

THE UNIVERSITY OF MANITOBA

BIOKINETICS OF LOW TEMPERATURE
WASTE ASSIMILATION

by

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A THESIS

SUBMITTED TO THE FACULTY OF GRADUATE STUDIES
IN PARTIAL FULFILMENT OF THE REQUIREMENTS FOR THE
DEGREE OF DOCTOR OF PHILOSOPHY

DEPARTMENT OF CIVIL ENGINEERING

WINNIPEG, MANITOBA

OCTOBER 1976

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BRIAN H. TOPNIK

A dissertation submitted to the Faculty of Graduate Studies of
the University of Manitoba in partial fulfillment of the requirements
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ABSTRACT

During the winter, in northern countries such as Canada, it is not uncommon for sewage to approach freezing temperatures. To simulate low temperature wastewater treatment, a laboratory investigation was conducted on a 20 litre per day continuous-flow extended aeration plant at temperatures ranging from 20°C to 0°C using typical raw domestic sewage as feed. The kinetic growth and substrate utilization parameters were determined after steady state operation at a given temperature. The kinetic constants were calculated from straight line equations based upon the fundamental Monod, and cell and substrate mass balance equations.

After a continuous test period of 261 days, no apparent temperature or substrate concentration effects were noted for either the growth or substrate utilization constants. The following are the average kinetic constants as a result of this investigation.

- (a) yield constant, $y = 0.33 \frac{\text{wt VSS}}{\text{wt BOD}_5}$;
- (b) decay constant $k_d = 0.97 \times 10^{-3} / \text{hr}$ based on BOD_5 ;
- (c) half-velocity coefficient = $K_s = 6 \text{ mg/l}$ based on BOD_5 ; and,

- (d) maximum substrate utilization rate, $\mu_{\max} =$
 1.27×10^{-3} /hr based on BOD₅.

The overall BOD₅ removal rate of the unit followed a zero-order relationship given as

$$K_t = 3.62 \times 10^{-4}T + 2 \times 10^{-3},$$

where K_t = BOD₅ removal rate (day^{-1}); and

T = temperature, °C.

Nitrification was found to occur at all temperatures including 0°C at which temperature approximately 18% of the feed ammonia nitrogen was oxidized to nitrate.

The oxygen uptake rate was found to be temperature dependent in accordance with the equation given below.

$$\text{Oxygen Uptake Rate} = 5.7 T^{0.323},$$

where oxygen uptake rate is in mg/l/hr.

The apparent high BOD₅ and COD removal efficiencies at low temperatures (90% and 91% at 0°C respectively) were considered to have occurred due to cellular food storage rather than due to cell replication. This theory was substantiated by other test parameters such as mixed liquor and effluent solids concentrations, settleability, turbidity, nitrification, and total phosphorus removals.

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BIOKINETICS OF LOW TEMPERATURE

WASTE ASSIMILATION

1. INTRODUCTION

With ever increasing public concern about the environment, it is becoming increasingly important that waste treatment technology keep pace with the demands for more complete and economical treatment. This includes waste treatment techniques currently practiced as well as the development of new concepts.

1.1. STATEMENT OF THE PROBLEM

Although many forms of waste water treatment have been developed over the years, one that has been used with considerable success since the turn of the century is the biological process developed in England known as activated sludge (1)*.

* The numbers in parentheses in the text indicate references in the Bibliography.

The activated sludge process is one where wastewater is aerated in the presence of a large quantity of activated sludge "floc" for a predetermined time and in the presence of dissolved oxygen. The mixture is then settled in a settling tank where the solids flocculate, settle out, and are recovered for reuse in the aeration tank with excess sludge being wasted.

The most significant part of the activated sludge process is the role played by the microorganisms which make up the "floc". To design and operate an activated sludge system efficiently, it is of utmost importance to understand the operating conditions which will optimize the microorganisms' growth and replication.

The key role of bacteria in nature is to decompose organic matter. This is also true in the activated sludge process where bacteria are thought to be the most important microorganisms responsible for degradation of organic matter in the waste influent (2) (3). The bacteria metabolize organic material to produce protoplasm, i.e. new cells. This is done by breaking down the organic molecule and synthesizing complex protoplasm. This requires energy and it is derived from the organic being metabolized.

Although the bacteria actually degrade the organic waste, the metabolic activities of other microorganisms, such as protozoa which feed on the bacteria, are also significant

(3)(4). As with all living organisms, environmental conditions will determine how successful this microbial ecosystem will perform, i.e. degrade the organic waste. In activated sludge, some of the more important environmental conditions are pH, nutrients, trace elements, toxic elements, acclimation, and temperature.

Metcalf and Eddy (2), McKinney (3) and many others have stated that, "one of the most important factors affecting microbial growth is temperature". As well as physiological effects on the activated sludge process, temperature will also affect physical properties of the process such as gas transfer rates and settling characteristics of the biological solids.

Generally, the microbial growth, and hence waste purification potential, will decrease as the temperature is decreased. This fact is of paramount importance in the design of biological waste treatment facilities for operation at temperatures approaching freezing. The activated sludge process is often applied to waste treatment under freezing conditions. It therefore becomes necessary to have an indepth understanding of the microbiological behaviour at temperatures approaching 0°C.

1.2. REASON FOR STUDY

In engineering processes involving chemical reactions, it is well known that temperature has an effect on reaction rates. This is often approximated by an abbreviation of the Van't Hoff-Arrhenius equation by saying that the rate doubles for each 10°C rise in temperature (5). This indicates that temperature exerts an effect on the reaction in accordance with some mathematical power rule. This same approach is often applied to biological systems by stating that the temperature effects tend to follow a first order equation dependent on a temperature-activity coefficient often symbolized as θ (2)(5)(6). The temperature coefficient, θ , is somewhat limited in practical universal application because of its variation with temperature (5)(6)(7)(8). Temperature studies on activated sludge process behaviour in the past has largely been concentrated at temperatures near 20°C , although work by Carpenter and others (7) included data down to 2°C . Grube and Murphy (9) found that temperatures in an oxidation ditch in Alaska did not drop below $+1.9^{\circ}\text{C}$ during a period of intense cold weather (Air @ -32°C) and low sewage flows. In spite of the fact that the influent appears to be able to maintain greater than $+2^{\circ}\text{C}$ temperature in a waste treatment facility, it does not ensure that 0°C or even colder temperature will not occur.

Studies carried out on biodegradability of wastes are most often performed in the laboratory using synthetic sewage and batch process type of flows (10)(11)(12). The data obtained from this type of laboratory experiment is then used to predict performance in full scale operating treatment units. This approach has been criticized by some investigators both favourably and otherwise (13)(14). It would seem reasonable to assume that the closer conditions in the laboratory simulate operating conditions in the field, the more realistic the results would be. For this reason activated sludge behaviour investigations for this study were performed using accepted sanitary engineering design criteria as laid out by Great Lakes-Upper Mississippi River Board of State Sanitary Engineers commonly known as 10 State Standards (15). A laboratory model of the continuous flow extended aeration modification at the activated sludge process was designed using typical raw domestic sewage as feed.

