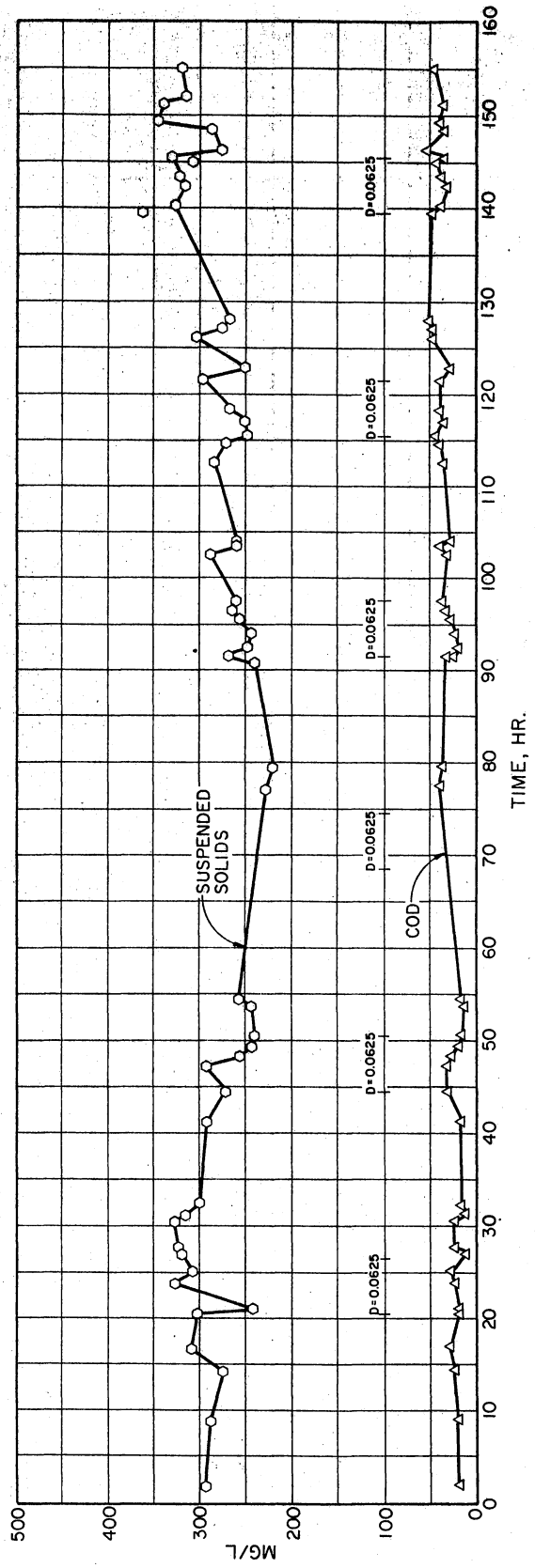
of the first shock in the cycle. It is emphasized, however, that all units were run for a period of 4-5 days in the steady state prior to beginning a shock loading series. The hydraulic shock load portion of the study will be presented initially, followed by the quantitative shock loads in increasing order of severity (750 mg/l, 1000 mg/l, and 2000 mg/l). The quantitative shock loads at each concentration were operated in duplicate with one reactor dilution rate set to correspond to 0.0625 $hr^{-1}$ ($\bar{t}$ = 16 hours), and the other set to 0.125 $hr^{-1}$ ($\bar{t}$ = 8 hours). Both reactors were operated in parallel, utilizing normal substrate and shock feed from the same feed reservoir. The changes imposed on either the dilution rate or influent substrate concentration and the duration of the shock load, are indicated on the graphs. In all cases, the shock load was begun at a specified time (e.g., 11:00 A. M.) on the first and every succeeding day of the experiment; however, slight deviations did occur occasionally, due to unavoidable scheduling conflicts and brief mechanical malfunctions.

Figure 16 shows the response of a once-through chemostat system to a periodic shock load consisting of a decrease in dilution rate from D = 0.125 $hr^{-1}$ to D = 0.0625 $hr^{-1}$. This corresponds to a change in detention time from eight to 16 hours. The system was operated at steady state at a dilution rate of 0.125 $hr^{-1}$ (8-hour detention time) prior to shock loading. As shown in Figure 16, the system was shocked for a 6-hour period on six successive days. During the experiment, the influent substrate concentration was maintained at approximately 500 mg/l dextrose. It can be seen that the system responded well to the 100 percent decrease in flow rate. There was virtually no leakage of substrate during the shock load, and purification efficiency throughout

Figure 16. Response of a Completely Mixed Once-through System to a Periodic Decrease in Dilution Rate From 0.125 $hr^{-1}$ to 0.0625 $hr^{-1}$ (feed concentration is maintained constant at 500 mg/l dextrose)

D = 0.125 $hr^{-1}$ for 18 hours/day

D = 0.0625 $hr^{-1}$ for 6 hours/day


MG/L
TIME, HR.
SUSPENDED SOLIDS
COD
p=0.0625
p=0.0625
p=0.0625
p=0.0625
p=0.0625
0
100
200
300
400
500
10
20
30
40
50
60
70
80
90
100
110
120
130
140
150
160

