STUDIES ON THE HYDROLYTICALLY-ASSISTED EXTENDED AERATION PROCESS AND ON PRE-HYDROLYSIS OF SLUDGE IN AEROBIC DIGESTION PROCESSES

By

HOMAYOON SAIDI Licentiate Degree of Engineering

Tabriz University

Tabriz, Iran

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Thesis Approved:

Thesis Ad

Dean of the Graduate College

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iii

TABLE OF CONTENTS

Chapter	^ ·	Page
I.	INTRODUCTION	1
II.	LITERATURE REVIEW	3
III.	MATERIALS AND METHODS	8
	 A. Experimental Design 1. Studies on Operation and Efficiency of an 	8
	Extended Aeration Process with Hydrolytic Pre- treatment of Some Return Sludge 2. Studies on Aerobic Digestion of Sludge After	- 8
	Hydrolysis	13
	Cells Taken From the Pilot Plants 4. Studies on the Endogenous Oxygen Uptake of	13
	Cells Grown in the Pilot Plant	17
	assisted Extended Aeration Activated Sludge to Shock Loadings	17
	From Extended Aeration Pilot Plants	18 19 19
IV.	RESULTS	2 1
	A. Phase I	21
1 1 1	Organic Loadings	21 33
1	Sludge	33 44
۷.	SUMMARY AND CONCLUSIONS	47
VI.	SUGGESTIONS FOR FUTURE WORK	50
BIBLIO	GRAPHY	51

LIST OF TABLES

Table		Page
Ι.	Composition of the Waste	11
II.	Composition of the Waste	13
III.	Effect of Conditions of Hydrolysis on Sludge Characteristics	46

LIST OF FIGURES

Figu	Figure	
1.	Longitudinal Section of Pilot Plant Aeration Basin and Settling Tank	10
2.	A General Scheme for Total Oxidation Consisting of an Activated Sludge Pilot Plant With Sludge Consistency TAnk (Aerator #2) and Aerobic Digestion of Hydrolyzed Excess Sludge	15
3.	Performance of a "Hydrolytically-assisted" Extended Aeration Process at High Organic Loadings	24
4.	Effect of Various Types of Shock Loads on the Operation of the Extended Aeration Pilot Plant	30
5.	Performance of an Aerobic Digestion Pilot Plant Employing Pre-hydrolyzed Sludge as Feed Stock	37
6.	Performance of the Aerobic Digestion Pilot Plant During a Period of Batch Operation Consisting of 280 mg/l Hydrolysate COD	42

CHAPTER I

INTRODUCTION

Control of water pollution has become one of the major public concerns of our time. Formerly, primary treatment for removal of settleable solids may have seemed adequate, but with industrial and population growth, secondary treatment has become mandatory today and, in some cases, there is need for treatment of the wastewater beyond this level. Secondary treatment by various biological processes offers perhaps the most economical method for removal of soluble organic matter. Since the success of the process depends upon the growth of microorganisms, the excess cells, or sludge, become a waste product of the purification plant which must be disposed of in some way. Biological means of accomplishing sludge reduction include both anaerobic and aerobic digestion. Some time ago, another alternative was suggested. Porges and his co-workers suggested that waste purification and sludge disposal could be accomplished concurrently by recycling all of the excess sludge to the aeration tank. If the aeration tank were made large enough to provide for an "extended" period of operation, the excess sludge might be autodigested, and if the loading were adjusted, a balanced condition of synthesis and aerobic digestion could lead to a steady value of biological solids concentration in the system. The process met with some acceptance by practicing engineers, but general non-acceptance by researchers in the field, and many prominent

investigators concluded that the concept of total oxidation of biological cells was not theoretically sound. In order to help resolve the controversy, a long-term investigation was begun in the bioengineering laboratories of the Oklahoma State University. After four years of research it was possible to conclude that the concept of total oxidation was not unsound from a biological standpoint. However, the steady state with respect to biological solids as had been predicted by the originators of the process was not achieved, and it was desirable to ascertain some engineering expedient by which control of the biological solids might be attained. This need led to the development of the "hydrolytic-assist" process, which has been under investigation in these laboratories for three years. It consists in the main of withdrawal, hydrolysis, and partial liquefaction of a portion of the sludge prior to its recycle to the aeration tank. The results to date indicate the process offers a possible way to "engineer" the extended aeration process. However, much work remains to be done and the current investigation was initiated for the purpose of gaining further insight into the characteristic behavior of the process. The scope of the present studies specifically involves pilot plant investigations in which the purification efficiency, nitrification tendency, and biological solids level were assessed at high organic loadings of 500 and 1000 mg/l carbon source. Also examined in rather long-term pilot plant experimentation was the possibility of feeding only sludge hydrolysate to an extended aeration process, i.e., using it essentially as a separate sludge disposal process. Also included are some studies to examine various conditions of hydrolysis.

CHAPTER II

LITERATURE REVIEW

The concept of total oxidation of biological sludges began in the early 1950s and arose out of the work on the treatment of skimmed milk wastes by Porges, et al. (1)(2)(3). They found that under sufficient aeration time and by regulating the inflowing feed loading, an activated sludge system "could maintain a balance, producing no excess sludge and discharging an effluent practically free of organic matter" (4). These findings caused a considerable amount of research to be undertaken, and in 1953, Kountz reported that ninety percent COD removal efficiency was accomplished during a one-year period in which no sludge wasting was practiced (5). Six years later, Kountz and Forney changed their conclusion and stated that a solids balance could be reached when some wastage was practiced, whereas for the case of total cell recycle they found in this later work that solids balance could not be reached within a reasonable time period because there was an accumulation of residue equal to twenty percent by weight of the solids produced (6). In 1960, Busch and Myrick, employing batch and continuous flow extended aeration pilot plants reported that no solids balance was observed after 103 days of operation, although they did find very high COD removal efficiencies. They concluded that it was impossible to reach a solids balance unless biological solids inadvertently carried over with the effluent were equal to the buildup rate of biological solids

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concentration in the system (7). Washington and Symons employed various substrates in total solids recycle systems, and reported that there was an accumulation of inert volatile nitrogen-containing materials, mainly extracellular polysaccharides. They concluded that such a buildup was inevitable, but that addition of nitrogen to the system improved the process (8). Later, Washington, Hetling, and Rao, using a batchfed extended aeration unit, found evidence for some autodigestion and/or lysis of the organisms in the mixed liquor (9). Ludzack made benchscale studies of the extended aeration process and found that twenty percent wastage of solids per week yielded a volatile suspended solids content of 75-80 percent in the sludge, and when sludge wastage was decreased to five percent per week, the volatile content of the sludge decreased to 60 percent. Finally, when no sludge wastage was practiced, the volatile content dropped from 60 to 55 percent (10). Eye, et al., after studying operational conditions of various extended aeration activated sludge plants, concluded that the accumulation of non-volatile suspended solids in aeration tanks was due to accumulation of mud and clay (11).

Evidence in favor of the concept of total oxidation was presented in 1971 by Thabaraj and Gaudy (12). They showed the results of batch experiments in which a prolonged period of endogenous metabolism of the biological solids, which had been grown up under balanced growth conditions, resulted in oxidation of an amount of solids essentially equal to the total amount of solids synthesized during the substrate removal period (12).