Design and control of the activated sludge process in the past was largely empirical and often became a "hit and miss" affair. Recently, investigators such as Lawrence and McCarty (16) have developed mathematical models to describe behaviour of biological waste treatment processes. At the present time limited information is available with regard to

kinetic constants which describe these mathematical models, especially for specific applications of a given biological process, such as extended aeration (2). There is even a greater lack of information relating the effects of temperature on these kinetic constants.

The extended aeration modification of the conventional activated sludge process has recently been applied to waste water treatment in the North (17)(18)(19). The documented operational problems of these cold weather applications suggests that a lack of knowledge exists for cold temperature biological waste treatment. This warranted an indepth study of the kinetic behaviour of extended aeration waste water treatment at near-freezing temperatures.

2. PURPOSE AND EXTENT OF INVESTIGATION

2.1. PURPOSE

The purpose or the objectives of this investigation may be categorized as primary or secondary as follows:

2.1.1. Primary Objectives

(1) To determine the Monod growth and substrate utilization kinetic given by the sludge yield coefficient, Y , the endogenous decay coefficient, k_d , the substrate utilization rate coefficient, μ_{max} , and the half-velocity coefficient, K_s , as a function of temperature from 20°C to 0°C using a continuous-flow laboratory extended-aeration plant and typical raw domestic sewage as feed.

(2) To determine the effect of low temperature on the rate of substrate removal efficiency for the laboratory extended-aeration process.

2.1.2. Secondary Objectives

(1) To determine the effect of low temperature on the 5-day biochemical oxygen demand test.

(2) To determine the effect of low temperature on nitrification at long detention times.

(3) To determine the effect of low temperature on the extended aeration oxygen uptake rate.

2.2. EXTENT OF INVESTIGATION

This investigation of low temperature extended aeration waste treatment was conducted in a walk-in environmental chamber capable of controlling the temperature within $\pm 0.5^{\circ}\text{C}$. The laboratory continuous flow extended aeration unit was constructed from acrylic plastic and was designed in accordance with 10 State Standards (15) to treat approximately 19 litres/day of screened raw domestic sewage obtained from the City of Winnipeg's South End Water Pollution Control Center. Although continuous flow in the true sense was not actually obtained, it was approached by feeding a calibrated amount of raw sewage intermittently from a reservoir which was stirred using a magnetic stirring device. The tests for this study were conducted for a period of 261 days beginning March 27, 1975 to December 16, 1975. The raw sewage feed for the unit was collected every Friday morning at about 11:00 a.m. Samples were stored in the environmental chamber at the temperature at which the test unit was operating.

The test extended aeration unit was operated at temperatures ranging from 20°C down to 0°C and then up to 10°C .

The test unit was attended to daily during the 261 day period and the following daily tests were conducted:

- (1) BOD on feed and effluent;
- (2) Suspended and volatile suspended solids on feed, mixed liquor and effluent;
- (3) Effluent turbidity;
- (4) Temperature;
- (5) Feed flow;
- (6) Sludge volume index; and
- (7) Feed and mixed liquor pH.

Other tests that were run intermittently throughout the test period included:

- (1) COD on feed and effluent;
- (2) Ammonia, organic and nitrate nitrogen on feed and effluent;
- (3) Total phosphorous on feed, mixed liquor, and effluent;
- (4) Oxygen uptake on mixed liquor.

3. LITERATURE REVIEW

In order to maximize the efficiency of any process, it is absolutely necessary to be completely familiar with the basic fundamentals upon which the process operates. This is true for a pure chemical reaction or a biochemical reaction such as the activated sludge wastewater treatment process. Basically this research was conducted to evaluate the behaviour of activated sludge at freezing temperatures. A thorough evaluation of the data obtained required an indepth study of the fundamentals of biological treatment. This was done via a literature search of work done by other investigators.

3.1. BASIC MICROBIOLOGICAL CONCEPTS

Biological treatment processes can be described by their oxygen dependency. When the system operates in the presence of free dissolved oxygen, it is known as aerobic. An anaerobic system is one where the terminal electron acceptor is chemically bound oxygen and operates in the absence of free dissolved oxygen. The discussion of any biological system must include some mention of bacteria, considered the basic life form in all types of biological waste treatment (2) (3) (20) (14).

3.1.1. Bacterial Growth Patterns

Bacterial growth patterns as they relate to sanitary engineering have been described most clearly by McKinney (3). The two ways McKinney describes the bacterial growth with respect to time are numbers of microorganisms or mass of microorganisms.

A typical pattern relating numbers of organisms versus time is shown below as Figure 1.

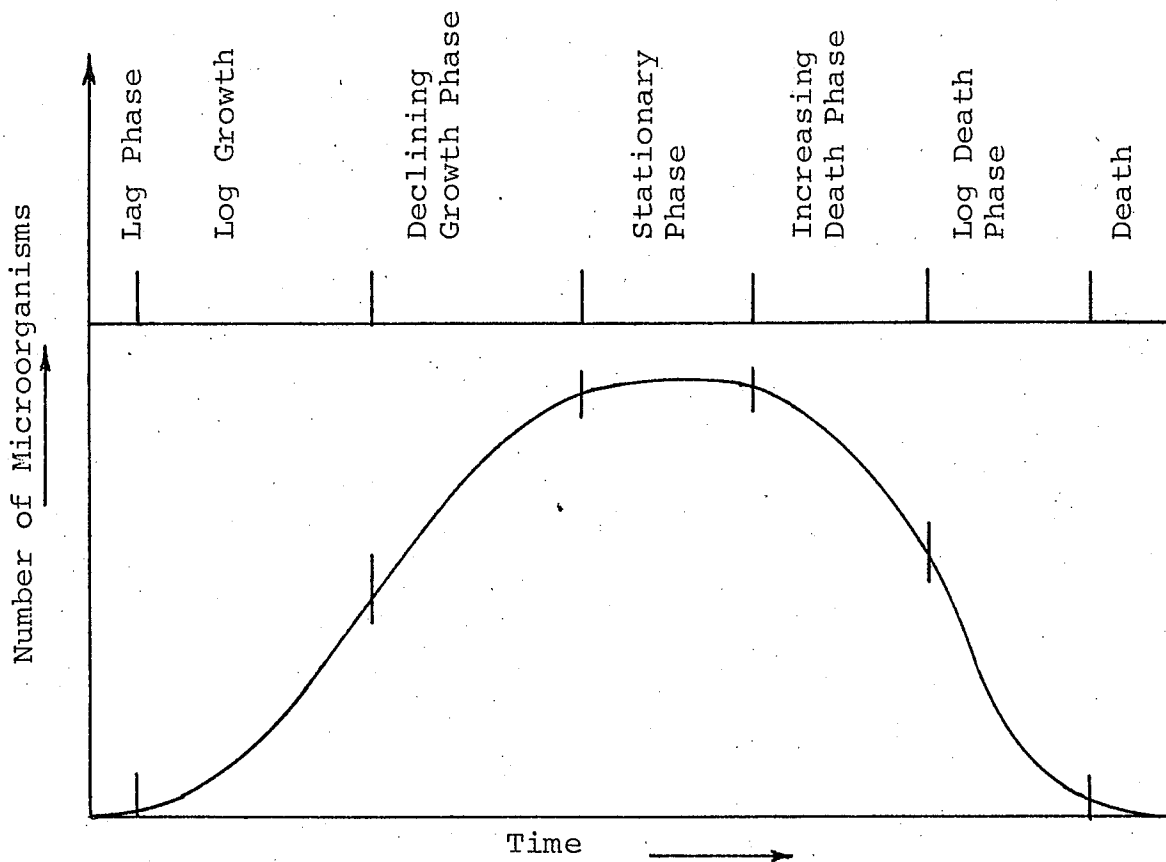


Fig. 1. Growth Pattern Based on Numbers of Microorganisms (after McKinney (3)).