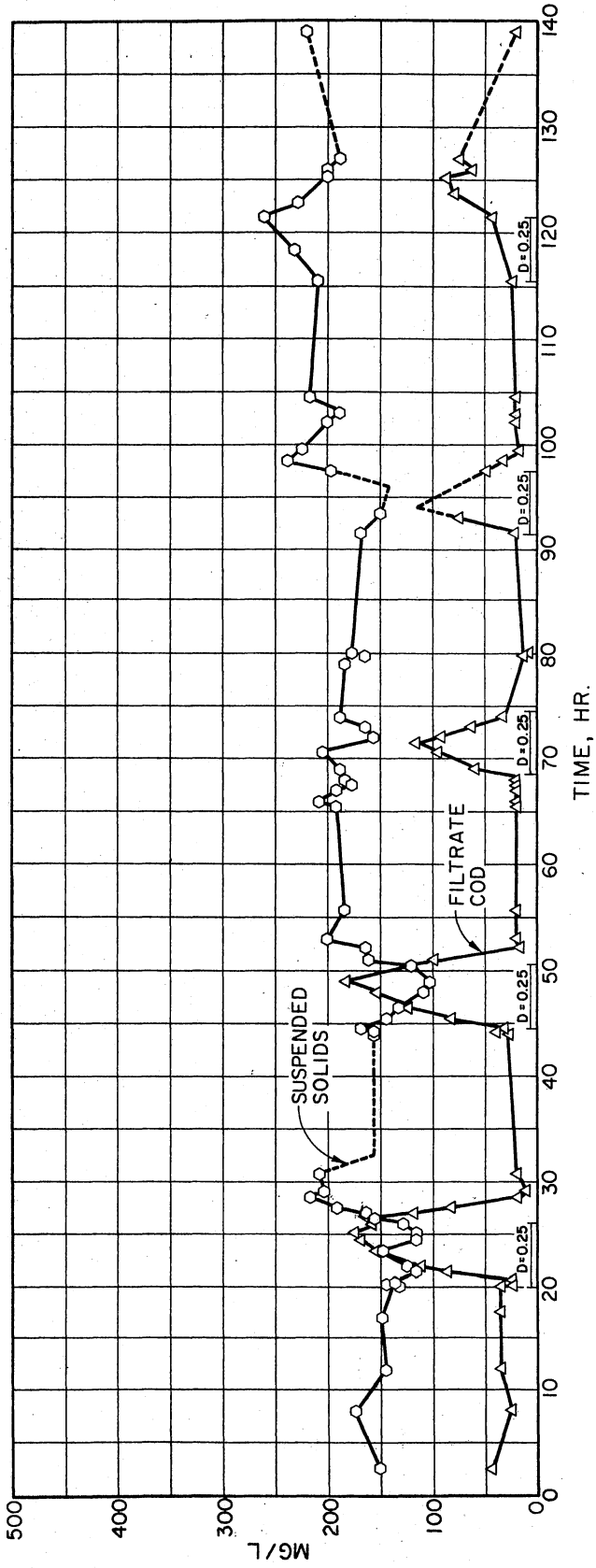
the experiment was maintained at above 90 percent. Biological solids were found to fluctuate slightly in response to the cyclic hydraulic shock load. During every shock load period, a moderate increase in biological solids occurred. These results suggest that the biological population, particularly those species with slow optimum growth rates, increased in number during the shock load. When normal operation was resumed and the flow rate increased, that portion of the population that could not acclimate quickly to a higher specific growth rate ($\mu$ = 0.125 $hr^{-1}$) was diluted from the system. In general, this shock load caused little or no changes in effluent substrate concentration and in this regard, the results are in general agreement with those reported by George (12) on step changes in dilution rate from D = 0.125 $hr^{-1}$ to D = 0.0625 $hr^{-1}$.

Figure 17 shows a series of shock loads consisting of periodic increases in dilution rate from D = 0.0625 $hr^{-1}$ to 0.25 $hr^{-1}$. As can be seen from Figure 17, this series of shock loads caused a rather severe disruption in the purification efficiency of the system. Prior to shock loading, the system was grown at steady state with a growth rate of $\mu$ = 0.0625 $hr^{-1}$; the feed concentration during the study was maintained at 500 mg/l dextrose. In all, five individual, daily shock loads were made during the experiment. The first two shock loads, beginning at hours 20 and 44.5, resulted in similar transient responses. As the shock load was imposed, the majority of the microorganisms in the reactor were unable to grow at a rate equal to or greater than the new dilution rate. As the dilution rate exceeded the growth rate, the microorganisms were lost from the system and the biological solids concentration decreased. This decrease in biological solids resulted in a

Figure 17. Response of a Completely Mixed Once-through System to a Periodic Increase in Dilution Rate From 0.0625 $hr^{-1}$ to 0.25 $hr^{-1}$ (feed concentration maintained constant at 500 mg/l dextrose)

D = 0.0625 $hr^{-1}$ for 18 hours/day

D = 0.25 $hr^{-1}$ for 6 hours/day


MG/L
0
100
200
300
400
500
TIME, HR.
0
10
20
30
40
50
60
70
80
90
100
110
120
130
140
D=0.25
D=0.25
D=0.25
D=0.25
D=0.25
SUSPENDED SOLIDS
FILTRATE COD

large scale increases in filtrate COD (from approximately 25 mg/l to 175 mg/l). Immediately following the termination of the shock load, the microorganisms left in the system responded immediately to the excess substrate in the reactor and increased substantially from approximately 125 mg/l to 200 mg/l. The effluent COD was returned to pre-shock levels in from two to three hours after termination of the shock load. Between hours 55 and 65, a change in predominance of microorganisms was observed in the reactor. During this period, the color of the mixed liquor in the system changed from white to light green. Also, the biomass concentration was slightly higher during the second cycle, indicating a change in cell yield. This suggests that the shock loading previous to hour 55 may have selectively eliminated a significant portion of the microbial population which could be maintained only at the low growth rate. Thus, the shock loading caused a change in the predominance of microorganisms in the heterogeneous population. During the third shock load (hours 68.5 to 74.5) it may be seen that although some decrease in biological solids did occur, the amount of leakage of filtrate COD was approximately one-third less than that observed in the previous two shock load periods. The somewhat higher concentration of biological solids and the probability that they were more nearly capable of surviving at high growth rates (because of the previous shock) may have been the cause for the reduction in the amount of substrate leakage from the reactor during this shock load. The fourth shock load in the series was insufficiently defined by the available data points. The hypothetical results, indicated by the dotted lines, are merely "educated guesses" bases on observations of previous results and the existing post-shock load data.