Results such as those just cited caused Gaudy and his co-workers to undertake rather extensive studies in regard to the concept of total

oxidation and the extended aeration process in general. Long-term pilot plant studies were undertaken in which there was obtained evidence for periods of biological solids accumulation and periods of de-accumulation due to autodigestion of the biological solids in the reactor (13)(14)(15). During these studies, positive retention of all biological solids was attained through centrifugation of the pilot plant effluent. Throughout this long period of study, the purification efficiency of the system remained very high and there was no evidence for the buildup of an inert fraction in the biological solids. There were indeed times when biological solids would have been lost in the plant effluent had the system not been backed up by centrifugation, and although this work proved from a basic standpoint that the total oxidation concept was not theoretically unsound, it was deemed desirable to seek some sort of engineering expedient which could assist or initiate a de-accumulation phase. Thus, in subsequent studies it was decided to investigate the use of the following practice. During times when biological solids concentration was climbing to concentrations which might begin to interfere with settling of the sludge, some sludge was withdrawn and hydrolyzed chemically. This chemically hydrolyzed or liquefied sludge was then fed back to the aeration tank along with incoming substrate. Studies accomplished under this mode of operation indicated it to be a potentially useful engineering modification (16)(17)(18).

In addition to these engineering studies, separate investigations were undertaken in the Oklahoma State University bioenvironmental engineering laboratories to gain insight into the so-called inert and/or non-biodegradable material of the sludge. The work of Symons and McKinney (19) had indicated that extracellular polysaccharides of

biological cells constituted a biologically-inert fraction. However, when extracellular polysaccharide was obtained from various species of microorganisms and fed to heterogeneous populations, it was observed to serve as an excellent source of carbon for growth (20)(21). Work in the bioengineering laboratories regarding the extended aeration process has also been undertaken to assess the ability of such systems to accommodate shock loadings. The work to date indicates that such systems can yield excellent COD removal efficiencies under steady and under shock loading conditions (13)(22)(23).

The new modification of the extended aeration process, i.e., incorporation of the hydrolytic pre-treatment of some of the sludge from the clarifier, was examined using both synthetic waste and a pulp-mill waste and was shown to be capable of rather good purification efficiency under fairly high organic loading conditions (14). When effluent from the pilot plant was examined for its potential effect upon receiving stream waters using an open jug technique to obtain the oxygen uptake curves, the effluent was found to be one that would not be expected to cause serious drawdown of the oxygen resource (24). Although on the basis of the results gathered in this laboratory, it is possible to conclude that the hydrolytically-assisted extended aeration process offers considerable promise for application in the field, it was felt that more experimental information was needed and that on the basis of such further work it might be possible to extend the scope of applicability of the new process. Accordingly, work was undertaken primarily to examine the following aspects:

1. Could the process be successfully operated at high substrate loading, e.g., 500 and 1000 mg/l COD so as to yield not only good COD

removal but a high degree of nitrification of the effluent?

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2. Would it be possible to employ a hydrolytically-assisted extended aeration process solely as a means of sludge disposal for various biological sludges?

CHAPTER III

MATERIALS AND METHODS

A. Experimental Design

To initiate the continuous flow pilot plant, a batch-activated sludge was developed using glucose and other nutrients. The sludge was initiated from an original seed obtained from the supernatant of the secondary clarifier of the Stillwater municipal sewage treatment plant.

1. Studies on Operation and Efficiency of an

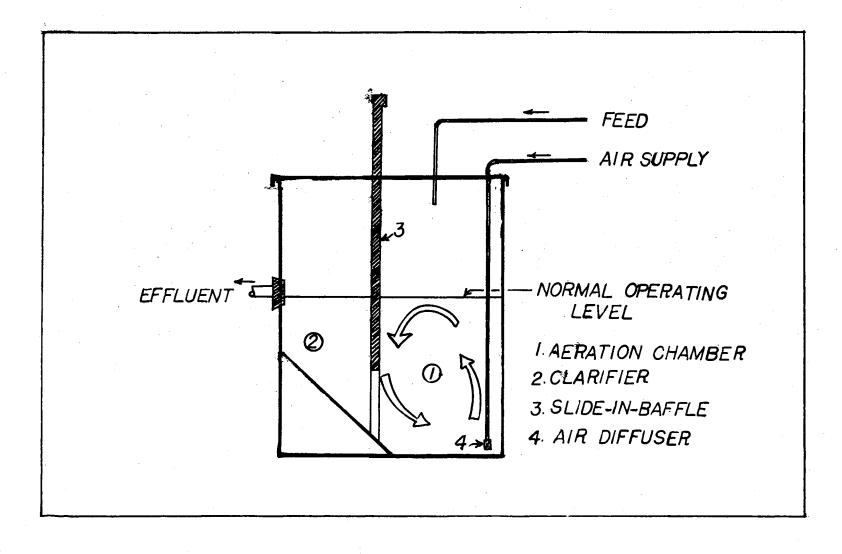
Extended Aeration Process With Hydrolytic Pre-

treatment of Some Return Sludge

The pilot plant unit used throughout these experiments was a plexiglass unit with a 6.2-liter aeration basin and a 3.2-liter settling chamber yielding a total volume of 9.4 liters (see Figure 1). The two compartments were separated by a movable baffle leaving a gap between it and the tank bottom so that mixed liquor could pass to the settling tank. Aeration was provided by sintered glass diffusers. The flow rate of 9.4 liters/day was provided by a minipump (Milton Roy Model MM2-B-96R) to allow a detention time of 16 hours in the aeration tank and eight hours in the settling tank with a total detention time of 24 hours. Temperature was maintained at $23^{\circ}C \stackrel{+}{-} 2.0$. The pH of the system was adjusted to 7.0 as needed, by making direct injection of

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Figure 1. Longitudinal Section of Pilot Plant Aeration Basin and Settling Tank



alkaline solution (sodium hydroxide) to the aerator and by continuous addition of buffer solution in the feed. Composition of the waste for 1000 mg/l of glucose is given in Table I. When the carbon source was 500 or 2000 mg/l of glucose, either half or double the values shown in Table I were employed.

TABLE I

COMPOSITION OF THE WASTE

	ومرجا المحافظة المستار بجد متحاصيت والمحافظين ومراور والتجريا متعاوية والمتحر والمحاف
Glucose	1000 mg/1
(NH ₄) ₂ S0 ₄	500 mg/1
MgS0 ₄ •7H ₂ 0	100 mg/1
FeC1 ₃ •6H ₂ 0	0.5 mg/1
CaC1 ₂ •2H ₂ 0	7.5 mg/1
MnS0 ₄ •H ₂ 0	10.0 mg/1
1.0 M Phosphate Buffer	
$pH = 7.0 (KH_2PO_4 + K_2HPO_4)$	10-15 m]/1

The COD of the feed was measured after addition of salts and carbon source in accordance with Table I, and a portion of cell hydrolysate to a feed reservoir filled to the desired volume by adding distilled water. The feed reservoir was cleaned each day and refilled. Alternate feed reservoirs were employed; one was being cleaned while

one was being placed in operation. The reservoir was cleaned with dichromate cleaning solution, and was rinsed free of spent solution with multiple tap water rinses and a final distilled water rinse. The feed lines were cleaned each day for a one-hour period by recirculating a mild Clorox solution through the lines and pump. This was followed by tap water rinses. The sampling procedure was as follows: Each day the feed line was removed from the unit and the suction end was removed from the reservoir and placed in the Clorox solution reservoir. Immediately after removing the feed line from the aeration tank of the pilot plant, the effluent port was stoppered, the clarification tank baffle was removed, and the total contents of the system momentarily mixed. At this time, a sample was taken to determine the biological solids concentration in the system, and the baffle was immediately re-set. Also during this time, samples were removed from the effluent collection reservoir (after thoroughly mixing it) for measurement of biological solids, filtrate COD, and total COD. Samples were also taken for NH_3-N , NO_2-N , and NO_3-N on the filtrate of the composite effluent sample. The samples for nitrogen analysis were frozen for later testing. The total sampling period usually required approximately one hour, and during this time the feed lines were being cleaned with Clorox solution, rinsed with tap water, and flushed with new feed. The line was then re-engaged with fresh medium, and the pilot plant set into continuous flow operation until the following day's sampling period. Weekly, a portion of settled sludge was withdrawn from the bottom of the clarifier and biological solids determinations were made prior to hydrolyzing it. After acidifying to pH 1, this portion of sludge was subjected to high temperature $(121^{\circ}C)$ and high pressure (15 psi) for

five hours in a laboratory autoclave. The hydrolysate was removed, cooled to room temperature, and finally neutralized. Equal portions were added to each daily feed over the next seven-day period.