This growth curve shows seven separate phases and is most suited to batch cultures involving small numbers, e.g. inoculated cultures. The complex heterogeneous cultures found in activated sludge usually are evaluated in terms of mass rather than numbers. A typical curve is shown below as Figure 2.

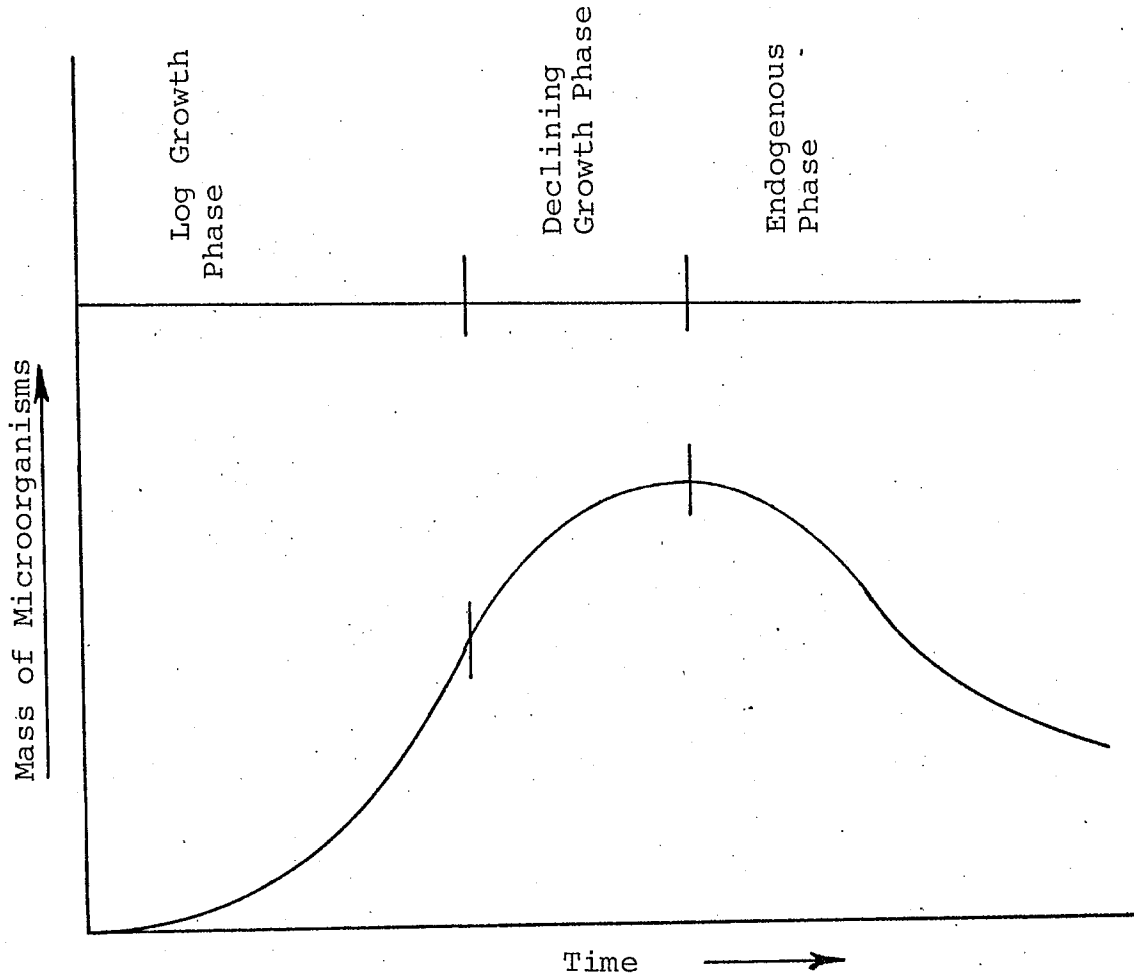


Fig. 2. Growth Pattern Based on Mass of Microorganisms (after McKinney (3)).

Figure 2 shows three main growth phases. A lag phase, not shown, may or may not be present. The apparent similarity between the pure culture and the complex culture is a most significant observation when comparing the two curves. The fundamental assumption, that the complex cultures behave similarly to the pure cultures, is often used by scientists to predict field performance from laboratory studies.

In the log phase an excess of food is always available to the microorganisms. The growth rate is at its maximum at the end of this phase. Since the rate of substrate uptake is so rapid throughout the log phase, it would appear that it would be desirable to operate at this point. This is not necessarily true, because the excess food which stimulates the rapid growth would show up in the effluent. This rapid growth condition also precludes efficient flocculation and clarification.

In the declining growth phase the plentiful food supply is no longer available. The rate of growth decreases as the food supply diminishes and the system energy begins to decline. This lower energy state allows the flocculation and settling of bacterial cells. The latter portion of this growth is therefore the point at which conventional activated sludge processes are usually operated. It may also be noted that at this point the sludge production is highest, which may present a solids handling problem.

The endogenous respiration phase is the one that is of greatest significance for this particular study. The food supply is at a minimum at this point; in fact, it may be considered to be in equilibrium with the microorganisms. McKinney (20) has defined endogenous respiration as the basal energy requirement of the bacteria, where basal energy is the small amount energy required for motion and enzyme activity. The basal energy requirement is thought to be a continuous reaction resulting in the metabolism of protoplasm. The bacteria utilize their own cellular constituents in this phase in order to maintain life (21). The total mass as a result of this, will tend to decrease and because of the limited food supplies, energy is at a minimum. This creates a condition suitable to settleability of biological solids desirable in the waste treatment process.

The mass decreasing tendency of endogenous respiration might lead to the notion that a system could continue to consume itself so that no excess sludge would need to be wasted, i.e. total oxidation. However, as Busch and Myrick (14) and others (20) (21) (22) (23) (24) have noted, sludge accumulation does in fact occur even with a long detention period.

3.1.2. Oxygen Relationships

The basic processes that describe activated sludge can be given as: (25)

(a) basic respiration and metabolism of aerobic bacterial cells;

(b) mass transfer of nutrients in the substrate and dissolved oxygen to the cell surface and removal of the metabolic waste products;

(c) the solution of molecular oxygen into the waste liquid; and,

(d) the flocculation of individual microorganisms into a settleable mass.

It is of significance that three of the four basic assumptions refer directly to the word oxygen. The fourth assumption indirectly refers to oxygen in that "settleable mass" often is expressed in terms of oxygen equivalence (26). It becomes readily apparent that the role of oxygen in the activated sludge process is of importance.

McKinney (20) offers the following three arguments for the use of the "oxygen equivalent scale" for evaluating system kinetics.

(1) The five-day biochemical oxygen demand test is used as a measure of the organic matter being metabolized;

(b) The oxygen uptake is related to the energy expended for both synthesis and endogenous respiration;

(c) The oxygen equivalent of protoplasm can be obtained from a chemical oxygen demand analysis.