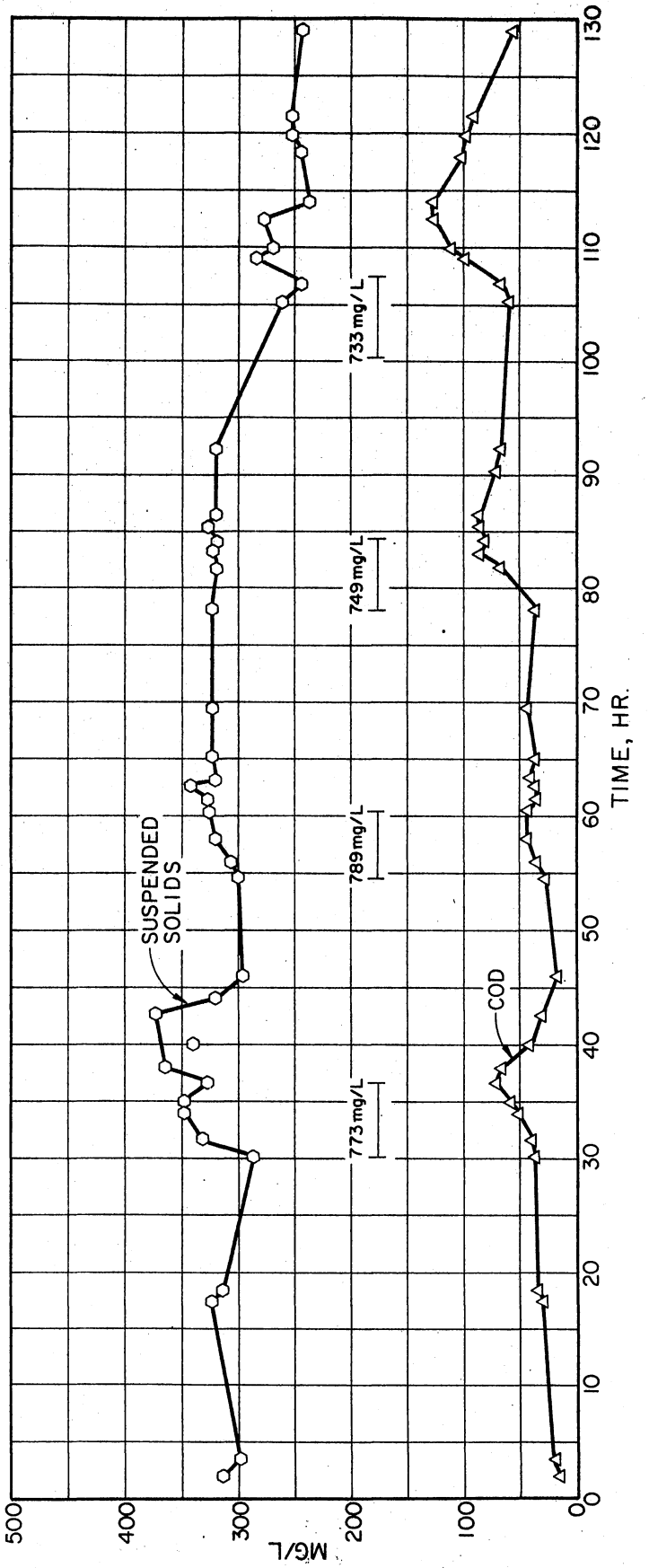
The fifth and final shock load (hour 120.5 to hour 126.5) shows an even greater decrease in substrate leakage, resulting from the shock load. Filtrate COD rose from 20 mg/l to 85 mg/l, approximately one-half the leakage observed in the initial shock load of the experiment. Furthermore, the solid dilute-out and substrate leakage were delayed until removal of the shock. The results suggest that it is possible for a biological system to become acclimated to cyclic hydraulic shock loads, possibly by selective removal of the lesser tolerant species of microorganisms. The predominant microorganisms remaining can be assumed to have a greater capacity for accommodation of the shock load; e.g., they are capable of growing at rates near or greater than the shock load dilution rate and can shift growth rate rapidly in response to the environmental change. The results of this experiment are in general agreement with those found by Antone (62) in a similar study of longer duration. The present experiment was terminated prematurely due to the presence of a great number of large white colonies in the reactor, which appeared after time 140 hours (Figure 17). The large colonies were difficult to mix completely and caused high variability in suspended solids data.

Figure 18 shows the response of a completely mixed system to a change in influent COD from 500 mg/l to 750 mg/l. The quantitative shock load was administered for a 6-hour period on four successive days at a dilution rate of 0.0625 $hr^{-1}$ (16-hour detention time). As a result of the first and fourth shock loads (hours 30 to 45 and 105 to 115), the biological solids showed a relatively high degree of fluctuation. In response to the quantitative shock load, the biological solids increased moderately (8 to 14 percent). This increase in

Figure 18. Response of a Completely Mixed Once-through System to a Periodic Increase in Influent Substrate Concentration From 500 mg/l Dextrose to 750 mg/l Dextrose at D = 0.0625 $hr^{-1}$

$S_i$ = 500 mg/l dextrose for 18 hours

$S_i$ = 750 mg/l dextrose for 6 hours


SUSPENDED SOLIDS
COD
773 mg/L
789 mg/L
749 mg/L
733 mg/L
MG/L
TIME, HR.
0
100
200
300
400
500
10
20
30
40
50
60
70
80
90
100
110
120
130