2. Studies on Aerobic Digestion of

Sludge After Hydrolysis

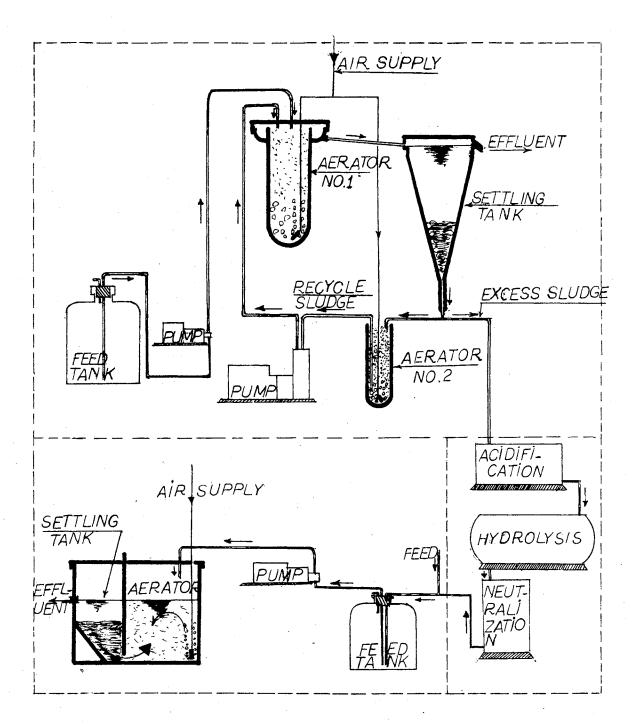
The general mode of operation of the system was the same as outlined above. However, the feed composition (see Table II) and the sludge from which the hydrolysate was obtained came, primarily, from a completely mixed laboratory activated sludge plant which was being operated as shown in the upper portion of Figure 2. The excess sludge from this laboratory pilot plant was hydrolyzed and fed along with other nutrients. Also, there were times when sludges were fed from various other laboratory pilot plant operations. These will be explained in the Results section. During this phase there was no hydrolysis of sludge developed in the aerobic digestion unit, i.e., it was not hydrolyticallyassisted.

3. Studies on Growth and Substrate Removal of the Cells Taken From the Pilot Plants

a) For measuring growth rate and other biological constants, a shaker apparatus (Eberbach Corporation, Ann Arbor, Michigan) was adjusted to 100 osc/min. Different substrate concentrations ranging from 250 to 1000 mg/l glucose were used, and mineral salts were added in proportion to concentrations shown in Table I. The total volume of samples in each flask was 50 ml, and the cells which were taken from the aeration chamber of the pilot plant were immediately added to each flask to

Figure 2. A General Scheme for Total Oxidation Consisting of an Activated Sludge Pilot Plant With Sludge Consistency Tank (Aerator #2) and Aerobic Digestion of Hydrolyzed Excess Sludge

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give a suspended solids level of approximately 50 mg/l. Growth was measured by optical density using a Bausch-Lomb spectronic 20 colorimeter. The biological constants, maximum specific growth rate, μ_{max} , and saturation constant, K_s, were obtained using a straight line form of the hyperbolic relationship between specific growth rate and limiting nutrient concentration suggested by Monod (25).

TABLE II

COMPOSITION OF THE WASTE

Hydrolysate	400-800 m1
(NH ₄) ₂ SO ₄	150 mg/l
MgS0 ₄ •7H ₂ 0	30 mg/1
FeC1 ₃ ·6H ₂ 0	0.15 mg/1
CaCl ₂ •2H ₂ O	2.25 mg/1
MnS0 ₄ •H ₂ 0	3.0 mg/1
1.0 M Phosphate Buffer	· *
$pH = 7.0 (KH_3PO_4 + K_2HPO_4)$	10-15 m1/1

b) For measuring yield coefficient, a large volume of feed with a glucose concentration of 1000 mg/l (with salts concentration as per Table I), and 50 mg/l of biological solids as initial inoculum were aerated in a batch reactor. Immediately after mixing biological solids

and substrate, a sample was removed and filtered to measure initial suspended solids and initial COD concentration (filtrate). Periodic samples were taken during the ensuing course of growth and substrate removal. The sample for biological solids and filtrate COD at the time of COD removal were used to calculate the cell yield as follows:

$Y = \frac{\text{change in biological solids concentration}}{\text{change in COD}}$ (1)

<u>4. Studies on the Endogenous Oxygen Uptake of</u> <u>Cells Grown in the Pilot Plant</u>

A sample of mixed liquor was removed from the aeration tank, and biological solids determination was made. Then a 40-ml sample of the suspended solids was placed in a Warburg flask (1.5 ml of 20 percent KOH in the center well). Oxygen uptake was then measured using the Warburg respirometer (Gilson Medical Electronics, Middleton, Wisc.) with the oscillation rate set at 105 osc/min. The suspension of cells was not washed free of substrate because in all cases the soluble substrate in the aeration tank was negligible. The average hourly uptake rate during the linear portion of the accumulated 0_2 uptake curve was divided by the initial biological solids concentration in the flask and reported as endogenous 0_2 uptake rate in mg/l 0_2 /gm sludge.

5. Studies on the Response of Hydrolytically-

assisted Extended Aeration Activated Sludge

to Shock Loadings

a) Feed containing 9.4 times the given values in Table II was

introduced to the pilot plant after stopping continuous flow operation and removing the baffle. The COD and biological solids concentration were measured at periodic intervals and a plot of COD remaining and biological solids produced vs. time was made.

b) During regular continuous flow operation, the system was subjected to a change in feed glucose COD (quantitative shock load) from 1000 to 3600 mg/l. Clarifier supernatant COD was periodically determined to assess the ability of the system to accomodate the shock.

c) During continuous flow operation, a combined quantitative and qualitative shock was administered for which the feed composition was changed from 1000 mg/l glucose COD to 3100 mg/l glucose COD + 5500 mg/l sorbitol COD. The mineral salts concentration was increased in proportion to the amount of glucose in the shock, but it was not increased in accordance with the sorbitol concentration. COD, glucostat, and periodate determinations were made on the plant effluent to assess the results of the shock.

d) The system was subjected to hydraulic shock load with feed remaining at 1000 mg/l glucose. The flow rate was doubled, i.e., total detention time reduced from 24 to 12 hours. Supernatant COD from the clarifier was measured to assess the ability of the system to accommodate the hydraulic shock loading.

6. Studies on Chemical Flocculation of Effluents From Extended Aeration Pilot Plants

a) FeCl₃·6H₂O at concentrations of 50, 100, 200, 300, and 400 mg/1 were added to beakers containing samples of the effluent. These samples were then subjected to jar tests. The Phipps and Bird apparatus

was employed. The conditions of the test were as follows: One minute of rapid mixing was followed by ten minutes of slow mixing for flocculation, and this period was followed by 30 minutes of quiescent settling. The effect of chemical treatment was assessed by measuring percent transmittance before and after treatment, as compared to the percent transmittance of a sample of untreated effluent.

b) Studies were also made in which $\text{FeCl}_3 \cdot 6\text{H}_20$ was employed at dosages of zero to 50 mg/l, and in which $\text{AL}_2(\text{SO}_4)_3$ was also employed at the same concentrations. The experimental procedure was as described above.