The basic equations describing the activity in biological treatment are given by McKinney (20) as follows:

Organic matter metabolized = protoplasm synthesized +
energy for synthesis; and,

Net protoplasm accumulated = protoplasm synthesized -
endogenous respiration.

These equations may be written in differential form as follows:

$$\frac{dF_m}{dt} = \frac{dM_s}{dt} + \frac{dO_s}{dt} ; \text{ and,}$$

$$\frac{dM_p}{dt} = \frac{dM_s}{dt} - \frac{dO_e}{dt}$$

where:

F_m = ultimate oxygen demand of organic matter being metabolized;

M_s = oxygen equivalent of protoplasm;

O_s = oxygen uptake for synthesis;

M_p = oxygen equivalent of net protoplasm; and,

O_e = endogenous respiration oxygen uptake rate.

The concept of using oxygen equivalence to relate energy and synthesis was investigated in greater detail by Burkhead and McKinney (27). A relationship between the heat of reaction, which is related to heat loss and oxygen utilization was established. The authors' conclusion was that the findings supported McKinney's (20) energy-synthesis concept based upon oxygen equivalence. The concept of using thermodynamic properties such as free energy changes and enthalpy to predict growth was objected to by Banerji (28) and others (29) (30). Open systems such as activated sludge are too complex and depend on too many environmental factors to apply exact laws such as given by classical thermodynamics. The non-standard approach used by investigators and the less than ideal method of measuring growth tends to favour empirical methods for establishing process kinetics.

In an aerobic system free dissolved oxygen must be supplied to the microorganism. The oxygen mass transfer depends upon the coefficient of diffusion and the concentration gradient. Kalinske (25) quotes Wuhrman as stating that the limiting factor in the activated sludge process may not in fact be free dissolved oxygen, but rather the nutrient concentration of the mixed liquor. This fact could lower the required free dissolved oxygen concentration for optimum operation. Ten State Standards (15) suggest that the minimum dissolved oxygen in the aeration tanks not

be less than 2 mg/l at any time. In the same reference (25) Mueller's work indicated that the oxygen uptake rates depend on the diffusion of oxygen through the floc matrix and this may be the controlling factor in oxygen utilization.

It would appear that the oxygen concentration does not necessarily influence the rate of bacterial respiration. However, in heavy floc suspensions this may not be true because localized volumes of oxygen depletion may occur as a result of inadequate agitation.

Rickard and Gaudy (31) reviewed the work done on agitation and microbiological activity by Tsao and Kempe, Imhoff, von der Emde, Pasveer and Zahradka. In their own experiments, Rickard and Gaudy determined the effect of agitation on the growth and substrate utilization of continuous flow heterogeneous batch systems. The results of their work indicated that an increase of oxygen uptake and decrease in solids yield occurred with increasing velocity gradient. Explanations for this phenomenon were given as:

- (a) changes in microorganism predominance;
- (b) increased frequency of contact between cells and substrate;
- (c) production of smaller floc particles with resultant improved penetration of substrate and oxygen i.e. increased surface area;

(d) increases in the rate of oxygen transfer across the cell-liquid interface; and,

(e) maintenance of a higher dissolved oxygen concentration (at all points in the system) with increasing turbulence.

Generally, it was thought that the increased oxygen uptake with increased turbulence probably resulted from reductions in the cell-liquid interface resistance to oxygen transfer as the amount of mixing energy increased. The decrease in biological solids that occurred primarily at the expense of cellular carbohydrate was believed to have resulted from an increased ratio of respiration to synthesis.

This same phenomenon was noted by Pasveer (32) using theoretical considerations and later verified experimentally (33). The test results were summarized as follows:

"...as a rule it will merely be necessary to increase the oxygenation capacity and degree of turbulence by increasing the supply of oxygen to the floc and accelerating the biochemical process ... the decreasing floc size is of major importance..."

The effect of turbulence on microorganism activity was also noted by Ali and Beutra (34) on biochemical oxygen demand studies. BOD progression studies on stirred and unstirred samples consisting of synthetic media, raw, settled and final wastewater, showed a significant 17 percent

increase in 5-day BOD when stirred. The deoxygenation rate constants increased in all samples except for the final effluent. Possible reasons for increased microbiological activity in the stirred samples were given as:

(a) increased rate of bacteriological activity and multiplication with increased turbulence;

(b) turbulence increases the rate of material transport into the cell and the rate of removal of by-products accumulating on the cell membrane; and,

(c) turbulence increases the contact between bacterial cells and substrate increasing the rate of assimilation.

Although turbulence appears to play several roles in wastewater purification, it very definitely appears to be significant in the all important cell oxygen delivery scheme. Kalinske (25) refers to agitation more in terms of micro scale, i.e. micro-turbulence that controls convective diffusion on a scale comparable to the floc size. The author (25) indicates that according to air requirements derived from standard design criteria enough oxygen is not available to provide a sufficient amount of agitation for obtaining maximum mass transfer conditions to the microbiological cells.

McKinney (20) had noted and documented the advantages of the completely mixed activated sludge process. By definition a completely mixed system is one where the

untreated wastes are instantaneously mixed throughout the entire aeration tank. The fact that this mixing occurs over a relatively short time frame implies that micro-turbulence conducive to increased microorganism activity is probably occurring.

3.1.3. Nitrogen and Phosphorous Considerations

Nutritional requirements for biological growth are extremely complex and can be broken into two major categories which include macro and micro nutrients. The macro nutrients receiving most attention are carbon, nitrogen and phosphorous. A deficiency of nutrients is usually not a problem with domestic wastes (3). Generally an ample supply of necessary nutrients are present to support microorganism growth. This certainly is not always true for industrial wastes. Helmers et al (35) (36) operated laboratory activated sludge units using industrial wastes. These units were fed three times daily and were operated at a solids concentration of 1500 mg/l and detention time of 7 hours. The following are conclusions of this particular study.

(a) critical nitrogen requirements based on BOD removal were estimated to be 3, 4, and 3 pounds per 100 pounds of BOD removed at 10°C, 20°C and 30°C respectively;

(b) phosphorous requirements under similar conditions were 0.6, 0.7, and 0.5 pounds per 100 pounds of BOD removed at 10°C, 20°C, and 30°C; and,

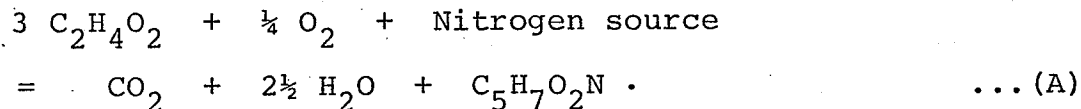
(c) the percent nitrogen content of dried activated sludge based on volatile matter is a good index of nitrogen deficiency. A value of less than 7 percent is indicative of a critical deficiency.

This work agrees favourably with accepted nutrient ratios of C:N:P of 100:5:1 (2) (3).