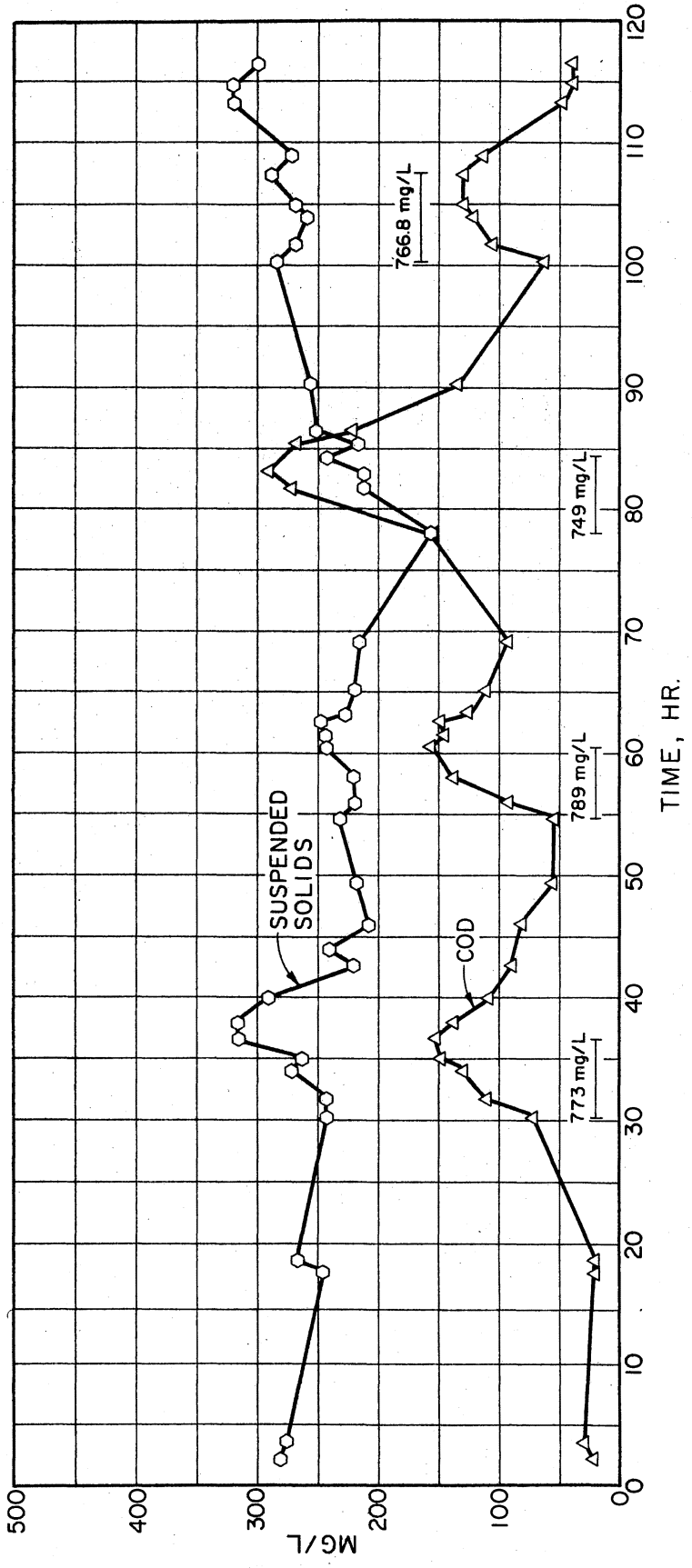
biomass was not, however, sufficient to prevent a 50 to 55 percent increase in filtrate COD in the system. During the second shock load (hours 54.5 to 60.5), the biological solids rose gradually from 300 to 345 mg/l, and filtrate COD increased slightly, from 25 to just above 40 mg/l. The purification efficiency during this pulse shock decreased only slightly, from 95 percent to 94 percent. The biological solids during the third shock load (hours 78 to 84.5) remained constant throughout the period of shock loading. The filtrate COD increased from 35 mg/l at hour 78, to 85 mg/l at hour 83. In general, few firm conclusions can be drawn from these data. However, with the possible exception of the first shock load, a trend could have been developing in the system. From pulse two to pulse four, a decrease in biological solids concentration and a corresponding increase in substrate leakage was observed. The variation of the separate responses observed during each pulse shock load indicates that the shock loading had an effect upon either the substrate utilization efficiency of the cells or predominance of microorganisms found in the system. The first pulse shock load resulted in a sharp fluctuation in biological solids concentration, while in shock three, the solids level remained steady throughout the sampling period. In both cases, however, the amount of substrate leakage was approximately the same. An explanation of this observation could be due to a change in cell yield or removal of substrate for use as internal storage products.

Figure 19 shows the response of a quantitative shock load applied in an identical manner as the one previously reported. The microorganisms were grown, however, at a higher dilution rate of 0.125 $hr^{-1}$ (8-hour detention time). As in the previous shock load study, the system

Figure 19. Response of a Completely Mixed Once-through System to a Periodic Increase in Influent Substrate Concentration From 500 mg/l Dextrose to 750 mg/l Dextrose at D = 0.125 $hr^{-1}$

$S_i$ = 500 mg/l dextrose for 18 hours

$S_i$ = 750 mg/l dextrose for 6 hours


SUSPENDED SOLIDS
COD
766.8 mg/L
749 mg/L
789 mg/L
773 mg/L
MG/L
TIME, HR.
0
100
200
300
400
500
10
20
30
40
50
60
70
80
90
100
110
120

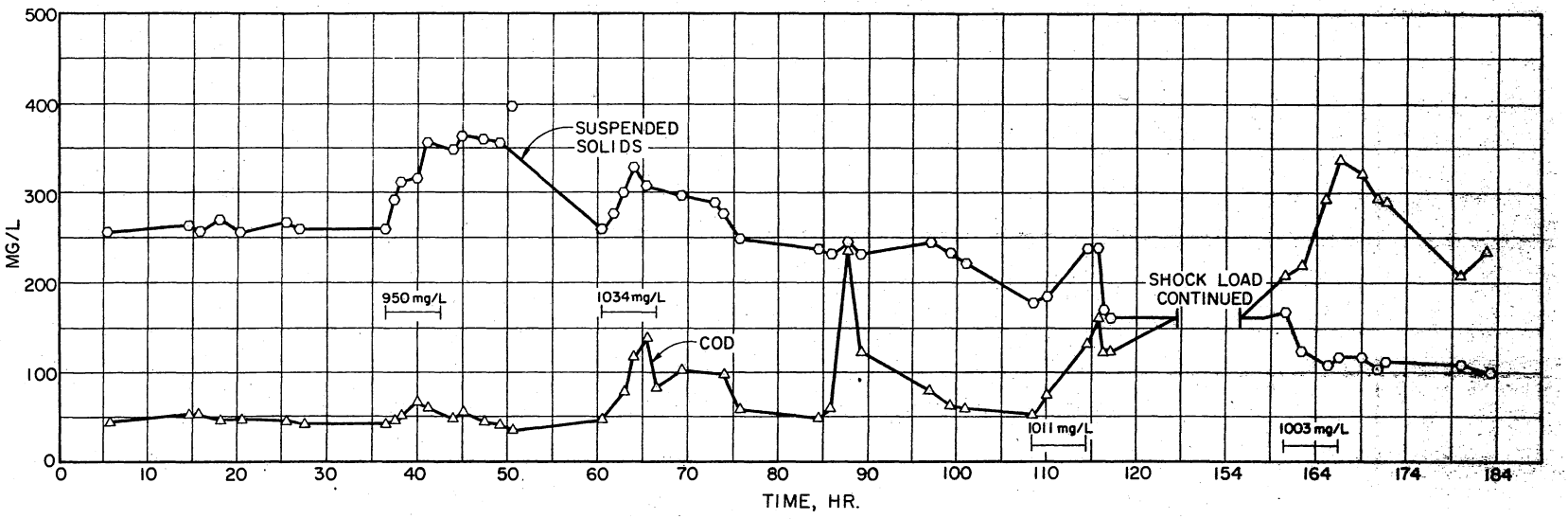
was shocked from 500 mg/l dextrose to 750 mg/l dextrose. The shock was applied on four separate days for a 6-hour period per day. It can be seen from Figure 19 that the system responded less favorably than did the unit shocked similarly at a dilution rate of 0.0625 $hr^{-1}$. The first through the third shocks did, however, result in biomass responses very similar to those previously reported (Figure 18). The initial shock load caused a transient rise in biological solids from 240 mg/l to 315 mg/l. The second and third pulse shock load exhibited the same lack of fluctuation or immediate biomass response which characterized the second and third pulse shocks of the previous study. However, the purification efficiency was severely disrupted at the higher dilution rate. From hour 78 to hour 115, a general rise in biological solids concentration was observed in the reactor. The higher concentration of biological solids reduced the amount of substrate leaking from the system during the final pulse shock load. The data obtained from both 500 mg/l and 750 mg/l glucose shock loads (D = 0.0625 and D = 0.125 $hr^{-1}$) seems to indicate that the initial or first shock load response is similar to systems subjected to step changes in influent substrate concentration. The succeeding shock loads cause responses that are made more complex because of the previous disruption of the steady state. That is to say, that before the system can reach a new steady state following a shock load, it is again shock loaded. The additional disruption brought about by the consecutive shock loads is responsible for the variation in response from shock load to shock load. This variation, as indicated in the discussion on Figure 18, could be the result of changes in the metabolic response time of the cells or of changes in predominance in the heterogeneous populations.