7. Studies on Conditions of Hydrolysis

Sludges from different sources were subjected to five hours' hydrolysis at 121^OC and 15 psi under varying conditions of pH and biological solids concentration. Analyses were made for biological solids, COD of the mixed liquor, protein content, and carbohydrate content before and after hydrolysis. Concentrated sulfuric acid (36 N) and sodium hydroxide (20 N) were employed in these experiments to adjust the pH.

B. Analytical Methods

Chemical Øxygen Demand (COD), biological solids concentration employing membrane filter technique (Millipore Corporation, Bedford, Mass., HA .45 micron), NO_3 -N (Brucine Method), and NO_2 -N (Diazotization Method) were run in accordance with recommendations set forth in Standard Methods for the Treatment of Water and Wastewater (26). Ammonia nitrogen was measured using a method developed by Ecker and Lockhart (27). Anthrone, biuret, and periodate tests were run in accordance with the recommendations given by Ramanathan, Gaudy, and Cook (28). pH was determined periodically with a Beckman expandomatic SS-2 pH-meter. Dissolved oxygen (DO) was measured using a DO analyzer (Weston and STack, Malvern, Pa.).

was operated as a batch process. After the biological solids concentration had attained a level of 3500 mg/l, some of the solids were placed in a second extended aeration unit which served to initiate the second phase of the investigation, i.e., aerobic digestion of excess sludge. At this time, the first unit (hereafter referred to as Unit #1), contained a biological solids concentration of 2000 mg/l. On May 13, microscopic observation revealed the presence of a considerable population of filamentous organisms. The amount of filamentous organisms in the system increased until May 24, at which time a serious bulking problem had accrued. At this time it was decided to withdraw half the sludge from Unit #1, acidify, hydrolyze, neutralize it, and feed it back to the aeration tank along with the 500 mg/l of incoming glucose synthetic waste. By June 22, the bulking problem had been alleviated. This day was selected as day zero (see Figure 3), and from this day onward a rather complete set of performance data was obtained. It can be seen at the top of Figure 3 that the feed to the unit at this time consisted of 500 mg/l glucose, plus 80 mg/l COD contributed from the cells which had been previously hydrolyzed. Also, over the first 27 days of operation, two 900-ml portions of sludge were withdrawn from the clarifier for hydrolysis and refeeding to the unit in accordance with the hydrolysis schedule which had been previously made at a lower organic loading. During the 27-day period at this loading, the purification efficiency based on COD of the filtrate ranged from 92 to 100 percent with an average of 96 percent, and based upon supernatant COD, the purification efficiency ranged from 76 to 93 percent with an average of 87 percent. It is most significant to note that during this period there was little or no ammonia leakage from the system. It

CHAPTER IV

RESULTS

The results of this investigation will be presented in two major sections. The first deals with the operation of an extended aeration process utilizing the "hydrolytic assist" for the treatment of a synthetic waste at rather high organic loadings. The second phase of this chapter deals with the presentation of results on the operation of a "hydrolytically-assisted" aerobic digestion process utilizing excess sludge from various pilot plant operations in the laboratory.

A. Phase I

 Studies on the Use of the Hydrolyticallyassisted Extended Aeration Process at High

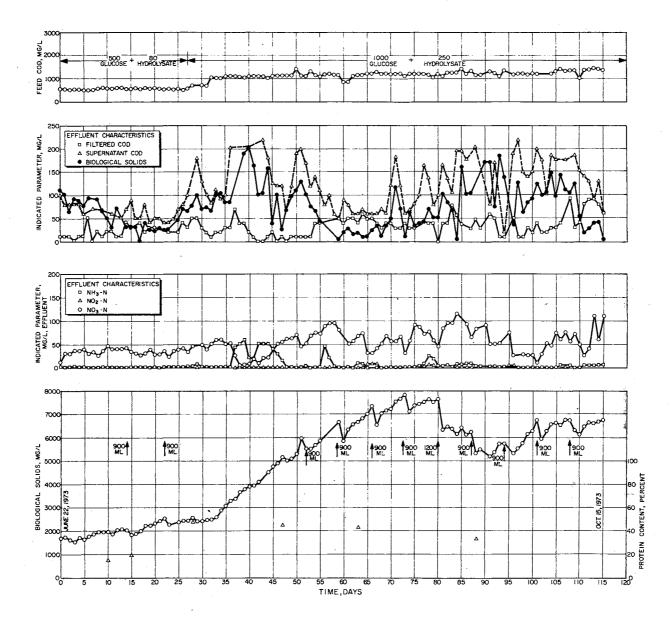
Organic Loadings

The major purpose of this phase of the investigation was to determine whether the hydrolytically-assisted extended aeration process could deliver, in addition to a well-treated effluent, a highly nitrified effluent. Since previous studies had shown that a highly nitrified effluent could be produced at glucose loadings of slightly higher than 300 mg/l, the initial loading in this study was a glucose concentration of 500 mg/l.

The unit was put into operation on May 1, 1973, and for some time

Figure 3. Performance of a "Hydrolytically-assisted" Extended Aeration Process at High Organic Loadings

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should be recalled that the COD:nitrogen ratio in the synthetic waste was 10:1. Essentially all nitrogen which appeared in the effluent was in the form of nitrate-nitrogen, i.e., the system produced a highly nitrified effluent, thus retaining one of the significant advantages of an extended aeration process.

On days 10 and 15, protein analyses were run, and these appear abnormally low; however, it can be seen that by day 28, the protein content was above 40 percent. During the entire 27-day period of operation at this loading, there was a tendency for the sludge in the clarifier to rise. The rising sludge could not be attributed to the usual cause for such a phenomenon, i.e., denitrification, since the effluent was highly nitrified. It was believed that the source of gas bubbles causing the sludge to exhibit the rising tendency was compressed air from the aeration tank, and efforts were made to correct the problem by adjusting the baffle height.

Even though it seems apparent that the biological solids concentration was still rising and had not attained a semi-balanced condition, it was amply apparent that the system was nitrifying, and the decision was made to increase the organic loading to 1000 mg/l glucose in the synthetic waste. On day 27, the feed concentration was increased from 500 to 700 mg/l glucose plus 80 mg/l hydrolysate COD, and approximately one week later was raised to 1000 mg/l glucose plus 250 mg/l hydrolysate COD. Over the next 40 days, the biological solids concentration continued to climb even though on day 51 the withdrawal and hydrolysis schedule was begun. By day 80, it appeared that the biological solids concentration was beginning to attain a balanced condition. However, since it had been rising previous to this time, 1200 ml rather than 900 ml were withdrawn for hydrolysis. The withdrawal, hydrolysis, and refeeding of this larger volume of sludge may or may not have been the cause of the increase in effluent biological solids and supernatant concentrations, but this occurrence did precipitate a decision to return to the 900-ml level of withdrawal. This procedure was followed by the ensuing 3-4 week period; the mixed liquor biological solids concentration did appear to attain a semi-level condition with a mean of approximately 6000 mg/l. It will be noted that the effluent characteristics with respect to filtrate COD remained excellent throughout the entire period of operation at the higher substrate loading level. The efficiency of removal varied from 83 to 100 percent, with an average of 96 percent. Based on supernatant COD, the efficiency of removal ranged from 64 to 95 percent, with an average of 87 percent. During the final 4-week period of operation at this loading level, the average removal efficiency based on the COD of the filtrate (ranging from 74 to 98 percent) was 95 percent, and based on supernatant COD, the efficiency was 86 percent (ranging from 67 to 95 percent). Also, even at this higher loading level, there was ample evidence that the system could be expected to provide a rather highly nitrified effluent. Periods of low nitrification (e.g., between days 35 and 45, and days 95 and 113) tended to coincide with periods of excessive leakage of biological solids from the system. Thus, it appeared that the nitrifying organisms during these periods of low nitrification were the ones being washed out of the system.