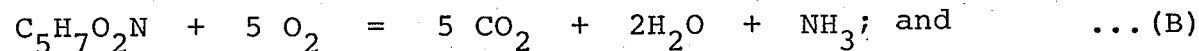
The empirical nutrient relationships have received further attention by Symons and McKinney (37). Their basic equation for the removal of organics is written as:

Total organic removed = oxidation + synthesis

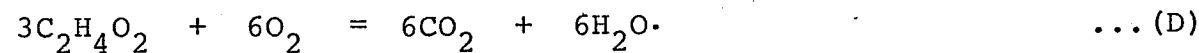
Using acetate as an example



Equation (A) is followed by:



Summation of equations (B) and (C) gives



Equation (D) states that all the carbon has been converted to CO_2 and that no additional nitrogen is required when the system has reached equilibrium. It also implies that no sludge accumulation occurs at equilibrium. Symons and McKinney (36) have put forth the theory that,

"...inasmuch as all of the organic transformations that take place inside a bacterial cell, except one, the production of certain polysaccharides, are reversible, all but one compound that a cell synthesizes can be degraded."

This theory was supported by microscopic examinations of Alcian-blue stained sludge slides which showed that the material which had accumulated was predominantly polysaccharide material.

The concept of using nitrogen to describe the system performance was developed further by describing the significance of nitrogen concentration in the wastewater feed.

Case I (High concentration)

A high concentration was assumed to be about 50 mg/l as N. After continued operation at this level, an increase of total solids mass was discovered. This was accompanied by a steady increase in protoplasm (organic nitrogen in the sludge) and an increase in polysaccharide (decrease in percent

nitrogen in the sludge).

Case II (Moderate concentration)

A moderate concentration of nitrogen in wastewater was assumed to be 12.5 mg/l. After continued operation the protoplasm concentration, as measured by the organic nitrogen in the sludge, was found to be constant. An increase of total solid mass was determined and was associated with an increase of polysaccharide (declining percentage of organic N in the sludge).

Case III (Nitrogen absent)

Continued operation resulted in a solids increase due to an increase of polysaccharides. The protoplasm as measured by organic nitrogen decreased indicating a dying system.

It becomes obvious that sufficient nitrogen and phosphorus concentration above the critical levels must be present for satisfactory operation of an activated sludge plant. A nitrogen-deficient system may be recognized by way of microscopic checks of the sludge. Fungi do not require as much nitrogen for the manufacture of protoplasm. This fact will tend to favour fungi growth over normal flora at low nitrogen concentrations (3). Fungi will remove

organic substrate from the wastewater in a manner similar to bacteria, but the filamentous growths often preclude satisfactory settling. Good settling is essential for proper operation of the secondary clarifiers in an activated sludge process.

Recent research by Sherrard and Benefield (38) describe a method that may be used to account for wastewater carbon, nitrogen and phosphorous and to specify the form in which each may be found after treatment. Basically the kinetics are based upon the mean cell residence time concept and equations set forth by Lawrence and McCarty (16). A mass balance is computed for each nutrient in the system as follows:

(a) carbon exists as effluent COD, waste sludge and CO_2 ;

(b) nitrogen appears as waste sludge, ammonia or nitrate; and,

(c) phosphorous is incorporated into the sludge produced from the carbon and nitrogen or it passes into the effluent. The sludge composition is assumed to be constant at $\text{C}_{60}\text{H}_{87}\text{O}_{23}\text{N}_{12}\text{P}$.

The concept of using growth kinetics to estimate the fate of nutrients in a biological system is of value. The greatest problem in using the mathematical kinetic model approach is to establish the growth constants.

3.1.4. Cell Growth Measurement

In biological processes the performance level is dependent upon the microbiological growth within the system. It is necessary therefore, to be able to measure this growth quantitatively. The complexity of the activated sludge process makes it difficult to obtain an exact measure of the amount of active cell tissue held in a system. It is general practice today that,

"...because of the simplicity of the test procedure involved, it has become standard practice to measure the weight of volatile suspended solids (VSS) contained in a system and assume that this weight approximates the amount of active organism tissue present." (39)

There is considerable controversy about using volatile solids as a measure of microbial growth. Jenkins et al (40) have stated that within the common operating range of the activated sludge process the use of volatile suspended solids as a measure of cell material does not cause serious errors in kinetic reactions. They did not find this to be true for high rates of operation.

Banerji (28) concluded that unless time-growth measurements are standardized and a more meaningful growth parameter other than mixed liquor suspended solids or

volatile suspended solids is adopted by the industry, discrepancies in sludge yield data will remain.

Eckenfelder (41) discussed the problem of using volatile solids in the calculation of growth kinetics. Stewart (39) mentioned that it is inevitable that a better parameter than volatile solids will be used to describe cell mass. He suggests that the use of deoxyribonucleic acid (DNA) would possibly be a more accurate measure of microorganisms since the amount of DNA present in the system gives a fairly accurate measure of the amount of active cell tissue present in the system. Genetelli (42) investigated DNA and nitrogen relationships in activated sludge. Hartman was quoted as reporting that the weight of nitrogen in the cells was a better estimate of viable organisms in the mixed liquor than volatile suspended solids. Symons and McKinney (36) similarly have used cell nitrogen to predict process biomass performance. Genetelli found that DNA was a more sensitive indicator than organic nitrogen for predicting cell mass. Using sludge nitrogen was not ruled out as being satisfactory as long as the waste water feed was low in organic nitrogen.

Another technique that has been investigated to obtain a more accurate measure of active biomass in activated sludge is the use of adenosine triphosphate. Biospherics Incorporated (43) developed methods for extracting ATP from sludge and

mixed liquor, and for determining ATP using the firefly bioluminescent procedure. Tests were conducted on laboratory units, pilot plant units and full scale municipal sewage treatment plants. Changes in ATP concentrations which indicated biological changes preceded conventional volatile sludge changes by as much as 24 hours. Successful results indicated some potential for ATP as a method of predicting biomass activity. In spite of improved accuracy and biomass prediction using more scientific techniques, volatile suspended solids will still be used for sometime due mainly to the test simplicity.

It is generally accepted that sludge accumulation does in fact occur in activated sludge processes. Washington and Symons (44) have studied volatile sludge accumulations in activated sludges and made some investigations into the composition of the accumulated solids. Kountz and Fourney are quoted by the authors as stating that 11.6 to 14.5 percent of the ultimate BOD fed to their reactors appeared as solid accumulation. Washington and Symons (44) claim that since the active mass of sludge is a constant for a given loading at equilibrium the accumulating solids must be biologically inactive and subject to metabolism by the microbial population. They also state that the accumulation occurs independently of the type of substrate or loading

schedule. It is thought that the inert material is the remains of cell capsular and external slime; a theory similarly expressed by McKinney (20). A conclusion of the experiments was that the composition of the inert volatile solids averaged as:

- (a) polysaccharide - 47 to 56%;
- (b) protein - 39 to 47%; and,
- (c) fats - 3 to 8%.

Hoover et al (26) studied the oxidation of milk solids by activated sludge. A detailed chemical analysis of the cell tissue yielded a chemical formula of $C_5H_7NO_2$. This formula has generally been accepted in the sanitary engineering field (2) (38).

3.2. Kinetic Models for Biological Treatment

Although the activated sludge process has been used for many years, most design criteria were largely empirical, developed from experience and operational data. An example of this is the design criteria published by Ten State Standards (15). A reasonable amount of success has been achieved using this design base. As the process developed, it became increasingly clear that more scientific design criteria needed to be developed using the fundamentals of biological growth.