Figure 20 shows the response of a completely mixed once-through system to a periodic 100 percent increase in influent feed COD. For six hours on each of six consecutive days, the concentration of dextrose in the feed stream was increased from 500 mg/l to 1000 mg/l of dextrose. The dilution rate was maintained at 0.0625 $hr^{-1}$. Figure 20 shows that during the first shock load, the system accommodated the quantitative increase in substrate very nicely. The filtrate COD rose slightly from 40 to 65 mg/l. During the second and third puse shock, the leakage in the effluent increased to 135 mg/l and 235 mg/l, respectively. From time 45 hours until the termination of the experiment, a general trend of decreasing biological solids and increasing filtrate COD was in evidence. During the fourth shock load, it appeared that a moderate response toward accommodation of the shock load occurred between hours 110 to 120, since the biological solids level increased from approximately 180 mg/l to 235 mg/l. However, there was a corresponding increase in filtrate COD from 50 mg/l to 160 mg/l. This leakage of substrate, though severe, is approximately 75 mg/l below the COD value noted in pulse number three. The unit was shock loaded for six hours between days four and five, but was not examined for COD and biological solids. The results of pulse shock load six (hours 165 to 171) further indicate that the cumulative effects of periodic disruption of the system caused a severe depression of biological solids activity and concentration. The filtrate COD increased during this final pulse shock load to an extremely high value of 335 mg/l COD. It is possible that some type of metabolic inhibition or suppression of enzyme activity was set in motion in the biomass due to the repeated quantitative shock loads. While there are

Figure 20. Response of a Completely Mixed Once-through System to a Periodic Increase in Influent Substrate Concentration From 500 mg/l Dextrose to 1000 mg/l Dextrose at D = 0.0625 $hr^{-1}$

$S_i$ = 500 mg/l dextrose for 18 hours

$S_i$ = 1000 mg/l dextrose for 6 hours


500
400
300
200
100
0
MG/L
SUSPENDED
SOLIDS
COD
950 mg/L
1034 mg/L
1011 mg/L
1003 mg/L
SHOCK LOAD
CONTINUED
0
10
20
30
40
50
60
70
80
90
100
110
120
154
164
174
184
TIME, HR.

insufficient data available at this time for accurate examination of the metabolic reasons for the tendency of the system for failure due to these repeated quantitative shocks, one general theory will be suggested. Studies by Engelbrecht and Gaudy (62) on oxidative assimilation have shown that microorganisms are capable of converting a large fraction of available organic material into internal storage products in the absence of a nitrogen source. Other studies have shown that even in the presence of a nitrogen source, cells may initially synthesize non-proteinaceous cellular materials then sequentially produce protein and nucleic acids prior to replication (64)(65)(66). The storage products may be of such constituents as carbohydrate and/or poly-β-hydroxybutyrate. Gaudy and Gaudy (67) have employed the sequential synthesis to explain the function of the biosorption process, and Komolrit, Gaudy and Gaudy (64) proposed a process modification which makes use of this metabolic phenomenon. It seems possible that during shock loading, the cells may initially remove a large portion of shock load material for use as storage products. If a step change were being applied, these products could then be used over a period of time for replication, i.e., increase in numbers in the biomass. After some time, the system would utilize all excess storage products and return to a new steady state.

In the present study, the microorganisms may remove large amounts of glucose for use as storage products after the initial shock load. Upon subsequent shock loads, the microorganisms may not have completed the conversion of internal storage products to materials for replication before the new shock is administered. Over a period of time, the effects of this response to shock loads become additive and the system

fails.

Figure 21 shows the response of a once-through system to an increase in influent concentration from 500 mg/l dextrose to 1000 mg/l dextrose at D = 0.125 $hr^{-1}$. This experiment was run parallel with the one reported previously (see Figure 20). The unit was shocked for six hours on each of six succeeding days. As can be seen in Figure 21, this set of shock loads exhibited a greater amount of leakage than did those imposed at a dilution rate of 0.0625 $hr^{-1}$. During the initial pulse shock load, the biological solids were unable to increase to a high enough concentration to prevent a leakage of 350 mg/l of COD from the reactor. The biological solids level recovered moderately between hours 40 and 45. As before, the biological solids increased after termination of the shock load. The biological solids from shock one through shock six tended to decrease in concentration and in ability to respond effectively to quantitative increases in influent substrate concentration. The peak concentration of filtrate COD, leaking from the reactor during the six shock loads, remained fairly constant at approximately 375 mg/l. The biological solids from 154 to 169 hours responded only slightly to the 100 percent increase in influent COD, and exhibited a marked decrease in ability to accommodate the shock loadings. These results are in general agreement with those reported in Figure 20, and provide further indication that cyclic quantitative shock loads were not readily accommodated by this once-through system.

Figure 22 shows the response of a completely mixed once-through system to an increase in influent substrate concentration from 500 mg/l dextrose to 2000 mg/l dextrose. The shock load was administered in 6-hours per day pulse loads for a period of seven days at D = 0625 $hr^{-1}$.