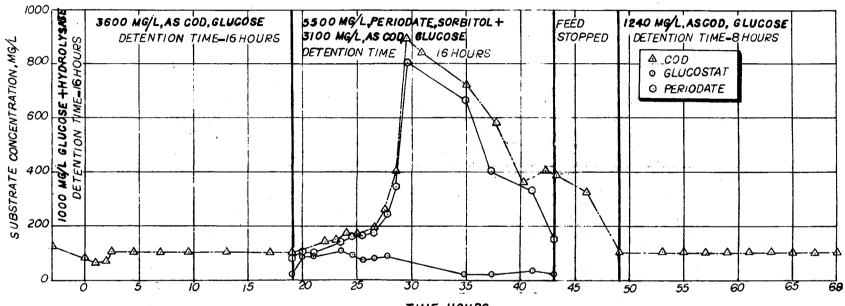
It is noted that the amount of 1 M phosphate buffer which was added to the feed was 15 ml/l. This was not sufficient buffer to maintain the pH at 7. There were times when the pH, if not adjusted by sodium

hydroxide, would have dropped below 6.4. In order to maintain the pH closer to 7, it was adjusted as needed (sometimes two or three times per day) with sodium hydroxide. This procedure was followed in lieu of increasing the concentration of buffer in the feed since it was observed in the preliminary studies with the unit that addition of very high amounts of phosphorus corresponded to periods of decreasing nitrification. Thus the mode of pH control could have militated against the desired objective of assessing the ability of the unit to nitrify at the organic loadings under study. The addition of 30 ml of 1 M phosphate buffer per liter corresponds to a potassium phosphate concentration of approximately 4800 mg/l. It is believed that potassium phosphate concentrations in the range of 4-5 grams/liter approach the toxic threshold limit for most bacteria (29). It seems possible that the toxic limit for the nitrifying bacteria could be somewhat lower than this value.

At this point in the study, it appeared that there was little doubt that an extended aeration process employing the "hydrolytic assist" could be operated at an incoming substrate concentration of 1000 mg/l with excellent substrate removal efficiency and with the delivery of a highly nitrified effluent. While it would have been interesting to simply increase the loading to determine if high substrate removal efficiency along with nitrification could be accomplished at higher loading levels, it seemed desirable to leave such studies for another investigator, because the author had become interested in assessing the ability of this system to accommodate various types of shock loadings successfully. It was especially interesting to do shock loading studies because at this time it appeared that the sludge was

settling extremely well and one could test both biochemical and overall efficiency in response to shock loadings. Accordingly, on October 16, 1973, the incoming substrate concentration was changed from 1000 mg/lglucose + 250 mg/l hydrolysate COD to a glucose COD concentration of 3600 mg/l. The samples were collected for measurement of biological solids in the effluent and effluent COD in the filtrate as well as the effluent supernatant COD from the clarifier. A few samples for biological solids concentration in the reactor were also taken. The response to the shock is shown in Figure 4. Only the supernatant COD concentrations were plotted, since the COD of the filtrate corresponded to the COD of the supernatant, i.e., the biological solids concentration in the effluent was negligible. There was throughout the extent of this shock loading and those which followed it, which were even more severe, never any instance of loss of sludge settleability. It is seen that the three-fold increase in influent COD had essentially no effect on the system. At the time of administering the shock (time zero on the X axis), the biological solids concentration in the system was $6510 \text{ mg/l}_{\circ}$ and at the time of terminating this particular loading, i.e., 19 hours later, the biological solids concentration had risen to 9670 mg/l. It is clear from the results shown in Figure 4 that the system successfully responded to the shock and after 19 hours, biological solids concentration was purposely lowered to approximately the level at time zero (6500 mg/1), and the system was administered a very severe combined quantitative and qualitative shock loading. The feed concentration was changed to a total COD of 8600 mg/1, which consisted of two parts sorbitol-one part glucose. To assess the response to this shock loading, separate analysis was made for glucose and for sorbitol. It is

Figure 4. Effect of Various Types of Shock Loads on the Operation of the Extended Aeration Pilot Plant



TIME, HOURS

seen in Figure 4 that the maximum substrate leakage amounted to approximately 900 mg/l COD, 800 mg/l of which registered as sorbitol by the periodate test. In the early period after changing the feed concentration, there was also some small amount of glucose leakage from the system. Even at the maximum COD leakage, the overall efficiency of COD removal was approximately 90 percent. Over the next fifteen hours the system recovered rapidly, and since it was becoming clear that the effect of this change in type of substrate and concentration had been adequately monitored, the feed concentration was stopped and the system was operated as a batch unit for a few hours during which time the biological solids concentration which had risen to approximately 10,000 mg/l was again reduced to approximately the 6000 mg/l level in preparation for the next shock loading. Since it was amply apparent that the process could accommodate a considerably high quantitative shock load and an even more severe combined qualitative and quantitative shock loading, it was of interest next to assess its ability to take a hydraulic shock load. The inflowing feed was returned to glucose at approximately the 1000 mg/l level and the flow rate was doubled, i.e., the detention time was halved. It is seen that the process accommodated the shock load without any indication of unsteadiness in efficiency or settleability.

It is intersting to note that during the period of this hydraulic shock, the biological solids concentration in the process rose from 6140 to 7070 mg/l.

At various times during the Phase I studies small portions of sludge were taken to perform batch growth studies and to determine cell yield and endogenous oxygen uptake. A limited number of experiments was

conducted, and in some there was difficulty in measuring biological solids concentration. Therefore, only the approximate trend of the data will be mentioned here. In the growth studies (three were performed), all systems exhibited a lag of from 10 to 12 hours; the maximum specific growth rate was approximately 0.45 hr⁻¹--no reliable estimate for K_s could be obtained--but it did appear in all studies that it was well above 100 mg/l. Endogenous oxygen uptake rate amounted to 12 mg 0_2 /gm cells/hr during two measurements when sludge was exhibiting a rising trend and an examination made shortly after withdrawing 1200 ml sludge for hydrolysis (see day 80 on Figure 3), the endogenous uptake rate which was recorded for the sludge on day 84 was 29 mg 0_2 /gm cells/ hr. The average of the separate cell yield determinations was 0.58.

It can be seen in Figure 3 that at times, the effluent contained a considerable concentration of biological solids. For example, between days 35 and 45, biological solids concentration was, in general, between 100 and 200 mg/l. Since in previous studies in the OSU bioenvironmental engineering laboratories it was found that addition of flocculating chemicals sometimes assisted in clearing up an effluent, a decision was made to perform some flocculation studies relative to the effluent from this extended aeration pilot plant. Preparations were made to run these studies, but after day 50, the biological solids in the effluent dropped to very low levels and it appeared that the study would not be necessary. However, since the preparations for running the jar test had been made, and since it was anticipated that there would most probably be settling problems in the future, flocculation studies were made on the relatively good effluent which existed in the system on day 60. Various dosages of FeCl₃ (50-400 mg/l) were employed and the lowest

concentration of flocculating chemical used was successful in improving an already good effluent. At various times after day 60, ferric chloride was added as a slug dose when the biological solids concentration in the effluent was high. In all cases, immediately following the addition of ferric chloride, the effluent cleared up. However, in all cases the effect was very short-lived, indicating that continuous addition of flocculating chemical would be necessary. The practice was discontinued since it was of interest to determine whether the clarity of effluent would improve due to natural causes as the system was operated. However, another flocculation study (jar test) was made on day 93, when the biological solids concentration in the clarifier supernatant was nearly 200 mg/1. Both ferric chloride and aluminum sulfate were employed (at concentrations ranging from 10 to 50 mg/l). Both flocculating chemicals were effective, but alum appeared to give better results. The general trend of these results employing flocculating chemicals was similar to that observed by Yang, i.e., the effluent can be "cleaned up" by chemical flocculation but it would appear that it must be operated continuously for a protracted period after the onset of a turbid effluent.