3.2.1. Reactor Types

The behaviour of biological populations in waste treatment depends upon many factors; one of which is the hydraulic characteristics of the reactor vessel. The four principal types of reactors used in biological waste treatment are classified according to their hydraulic characteristics as batch, plug flow, complete mix, and arbitrary flow (2).

Batch flows are characterized by the fact that the flow entering or leaving the reactor is not continuous. An example of batch flow would be a conventional lagoon, a single stage anaerobic digestion unit, or a BOD incubation bottle. Arbitrary flow is a general flow description with no definite flow pattern and may be considered to be somewhere between plug flow and complete mix. The complete mix and plug flow systems are most common in modern waste treatment.

Complete mix systems are favoured for industrial and small domestic waste applications (45). Advantages of the complete-mix system have been described by several investigators including McKinney (46). Basically the advantages of a complete-mix system are: (45)

- (a) maximum equalization of oxygen uptake rate;
- (b) maximum dampening of shock loads;
- (c) maximum neutralization of CO_2 produced by respiration;

(d) reduction in the toxicity of a toxic material when the toxic material is biodegradeable and is present in low concentrations; and,

(e) provision of relatively constant environmental conditions for the biological mass.

Complete mixing occurs when the particles entering the tank are immediately dispersed throughout the tank. The hydraulics of complete mixing can be tested by the injection of a tracer into a continuous flow unit. The time-concentration curve would appear as shown in Figure 3.

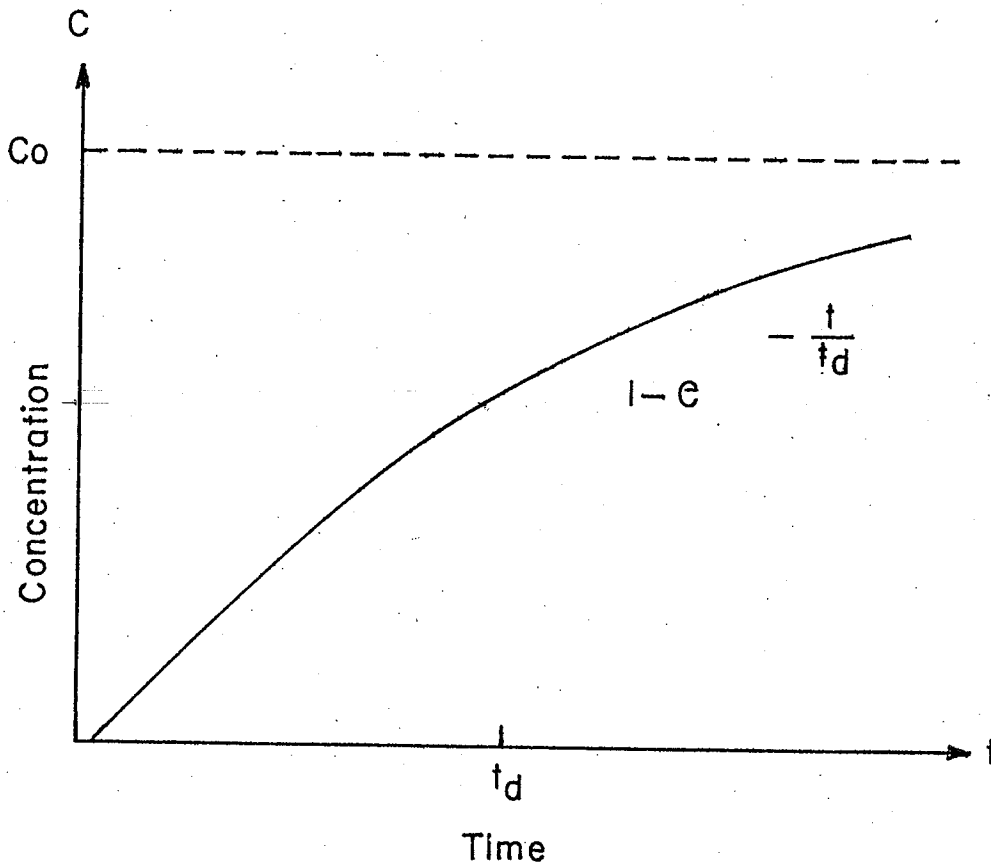


Fig. 3. Continuous Tracer Input for Complete-Mix System (after Metcalf and Eddy (2)).

This phenomenon can be described as follows:

(Rate of change in amount of tracer in reactor) = (rate of tracer inflow to reactor) - (rate of tracer outflow from reactor)

or mathematically as

$$\frac{dC(V)}{dt} = QC_0 - QC, \quad \dots (1)$$

where

C = effluent concentration of tracer at any time t;

V = volume of reactor;

Q = flow rate; and,

C₀ = influent concentration of tracer.

Rewriting and integrating equation (1) we find

$$\frac{dC}{dt} = \frac{Q}{V}(C_0 - C) \text{ and}$$

$$\int_0^C \frac{dC}{C_0 - C} = \frac{Q}{V} \int_0^t dt, \text{ that gives}$$

$$C = C_0(1 - e^{-t/(V/Q)}) \text{ or}$$

$$= C_0(1 - e^{-t/t_d}) \text{ where } t_d = \frac{V}{Q}$$

The hydraulics of the complete-mix system only will be presented since it is a characteristic of extended aeration, the activated sludge modification used in this investigation.

3.2.2. Basic Kinetic Model Equations

Investigators in the past have tried many different ways of describing or predicting the behaviour of biological waste treatment. This fact has caused some concern in the sanitary engineering field in that no standard approach has been taken so that results can be significantly cross referenced (28).

The development of kinetic growth equations is dependent upon the following factors (47):

- (a) growth rate;
- (b) a relationship between an essential nutrient and growth rate; and,
- (c) growth yield applied in conjunction with material balances.

One of the first researchers to investigate the use of kinetic models for continuous cultures was Monod. His works were researched by Gates and Marlar (48). They have indicated equations (1) and (2) below as the basic relationships proposed by Monod to define the interaction between nutrient utilization and bacterial growth and the dependency of the growth rate constant on the concentration of the growth controlling nutrient respectively. These equations are

$$\frac{dX^n}{d\theta} = \frac{1}{Y^o} \frac{dX^o}{d\theta}; \text{ and,} \quad \dots (1)$$

$$k = \frac{k^m X^n}{K + X^n} \quad \dots (2)$$

where X = concentration (mg/l); the superscript n and o refer to nutrient and organism concentration respectively.

Y^o = organism yield ($\frac{\text{mg organism}}{\text{mg nutrient removed}}$);

k = growth rate constant (time^{-1});

k^m = maximum growth rate constant (time^{-1});

θ = time; and,

K = the saturation or nutrient constant
(mg nutrient/l).

Although Monod applied his models to pure laboratory cultures, later researchers have applied his basic fundamentals to many models in sanitary engineering (47) (40) (46) (38) (49) (50) (51) (52). According to Schulze (53)

"...any mathematical model of the activated sludge process must contain formulations defining the kinetics of bacterial growth and specifically the relationship between growth rate and substrate concentration."

The relationship between growth rate and substrate utilization can be written as two basic equations (16). The first equation is an empirical relationship between net rate

of growth of microorganisms and rate of substrate utilization.

$$\frac{dX}{dt} = Y \left(\frac{dF}{dt} \right) - bX, \quad \dots (1)$$

where $\frac{dX}{dt}$ = net growth rate of microorganisms per unit volume of reactor;

Y = growth yield coefficient, mass/mass;

$\frac{dF}{dt}$ = rate of microbial substrate utilization per unit volume, mass/volume-time;

b = microorganism decay coefficient (time⁻¹); and,

X = microbial mass concentration (mass/volume).