Figure 21. Response of a Completely Mixed Once-through System to a Periodic Increase in Influent Substrate Concentration From 500 mg/l Dextrose to 1000 mg/l Dextrose at D = 0.125 $hr^{-1}$

$S_i$ = 500 mg/l dextrose for 18 hours

$S_i$ = 1000 mg/l dextrose for 6 hours

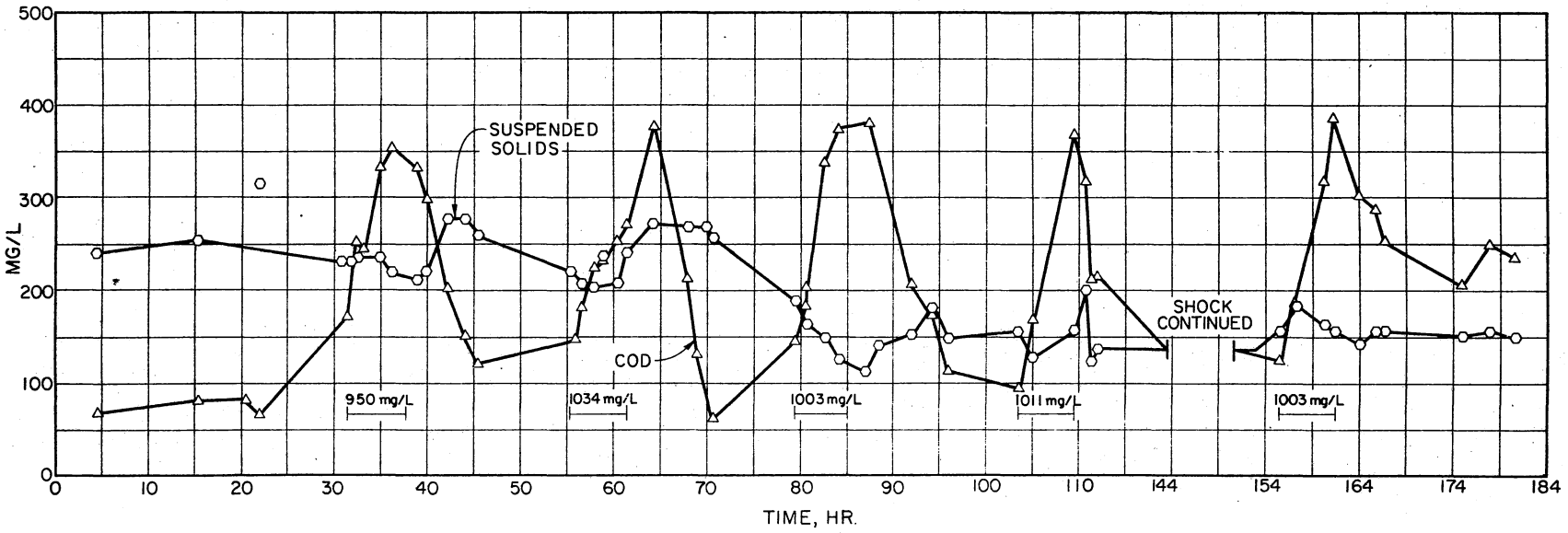
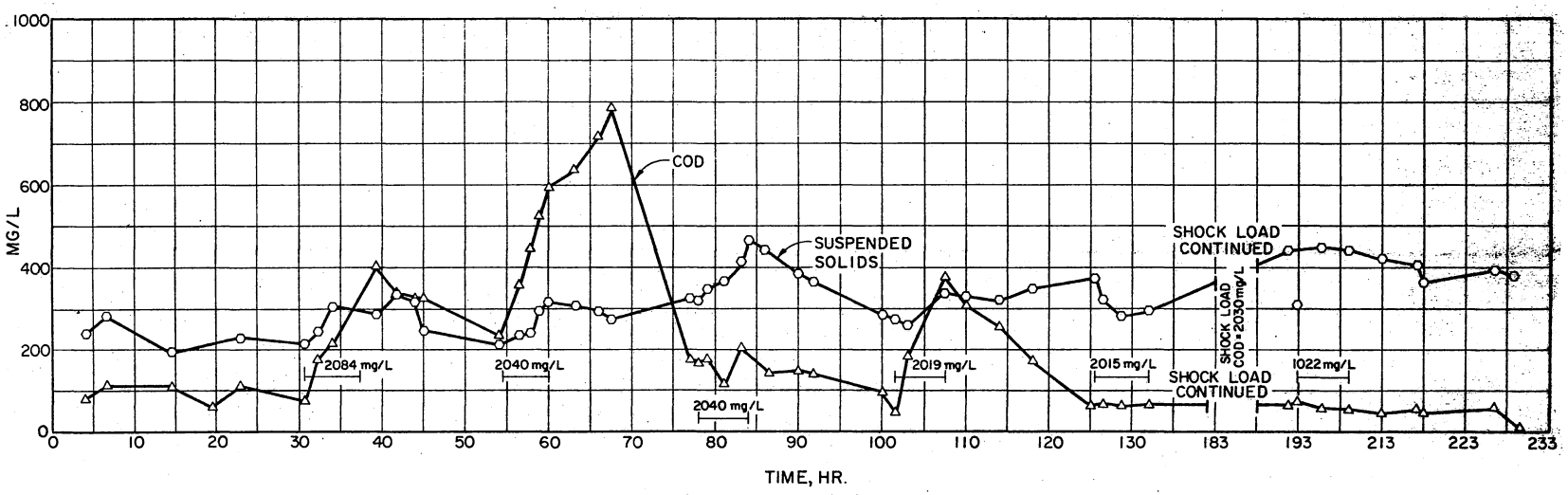

500
400
300
200
100
0
MG/L
SUSPENDED SOLIDS
COD
950 mg/L
1034 mg/L
1003 mg/L
1011 mg/L
1003 mg/L
SHOCK CONTINUED
0
10
20
30
40
50
60
70
80
90
100
110
144
154
164
174
184
TIME, HR.

Figure 22. Response of a Completely Mixed Once-through System to an Increase in Influent Substrate Concentration From 500 mg/l Dextrose to 2000 mg/l Dextrose at $D = 0.0625\ hr^{-1}$

$S_i$ = 500 mg/l dextrose for 18 hours

$S_i$ = 2000 mg/l dextrose for 6 hours


MG/L
1000
800
600
400
200
0
0
10
20
30
40
50
60
70
80
90
100
110
120
130
183
193
213
223
233
TIME, HR.
COD
SUSPENDED SOLIDS
2084 mg/L
2040 mg/L
2040 mg/L
2019 mg/L
2015 mg/L
1022 mg/L
SHOCK LOAD CONTINUED
SHOCK LOAD COD = 2030 mg/L
SHOCK LOAD CONTINUED

In response to the first periodic shock load, the concentration of biological solids increased from approximately 215 mg/l to 340 mg/l in eleven hours. Substrate leakage in the effluent reached a peak concentration of nearly 400 mg/l COD at hour 39. The substrate decreased to 230 mg/l COD just prior to the second shock, which was begun at hour 54.5. The second shock caused a 210 to 315 mg/l increase in biological solids concentration near time 60 hours. The filtrate COD, however, increased tremendously to 795 mg/l. Just prior to the third shock loading at hour 78, the effluent COD was returned to 185 mg/l. The system appeared to accommodate the quantitative increase of the third shock load. The level of biological solids rose gradually from 320 to 420 mg/l, and the filtrate COD increased moderately from 170 to 200 mg/l. During shock load four, the response indicated a marked reduction in the system's ability to accommodate the shock load effectively. Biological solids increased 90 mg/l in response to the substrate increase, while filtrate COD rose 330 mg/l.