B. Phase II

Studies on the Use of the Extended Aeration Process for Disposing of Hydrolyzed Secondary Sludge

This part of the study was devoted to an investigation of the feasibility of using a total oxidation system as a means of disposing of secondary biological sludge by hydrolyzing it and feeding it as a sole source of substrate to the extended aeration process.

In the early portion of this phase of the investigation, glucose was fed along with hydrolyzed sludge which consisted of excess sludge from a laboratory pilot plant. In later phases, hydrolyzed excess sludge from this pilot plant as well as from other pilot plants in the laboratory, was fed as sole source of substrate.

An extended aeration activated sludge was developed in the same manner as was the one described in Phase I. The synthetic medium was the same as that used in Phase I, except that carbon source and mineral concentrations were halved and 15 ml/l phosphate buffer was employed. In addition to 500 mg/l of glucose substrate, approximately 85 mg/l of hydrolysate COD was fed. On June 22, 1973, the biological solids concentration in the unit was 2200 mg/l, and this date was chosen as time zero and marked the beginning of an extensive monitoring period in which various parameters were measured in order to assess the performance of the system. During the period of this investigation, a colleague in the laboratory (R. Srinivasaraghavan) was performing experiments on a new design and operational model for activated sludge which had been recommended by Ramanathan and Gaudy (30). The flow diagram for this model was shown in the top of Figure 2. Aerator #2 acts as a sludge consistency tank so that the concentration of sludge in the recycle to aerator #1 can be maintained constant. It was the author's intention to determine if the waste or excess sludge could be biologically incinerated by hydrolyzing it and feeding it to an extended aeration unit as shown in the bottom portion of Figure 2. This possibility had been previously suggested by Gaudy and Gaudy (32). In attempting to gain some preliminary insight regarding this concept, the author's approach was to feed at first a mixture of glucose and hydrolyzed excess

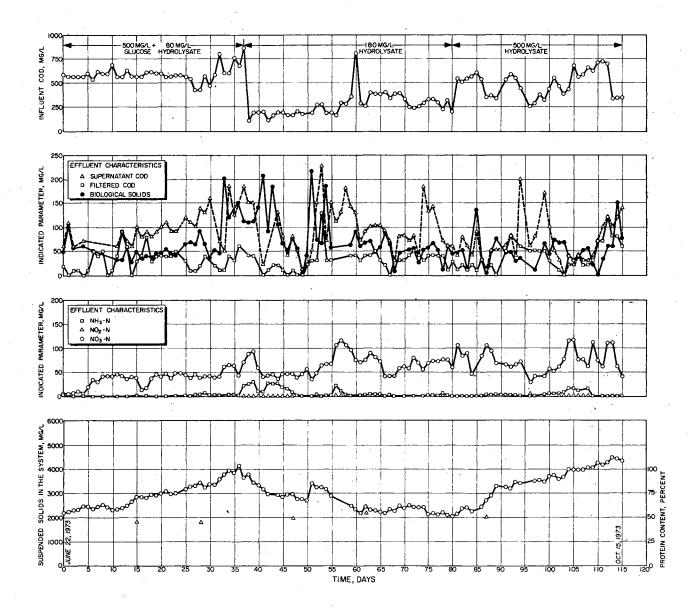
sludge, and then after a period of operation, take the system off glucose and feed only hydrolyzed excess sludge.

The results of the ensuing four months' study are shown in Figure 5. It is seen that during the first 37 days when the feed consisted of 500 mg/l glucose + 80 mg/l sludge hydrolysate, the performance may be adjudged satisfactory. The efficiency of COD removal based upon supernatant COD was 78 percent (ranging from 42 to 90 percent). The efficiency of COD removal based upon filtrate was 94 percent (ranging from 83 to 100 percent). During the first week of operation the system began to nitrify. It is seen that throughout the ensuing month, a rather highly nitrified effluent was produced, thus verifying the results presented in the previous section.

On day 37, the glucose in the feed was omitted and hydrolysate was increased to approximately 150-200 mg/l (see graph of feed at the top of Figure 5). The major purpose of this phase of the investigation was to determine if hydrolysate could be used as a sole source of carbon in a total oxidation system to produce a satisfactory aqueous effluent with regard to efficiency of removal of organic substrate and a highly nitrified effluent thus preventing the exertion of nitrogenous BOD in the receiving stream. During this period of the investigation, the buffer concentration was maintained at 15 ml/l. The mineral salts were as follows: MgSO₄·7H₂O, 30 mg/l; FeCl₃·6H₂O, 0.15 mg/l; MnSO₄·H₂O, 3 mg/l; and CaCl₂, 2.25 mg/l. The nitrogen source, ammonium sulfate, was added at a concentration of 150 mg/l. The system was operated at this loading level until day 80, and it can be seen in the figure that the effluent quality based on filtrate COD was rather good. The removal efficiency for filtrate COD was 89 percent, on average, and it ranged from 51 to

Figure 5. Performance of an Aerobic Digestion Pilot Plant Employing Pre-hydrolyzed Sludge as Feed Stock

-15.



100 percent. However, the removal efficiency based upon supernatant COD was 69 percent, ranging from a low of 31 to a high of 94 percent. The average biological solids concentration in the effluent was 77 mg/l, ranging from 27 to 220 mg/1. The effluent remained highly nitrified during this period. Early during the operation in this phase, there was a small amount of ammonia leakage in the effluent. It is interesting to note that the leakage of ammonia corresponded to a period of decreasing biological solids concentration in the aerator (see bottom graph of Figure 5). It is believed that the decrease in solids concentration is probably the result of two occurrences; first, the decreased organic feed coming into the unit could certainly be expected to cause more severe starvation conditions than had existed prior to stopping the glucose feed. Secondly, the rise in ammonia concentration even though the overall ammonia concentration in the feed had been reduced, indicates that the onset of this more severe starvation condition could possibly have enhanced lysis of some of the cells. In any event, this shock to the system did not seriously impair nitrification for any length of time, nor did it have any particular ill effects on the quality of the effluent. By August 9, 1973 (day 48), all of the excess sludge from the pilot plant runs of R. Srinivasaraghavan had been disposed of into this extended aeration process, and all other sludge fed during this phase of the operation, i.e., until day 80, was cell hydrolysate grown in a batch unit maintained especially for the purpose of feeding the process. It is seen that throughout this period, the results were such that use of the extended aeration process to treat cell hydrolysate obtained from excess cells in biological treatment units appeared to have some promise. It can also be seen that during

this phase of the operation, protein content (50 percent) was in general that of a viable healthy biological sludge.