The second basic equation relates the rate of substrate utilization to both the concentration of microorganisms in the reactor and to the concentration of substrate surrounding the organism. This can be described by a slightly modified version of the Monod equation, as

$$\frac{dF}{dt} = \frac{kSX}{K_s + S}, \quad \dots (2)$$

where k = maximum rate of substrate utilization per unit weight of microorganisms (time⁻¹);

S = concentration of substrate surrounding the microorganisms, mass/volume; and,

K_s = half velocity coefficient, equal to the substrate concentration when $\frac{dF}{dt} = \frac{1}{2}k$, mass/volume.

Equation (2) can be applied to a situation where S is very high ($S \gg K_s$) or S is very low ($S \ll K_s$). In most activated sludge applications, and especially extended aeration, the substrate concentration is generally low.

Equation (2) can then be written:

$$\frac{dF}{dt} = k^1 SX \quad \dots (3)$$

where $k^1 = \frac{k}{K_s}$.

Equation (3) is a first order discontinuous function and can be illustrated as follows (16) in Figure 4.

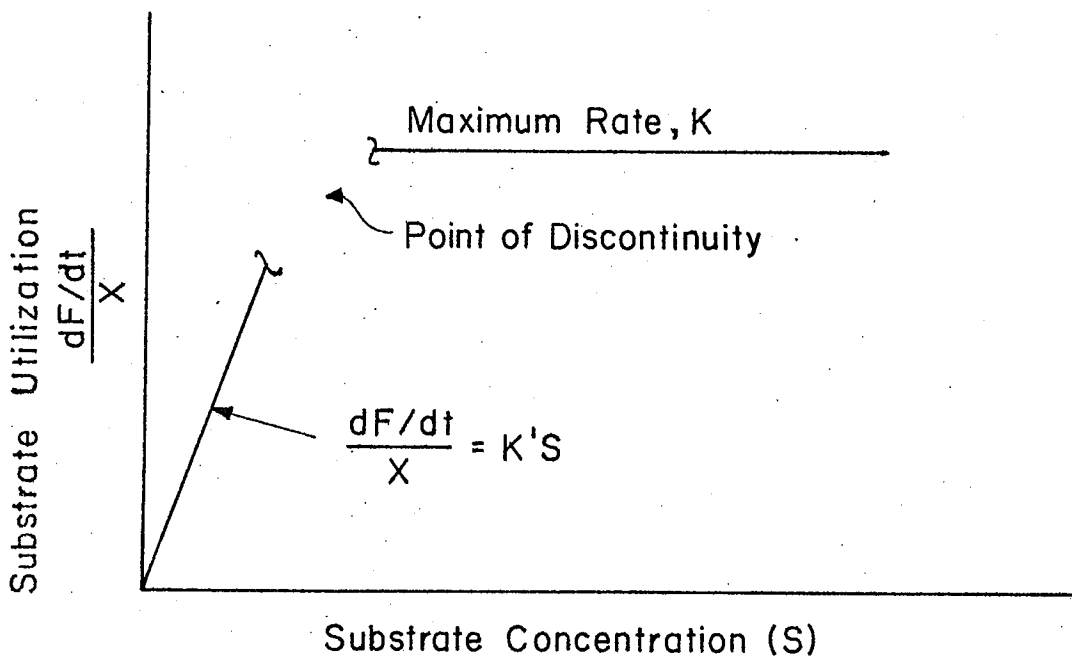


Fig. 4. Schematic Representation of Discontinuous Substrate Utilization Equation (after Lawrence and McCarty (16)).

Another basic approach that is similar to the Monod formulations is the Michaelis-Menten equation based on enzyme behaviour (2) (50). The bacterial cell is considered an enzyme molecule reacting with the substrate, S.

The specific growth rate equation can be written as:

$$k_1 = k_m \left(\frac{S}{S_n + S} \right), \quad \dots (4)$$

where k_1 = specific growth rate;

k_m = maximum growth rate;

S = substrate concentration; and,

S_n = substrate concentration at which $k_1 = \frac{k_m}{2}$.

The development of the mathematical models using either equations (2) or (4) is the same using either fundamental approach.

Reynolds and Yang (45) have summarized the fundamental equations necessary for model development as:

$$(a) \frac{dX}{dt} = \mu X;$$

$$(b) \frac{dX}{dt} = Y \frac{dS}{dt};$$

$$(c) \mu = \frac{\mu_{max} X}{K_s + S}; \text{ and,}$$

$$(d) \frac{dX}{dt} = k_e X.$$

where X = cell concentration;

Y = yield coefficient;

μ = growth rate;

S = substrate concentration;

K_s = half velocity coefficient; and,

k_e = endogenous respiration rate.

To evaluate the system it is necessary to write material balance equations of the system as:

(a) material balance of cells which may be written as;

Change in Reactor	=	Increase due to Recycled flow	+	Increase due to Growth
	-	Decrease due to endogenous respiration	-	Decrease due to effluent from reactor, and

(b) material balance of substrate which may be written as:

Change in Reactor	=	Increase due to Influent Flow	+	Increase due to Recycled Flow
	-	Decrease due to Growth	-	Decrease due to Effluent from Reactor.

3.2.3. Mathematical Model Development

To develop a mathematical model that may be applied to a particular system the basic assumptions set forth in

Section 3.2.2. and the hydraulic characteristics describe in Section 3.2.1. must be brought together and analyzed. It was mentioned earlier that the complete-mix activated sludge system was the most desirable for small systems such as the extended aeration process. Complete-mix systems can exist either as single stage without cellular recycle or single stage with cellular recycle. In the analyses of these systems a critical assumption is made in that the basic pure culture kinetic equations are applied to complex heterogeneous systems such as activated sludge.

It is also necessary to be aware of the significant assumption that microbial growth is limited by the absence of only one or one category of substances with all other requirements being adequate.

The development of mathematical models for complete mix systems will be limited to no recycle and cellular recycle. The former is given because of its simplicity demonstrating the analytical approach, whereas the latter most closely resembles the extended aeration process.

3.2.3.1. Complete Mix - No Recycle System (16)

The complete mix - no recycle system is most closely illustrated by a conventional anaerobic system, an aerated

lagoon, an aerobic lagoon, and dispersed growth modifications of the activated sludge process. A schematic diagram illustrating this is shown below as Figure 5.

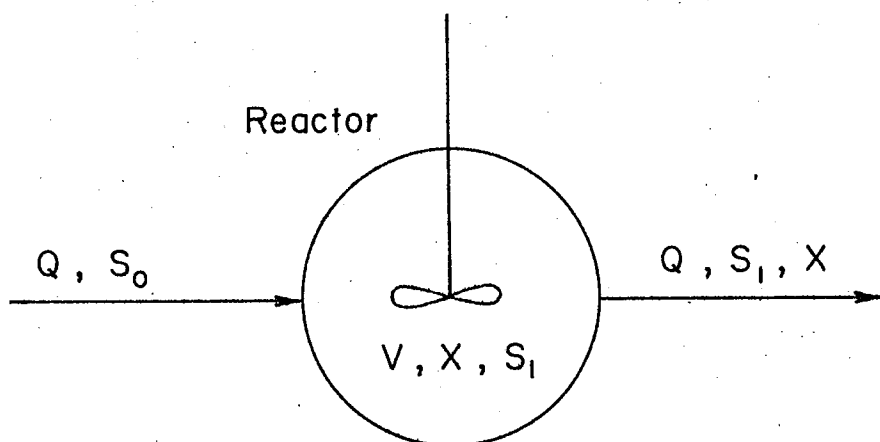


Fig. 5. Complete-Mix System With No Solids Recycle (after Lawrence and McCarty (16)).