After the fourth shock load, three more shocks of 2000 mg/l were applied. It is seen that during the fifth shock load, the effluent quality was excellent and biological solids did not rise. Between hour 130 and hour 137, data taken for the purpose of assessing existence of complete mixing the reactor indicated that such was the case. The biomass and effluent COD results during this shock load are viewed with skepticism, since the results are improbable in light of the previous results and the abnormally low cell yield implied by these results. During shocks six and seven, no data were taken. The author was on active duty during this 2-day period. Dr. Srinivasaraghavan changed the feed and applied the shock during this period but did not obtain samples.

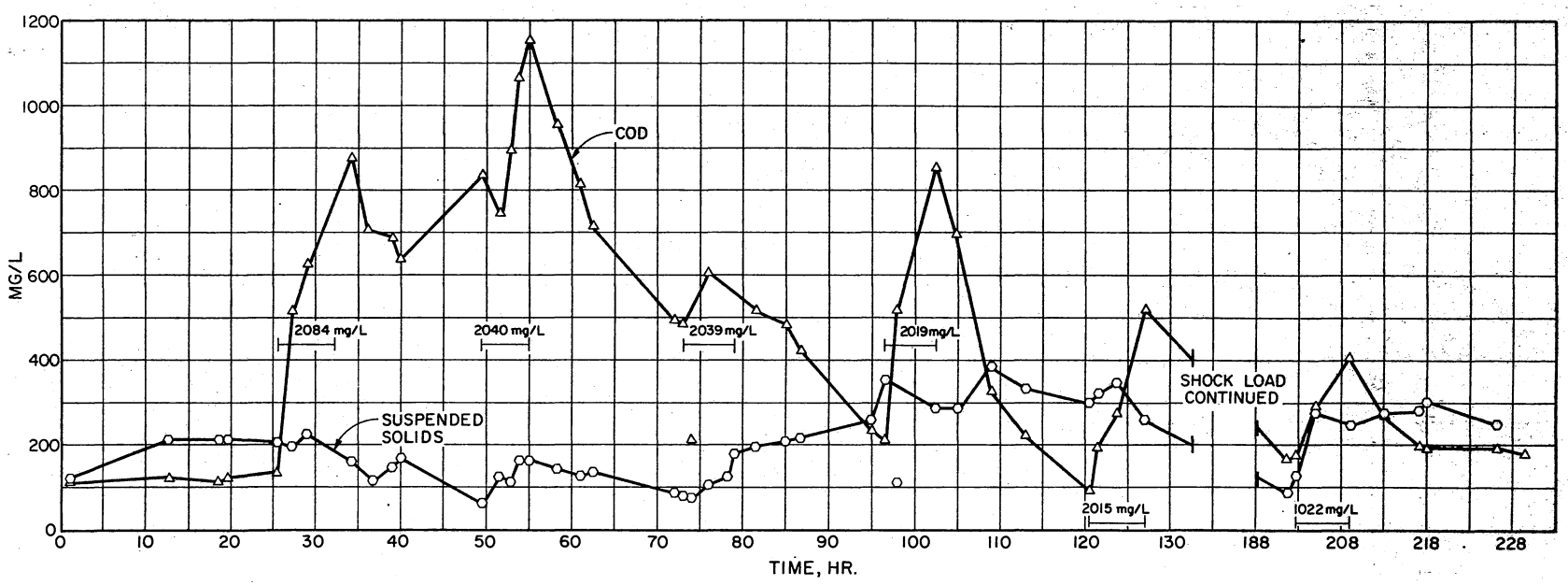
On the eighth day, the unit was again shock loaded but with a shock feed concentration of only 1000 mg/l dextrose. As can be seen in Figure 22, the relatively high initial biological solids concentration which existed in the reactor at this time enabled the system to accommodate the influent substrate increase successfully.

Figure 23 shows the response of a system shock loaded from 500 mg/l dextrose to 2000 mg/l dextrose at a dilution rate of 0.125 $hr^{-1}$. In response to all of the periodic shock loads, a substantial amount of substrate leakage was observed. Biological solids, for the most part, were relatively steady during the first three shock loads, fluctuating generally between 100 and 200 mg/l. As in the shock load shown in Figure 22, the first shock load of the series resulted in a large amount of substrate leakage in the system. The filtrate concentration had only partly recovered before the next highest shock load caused an enormous amount of substrate leakage (similar to that discussed during the second shock load of the previous study, Figure 22) from the unit. In response to the next shock load, the concentration of filtrate leakage was decreased from 1150 mg/l observed in response to the second shock load, to 600 mg/l observed in response to the third pulse shock. By the fourth shock load, a general increase in the biological solids level in the reactor was noted. Solids fluctuated in response to the fourth and fifth shock loads from approximately 280 to 390 mg/l. During shock four, a deterioration in the response of the system resulted in an increase in filtrate COD to near 850 mg/l. Shock load five elicited a somewhat better response than that shown by the system in response to the fourth shock load. Although the biological solids level was approximately equal to the level found during shock load four,

Figure 23. Response of a Completely Mixed Once-through System to an Increase in Influent Substrate Concentration From 500 mg/l Dextrose to 2000 mg/l Dextrose at D = 0.125 $hr^{-1}$

$S_i$ = 500 mg/l dextrose for 18 hours

$S_i$ = 2000 mg/l dextrose for 6 hours


1200
1000
800
600
400
200
0
MG/L
COD
SUSPENDED SOLIDS
2084 mg/L
2040 mg/L
2039 mg/L
2019 mg/L
2015 mg/L
1022 mg/L
SHOCK LOAD CONTINUED
0
10
20
30
40
50
60
70
80
90
100
110
120
130
188
208
218
228
TIME, HR.

the filtrate leakage was 300 mg/l less. After two days of similar shock loads (no data taken), the system was shock loaded for six hours at a decreased concentration of 1000 mg/l dextrose. As can be seen in Figure 23, the system responded in a manner similar to that described in the discussion of Figure 21 (1000 mg/l pulse shock load). The biological solids concentrations and amounts of substrate leakage are in good agreement with the data presented earlier on 1000 mg/l pulse-type shock loads.