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In the next phase of the study, a decision was made to increase the organic loading of cell hydrolysate in the influent feed to the process. Thus, on September 10, 1973, the feed concentration was increased to approximately 500 mg/l hydrolysate COD. All other constituents in the feed remained the same as they were in the previous phase. It is seen that during this phase of the investigation the biological solids concentration followed an increasing trend. The small batchscale reactor which was run to obtain hydrolysate was insufficient to supply the new feeding needs at the higher loading, and rather than increase its size, a decision was made to gather sludges from other pilot plants in the laboratory and use this extended aeration unit as a general sludge disposal facility for all of the excess sludge from other pilot plants operating in the laboratory. Thus, at times sludge from a trickling filter was hydrolyzed and fed, and at times various excess activated sludges were fed to the system. During this entire period of operation, i.e., from day 80 to day 115, the efficiency of substrate removal based upon filtrate COD was on average 91 percent, varying from a low of 75 to a high of 100 percent. Based upon supernatant COD, the average efficiency of COD removal was 83 percent, ranging from 54 to 98 percent; the system produced highly nitrified effluent. During this time most of the sludge which was fed to the unit was excess sludge from an activated sludge pilot plant being run for removal of COD by E. Stover. Also, as a point of interest, there were a few days in which the feed hydrolysate was derived from waste sludge from a laboratory trickling filter being operated by

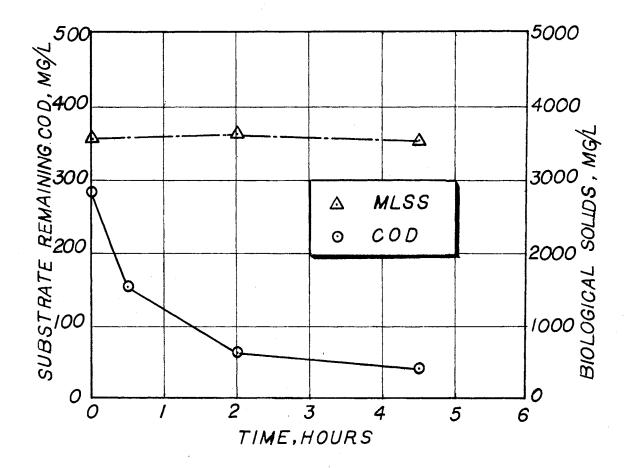
G. Marcangelli and T. Bentley. Also during this time, a small amount of sludge was fed with excess sludge from a laboratory pilot plant being run by R. Stall.

In summary, during the 115 days of operation, 140 liters of sludge at approximately 5000 mg/l solids were hydrolyzed and fed to this total oxidation unit.

It is recalled that P. Y. Yang, in conducting his Master's level research on the extended aeration process, and at times during his doctoral thesis research on the hydrolytically-assisted process, periodically stopped the incoming feed and fed an entire day's supply of feed as a slug dose or shock load. This had been done in order to assess the substrate removal capability of the sludge in the system at various times during the progress of the study, i.e., in his case, at various sludge ages. It was therefore of interest to gain some insight for comparative purposes regarding the removal kinetics when sludge hydrolysate was the sole feed to an extended aeration process. Accordingly, on day 98, the feed was stopped for 24 hours and the system was operated as a batch unit. After slug dosing with a day's supply of feed, biological solids concentration and filtrate COD were measured in order to gain some idea of the rapidity of purification. The results are shown in Figure 6, and they indicate a rapidity of removal fairly comparable to the rate observed in Yang's studies.

As with the previous phase of the investigation, small portions of sludge were taken at times to perform batch growth studies and to determine cell yield and endogenous oxygen uptake. As previously stated when reporting results of this type of study in the previous section, only a limited number of experiments were conducted and in some there

Figure 6. Performance of the Aerobic Digestion Pilot Plant During a Period of Batch Operation Consisting of 280 mg/l Hydrolysate COD



was difficulty in measuring biological solids concentration. Three growth studies were performed (on days 51, 64, and 84), and in all three systems exhibited a lag of approximately 10-12 hours, as before. Maximum specific growth rates recorded were, respectively, 0.49 hr⁻¹, 0.71 hr^{-1} , and 0.5 hr^{-1} . As with the previous experiments, insufficient data were obtained to determine an accurate estimate of K_{s} for these systems, but in general it appeared to be 100 mg/l or higher. Separate determinations for cell yield during batch growth studies were Only three such studies were performed, and the cell yield made. ranged from 0.35 to 0.6. The aim of these studies was simply to gain an idea as to the general similarity or difference in cell yield between this type of system and others which have been previously studied in the bioenvironmental engineering laboratory. On the basis of these studies, it would appear that one can expect the cell yield as well as other growth parameters such as $\mu_{\mbox{max}}$ and $\mbox{K}_{\mbox{s}}$ to be in approximately the same range for this type of system as it is for more conventional activated sludge systems. Endogenous oxygen uptake was also measured on these sludges during the same days in which the growth studies were made. These values were, respectively, 6, 12.1, and 13.0 mg 0_2 /gm cells/hr.

In this phase of the investigation on hydrolysate, it was also of interest to determine the utility of various chemical dosages to enhance flocculation of organic colloids in the effluent. Based upon results of the previous section, dosages of FeCl₃ and $Al_2(SO_4)_3$ ranging from 10 to 50 mg/l were applied to samples of effluent taken from the unit on September 27, 1973. In these jar studies, pH was adjusted to 6.6. The biological solids concentration of the effluent was 47 mg/l. It was observed that ferric chloride did not exhibit beneficial effect at any

of the dosages employed. However, aluminum sulfate produced an excellent cleared effluent even at the low dosage of 10 mg/l.

C. Studies on Conditions of Hydrolysis

In addition to the studies previously reported in this section, which were designed to examine the feasibility of the hydrolyticallyassisted process, it was of importance to gain further insight into the degree of solubility and biochemical composition of various biological sludges under different conditions of hydrolysis. A number of conditions were examined; for example, it was important to compare the degree of solubility for sludges hydrolyzed under acid conditions and under alkaline conditions. Also, it was of interest to determine if there were any gross effects of biological solids concentrations on the degree of sludge solubility after hydrolysis. Also, it was desirable to perform studies on a number of different sludges. In these studies the degree of solubility was determined by measuring biological solids concentration on the sludge sample before hydrolysis, and then running biological solids determination on the sample immediately after it was hydrolyzed and cooled. The membrane filter employed was a sterilizing filter, i.e., pore size was 0.45 nm. Also, the protein and carbohydrate content of the sample before and after hydrolysis was measured. Furthermore, it was of interest to determine if there was a loss of COD during the hydrolysis period. Biological solids were obtained by running two batch-activated sludge units; one was fed glucose (Unit A) and the other (type B) was fed bacto-peptone. A third type sludge (type C) was obtained from the secondary clarifier of a laboratory trickling filter which was at the time being operated by a fellow

student, G. Marcangeli. The results are shown in Table III. The results indicate that in general, alkaline hydrolysis appeared to be slightly better from the standpoint of dissolving the biological solids; however, it can also be seen that alkaline hydrolysis completely destroyed the protein in the sample. It is well known that alkaline hydrolysis of protein breaks down many amino acids. Also, it should be noted that at pH 12 if some of the amino acids were broken down completely, releasing ammonia, the ammonia would be stripped and lost from the system. Thus, from the standpoint of possible re-use of the nitrogen, alkaline hydrolysis would possibly be disadvantageous. On the other hand, for wastes containing an abundance of nitrogen, alkaline hydrolysis could effect some removal of nitrogen as ammonia. In general, acid hydrolysis decreased the protein content as measured by the biuret reaction. The decrease seemed to be severe with type A sludge, and no ready explanation is available. Also, with type A sludge there was a drastic decrease in carbohydrate content. However, this was not the case with types B and C sludge. It is also interesting to note that there were no decided trends in the degree of solubility with increasing or decreasing biological solids concentrations. For the type C sludge under conditions of acid hydrolysis, there was a decided decrease in percent solubility as biological solids concentration was. increased.

The table also shows the COD before hydrolysis and after hydrolysis. It is apparent both under acid and alkaline conditions, that during the 5-hour period in the autoclave at 15 psi and 121^OC there is a small but significant amount of digestion and/or stripping of some of the organic matter.

TABLE III

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EFFECT OF CONDITIONS OF HYDROLYSIS ON SLUDGE CHARACTERISTICS

Type of Sludge	Type A (Glucose-Batch Activated)						Type B (Bactopeptone Activated)						Type C (Sucrose-T			rickling Filter)			
рН	Ac	id pH =	1.0	Alk. pH = 12.0			Acid pH = 1.0			Alk. pH = 12.0			Acid pH = 1.0			Alk. pH = 12.0			
Concentration of Sludge, mg/l	2,750	5,500	10,000	2,750	5,500	10,000	1,950	3,900	5,850	1,950	3,900	5,850	4,500	9,000	18,000	4,500	9,000	18,000	
Biological Solids After Treatment, mg/l	-	2,700	3,180	450	200	1,570	1,160	330	470	940	80	770	1,200	2,980	9,040	-	-	_	
COD Before Treatment, mg/l	7,600	11,800	13,800	6,200	11,000	13,000	5,800	10,800	16,600	5,800	11,000	16,000	6,200	12,000	-	6,400	12,400	-	
COD After Treatment, mg/l	4,200	8,600	10,000	4,200	7,000	9,600	4,400	8,400	12,800	_	6,400	11,200	4,400	8,000	15,000	4,400	9,200	15,800	
% COD Reduced	44	27	27	32	36	26	24	22	22	_	41	30	29	33		31	25		
<u>% Solids Reduced</u>	-	_ 50	68	83	96	84	40	91	91	51	97	85	73	66	49		-		
Protein Content % Before Treatment	66						73						50						
Protein Content % After Treatment	23			•	-			57			_			44			-		
Carbohydrate Content % Before Treatment	34						18					8							
Carbohydrate Content % After Treatment	18				22			22			21			10			10		

46

total oxidation of excess sludge when the sludge fed to the system has been previously hydrolyzed. Thus, such a process may be able to serve as one employed solely as a means of disposing of excess sludge for various types of biological treatment processes. The Phase II results offer some indication that total plant flow sheet suggested by Gaudy and Gaudy (32) offers promise for the future.

In both Phase I and Phase II the efficiency based on filtrate COD was very good, but with regard to total COD based upon clarifier supernatant, there were times when the degree of treatment was not satisfactory because of organic suspended solids carryover. Such carryover was usually not due to rising or bulking sludge, but more to the nonclarity of the supernatant. Thus, with this system as with all other biological treatment systems which depend primarily on the quiescent sedimentation process, there may be need at times to "back up" the final clarifier with other separation processes to "polish" the efflent, e.g., filtration, chemical dosage, etc.

The results of the auxiliary growth studies indicate that the general characteristics of these biomasses were similar to those previously observed for hydrolytically-assisted extended aeration activated sludges. Although only a few experiments were run, the μ_{max} , K_s , and cell yield values were within the range of values previously observed. Also, this sludge exhibited a lag in the growth studies wherein a small inoculum was employed at various initial substrate concentrations, and the endogenous 0_2 uptake was "typical" of those previously found. It was in general higher than that for a normal or nonhydrolyticallyassisted extended aeration process, and lower than the values found for a young cell suspension.

CHAPTER V

SUMMARY AND CONCLUSIONS

The present investigation was an extension of a long-term study in a the bioengineering laboratory of Oklahoma State University into the theory of total oxidation of organic biomass and application of the theory to the extended aeration process. The latter aspect led previous workers in this laboratory to the engineering concept of the "hydrolytic assist." The present study was addressed primarily to further study of the "hydrolytic assist" and it is felt that it has added significant data in substantiation of the utility of the process and has extended the knowledge in regard to two very important areas. Phase I provided a definite indication that the process could be successfully employed at loadings which would be considered rather high for the extended aeration process. The 500-1000 mg/l feeding level corresponds to 47 and 94 lbs COD/1000 cu ft of aeration volume per day, whereas the usual loading for the extended aeration process is 15 lbs $BOD_5/1000$ cu ft of aeration volume per day (31). Furthermore, the results indicate that even at the 1000-mg/l feeding level a rather highly nitrified effluent can be produced. Thus, the advantageous nitrification tendencies of the extended aeration process are maintained with the hydrolyticallyassisted process at high organic loadings.

Phase II results provided an indication that an extended aeration process can serve as an aerobic digestion unit providing essentially

In general, the studies were made to gain fur ther insight in possible ways to hydrolize the biomass tended to affirm the use of the acid hydrolysis process and the conditions of time, pressure, and temperature currently being employed in the pilot plant work.

Although by the criteria of low endogenous O₂ uptake and apparent growth lag, this extended aeration sludge would be adjudged less active than sludges of younger "age," the results of the shock loading studies indicated a rather large capacity to assimilate various types of shock. Particularly interesting was the result of the hydraulic shock load. A halving of the mean hydraulic retention time had little or no effect upon the quality of filtrate COD, which was indeed an expectable result, but the fact that it had no effect on total COD seems especially significant in view of the fact that hydraulic shock is usually expected to have pronounced effects on settleability of the sludge. It should be remembered, however, that the settling time in the clarifier was rather long in any case, i.e., eight hours prior to the shock and four hours during the shock.

The general conclusion after completing this investigation is that the "hydrolytic assist" either to an extended aeration process or as a means of enhancing an aerobic sludge digestion process, offers considerable promise. There are, however, various avenues for further research which should be pursued, and some of these are given in the following section.

CHAPTER VI

SUGGESTIONS FOR FUTURE WORK

1. Although the results of the shock loading studies of this investigation indicate the system is extremely stable with respect to substrate leakage, it should be emphasized that studies on shock loads performed in this work were essentially of a preliminary nature. It would seem advantageous to mount more exhaustive studies on this aspect.

2. In the interest of gaining further insight regarding the hydrolytically-assisted extended aeration process, more studies should be made at various feeding levels and hydrolysis schedules to determine the average balance levels of biological solids concentration which develop at the various feeding and hydrolysis schedules. Operation at pre-selected biological solids levels with determination of the hydrol-ysis schedule required to satisfy the pre-selected condition would also be of value and interest regarding the process.

3. Regarding the hydrolytically-assisted extended aeration process for the treatment of waste streams, it would appear advisable to perform more studies on various whole wastes, such as those made by Scott on pulp and paper waste. Also in line with the studies on Phase II of the present investigation in which the aim was to determine if excess hydrolyzed sludge would be subject to essentially total oxidation in an aerobic digestion unit, it would be ideal to follow up such studies using actual secondary sludge from a treatment plant in the field rather than laboratory-developed biological solids.

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Homayoon Saidi

Candidate for the Degree of

Master of Science

Thesis: STUDIES ON THE HYDROLYTICALLY-ASSISTED EXTENDED AERATION PROCESS AND ON PRE-HYDROLYSIS OF SLUDGE IN AEROBIC DIGESTION PROCESSES

Major Field: Bioenvironmental Engineering

Biographical:

- Personal Data: Born in Rudsar, Iran, December, 30, 1945, the son of Mehrandokht-Arian Negad and Hasan Saidi.
- Education: Graduated from No. 1 Hadaf High School, Tehran, Iran, in September, 1964; completed requirements for the Licentiate Degree of Engineering from Tabriz University, Tabriz, Iran, in December, 1968; completed requirements for the Master of Science degree at Oklahoma State University in May, 1974.
- Professional Experience: Military service in Iranian Extention Corps, May, 1969, to May, 1971; supervisory engineer, December, 1968, to May, 1969; senior engineer, May, 1971, to October, 1971; research assistant in the bioengineering laboratories, Oklahoma State University, Stillwater, Oklahoma, September, 1972, to May, 1974.

Member in Professional Societies: Water Pollution Control Federation.