From the shock load studies on the response of a completely mixed once-through system presented herein, it can be seen that the results can be highly unpredictable. The normal variables inherent in a heterogeneous microbial population are compounded by the elements of external stress forced upon the system by the shock load. In addition, the additive effects of cyclic shock loads create an even greater burden upon the biological systems and thus, growth processes. Some of the cyclic shock loads studied here apparently never allow the biological population to reach a stable or functional state of equilibrium. The results, particularly for the higher concentration quantitative shock loads, show a tendency for either large-scale disruption of biological efficiency of the system or ultimate system failure.

# CHAPTER V

# SUMMARY AND CONCLUSIONS

## A. Hydrolytically-assisted Extended Aeration Process

1. They hydrolytically-assisted extended aeration process proved to be a highly efficient system for purification of organic wastewaters. In addition, the system was engineerable with respect to control of changes in biological solids concentrations.

2. Due to the high concentration of biological solids and the relatively long aeration period employed in the study, the process was capable of successfully accommodating a 5000 mg/l + glucose and hydrolysate shock load (a five-fold increase). During this severe shock load, the unit showed no apparent upset in purification efficiency. In addition, the COD removal based on filtrate was maintained above 94 percent for the duration of the experiment.

3. Large increases in hydrolysate concentration in the feed (COD = 2800 mg/l) tended to result in disruption of the system, as manifested by a high degree of leakage of filtrate COD. This shock load response was attributed to the complex organic nature of the hydrolysate.

4. Within the framework of the shock loads conducted, it was observed that ability to accommodate shock loading did not deteriorate with chronological age of the system.

5. While shock loads may cause an immediate enmasse biochemical response within the system, they can also cause a delayed response (aftereffect), which is in some cases due to changes in predominance of microorganisms and changes in cell yield. Thus this, which was noted by Thabaraj and Gaudy (61) in once-through systems, is also manifested in high cell recycle systems.

6. Rapid increases in biological solids concentrations in response to increases in organic loading may result in changes in the consistency of the sludge. Following biological solids increases resulting from quantitative shock loadings, the characteristics of the sludge changed from a dark brown, compact sludge, to a lighter, golden, bulky sludge. An accompanying increase in turbidity was observed incidental to several of the changes in sludge morphology.

In general, it was concluded that the hydrolytically-assisted extended aeration process can occupy a position of practical utility in the field. The process presents a suitable alternative for consideration for treatment of industrial wastes and small municipalities. The system offers excellent treatment efficiency, nutrient recycle, and elimination of requirement for sludge disposal equipment. It also is engineerable with respect to biological solids concentration and is resistant to many types of environmental changes or shock loads.

## B. Completely Mixed Once-through System

1. A completely mixed once-through system can successfully accommodate cyclic decrease in dilution rate from 0.125 $hr^{-1}$ to 0.0625 $hr^{-1}$ (one hundred percent increase in detention time). On the other hand, the system could not successfully respond to a four-fold increase in

dilution rate from 0.0625 $hr^{-1}$ to 0.25 $hr^{-1}$. At this decrease in detention time of from 16 to four hours, a large concentration of substrate leaked from the system. A trend was noted, however, in that the substrate removal efficiency of the unit increased with each successive shock load, indicating a possible acclimation of the cells resulting from a selective change in predominance.

2. In general, a trend toward an increase in disruption of the system following several periodic quantitative shock loads was noted. The disruption was identified by a trend of increasing filtrate COD concentration and deterioration in ability of the biological solids to increase in response to shock loads. In a majority of the quantitative shock loads studied, there was observed a tendency for the system to approach failure or large-scale disruption of purification efficiency following a series of quantitative shock loads.

3. The response of the system to quantitative shock loads was more successful at a dilution rate of 0.0625 $hr^{-1}$ than at a dilution rate of 0.125 $hr^{-1}$. In all instances, the amount of substrate leakage at the lower detention time ($D = 0.0625\ hr^{-1}$) was significantly less than the leakage observed at $D = 0.125\ hr^{-1}$ (8-hour detention time).

In general, results obtained from the completely mixed system showed a high degree of variability. The circumstances brought about by the cyclic shock loads created a situation in which the biomass was never completely free of one transient state or another. The present work was, to the author's knowledge, in all probability the first time a heterogeneous population was subjected to the periodic loads herein imposed, and it is evident from the results that the responses are as complete as they are interesting to study. It is concluded that

further work could be fruitfully pursued, and some possibilities for such investigations are listed in the following chapter.

# CHAPTER VI

## SUGGESTIONS FOR FUTURE WORK

The study of the response of biological systems to environmental stress has become increasingly important due to recent requirements for more stringent wastewater effluent standards. There is presently an enormous number of gaps in the understanding of microbial kinetics in the transient state and of effects of shock loads upon the biological efficiencies of treatment systems. In addition, the complex reactions and interactions at the physiological, morphological, and biochemical levels have not been sufficiently ascertained for a heterogeneous microbial population growing in the transient state. Before effective mathematical modeling and design of biological systems can be accomplished, a great deal of research energy must be expended at the basic and practical level toward attainment of definitive answers. Some suggestions for future work are described below.

1. A periodic series of shock loads corresponding to changes in flow and organic concentration characteristic of variations in flow found at on-line treatment plants should be imposed on a hydrolytically-assisted extended aeration pilot plant.

2. Step change shock loads of the hydrolytically-assisted extended aeration process should be continued to determine the parameters of operational stability of the system.

3. Further studies of the response of completely mixed, once

through systems to cyclic shock loads should be undertaken. The studies should be made on hydraulic and quantitative shock loads of longer duration than those herein reported, and should be extended to include changes in pH and temperature, and qualitative changes in carbon source.

4. Studies should be conducted to determine the effect of shock loads on changes in predominance of microorganisms in heterogeneous populations.

5. It is suggested that similar shock load studies of pure cultures be considered in order to assist in distinguishing between inter-species and intra-species responses to environmental changes.

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