The liquid wastes are assumed to flow into the reactor at a constant rate and to be instantaneously and homogeneously mixed with the reactor contents. The reactor volume is held constant by maintaining an outflow rate equal to the inflow

rate. The influent contains no active microorganisms and the reactor and effluent microorganism concentrations are equal.

A materials balance for the net rate of change of microbial mass in the system shown in Figure 5 may be written as

net rate of change of microbial mass = growth rate - washout rate, or

$$V \frac{dX}{dt} = Y \frac{dF}{dt} - bXV - QX \quad \dots (1)$$

where $\frac{dX}{dt}$ = net rate of change of microbial mass in the reactor, mass/volume - time .

for steady state conditions $\frac{dX}{dt} = 0$.

Thus equation (1) may be written as

$$\frac{Q}{V} = \frac{Y(dF/dt)}{X} - b \quad \dots (2)$$

and since $\frac{V}{Q} = \theta$, the hydraulic retention time

$$\frac{1}{\theta} = YU - b$$

where U = food to microorganism ratio.

The mean cell residence time is defined as (54) (55) (40):

$$\theta_c = \frac{X_T}{(\Delta X / \Delta t)_T}$$

where X_T = total active microbial mass in treatment system; and,

$\frac{(\Delta X)}{\Delta t}_T$ = total quantity of active microbial mass withdrawn daily including those intentionally wasted as well as those lost in the effluent.

For the complete-mix system with no solids recycle the mean cell residence time can be written as,

$$\theta_C = \frac{XV}{XQ} = \frac{V}{Q} = \theta;$$

i.e. the mean cell residence time is equal to the hydraulic retention time.

Substituting the modified Monod equation

$\frac{dF}{dt} = \frac{kSX}{K_s + S}$ into equation (2) results in

$$\frac{1}{\theta_C} = \frac{YkS_1}{K_s + S_1} - b \quad \dots (3)$$

and rearranging equation (3) to solve for the effluent concentration S_1 ,

$$S_1 = \frac{K_s(1+b(\theta_C))}{\theta_C(Yk-b)-1} \quad \dots (4)$$



It becomes apparent from equation (4) that the effluent concentration, S_1 , and hence efficiency, is directly related to the mean cell residence time, θ_c and the kinetic growth parameters K_s , k , Y , and b .

In a given biological reactor the kinetic constants are assumed to be fixed for a given operating condition and the controlling parameter becomes θ_c , i.e. the longer θ_c , the better the treatment efficiency.

It would be of value from practical considerations to have some knowledge of the minimum retention time. A minimum retention time determines the time at which cellular growth is not given the time to develop, with the result that the effluent concentration, S_1 equals the influent concentration, S_0 (2).

Rewriting equation (3) for minimum retention time, θ_c^M and letting $S_0 = S_1$, then

$$\frac{1}{\theta_c^M} = Y \frac{kS_0}{K_s + S_0} - b; \quad \dots (4)$$

and since $S_0 \gg K_s$

$$\frac{1}{\theta_c^M} = Yk - b. \quad \dots (5)$$

A basic assumption in the development of equation (5) is that the effluent concentration equals the influent concentration. This is not practical in fact and so the design θ_c varies from 2 to 20 times θ_c^M .

3.2.3.2. Complete Mix with Cellular Recycle (2)

Application of biological waste treatment in sanitary engineering frequently involves some form of cellular recycle. To evaluate the biokinetics of this type of system several assumptions in addition to the no solids recycle case are made so that:

- (a) waste stabilization occurs in the reactor only; and,
- (b) the volume for the calculation of θ_c is the reactor volume only.

These assumptions are not universally accepted and some investigators believe that the solids or at least a fraction of the solids in the secondary clarifier should be used in the calculation of θ_c (54). The assumptions given above result in a conservative design. A schematic of a complete-mix reactor with cellular recycle is shown below as Figure 6.

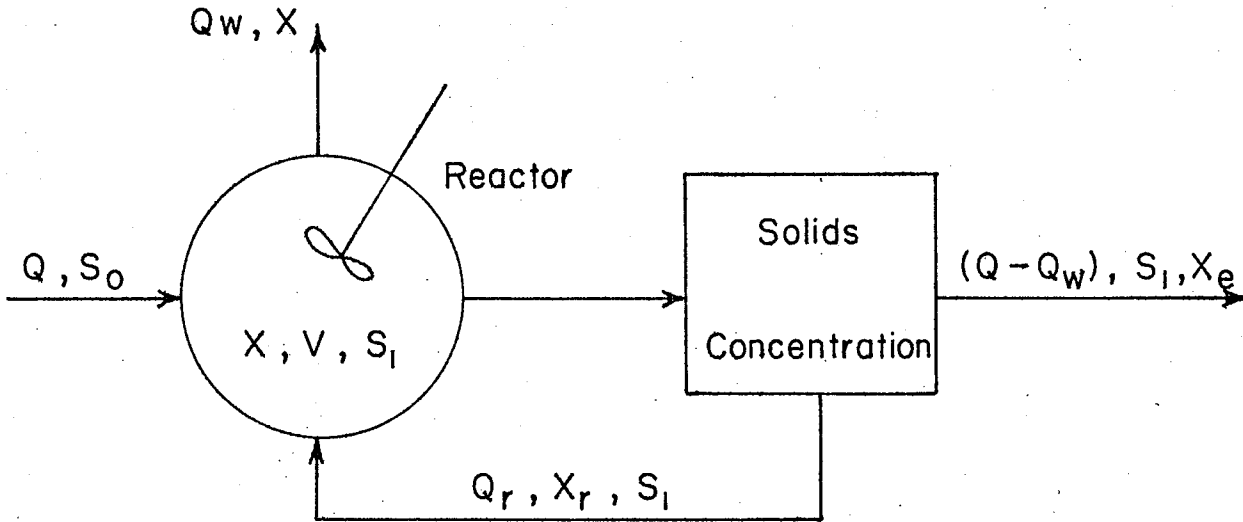


Fig. 6. Complete-Mix System With Solids Recycle (after Metcalf and Eddy (2)).

The system hydraulic retention time, $\theta_s = \frac{Vs}{Q}$
 where Vs = reactor plus settling chamber volume; and,
 Q = inflow rate.

The mean hydraulic retention time, $\theta = \frac{V}{Q}$
 where V = reactor volume.

From the definition of $\theta_c = \frac{X}{(\Delta X/\Delta t)}$,

$$\theta_c = \frac{VX}{Q_w X + (Q - Q_w) X_e}$$

where Q_w = flow rate of liquid containing cells wasted from the reactor; and,

X_e = microorganism concentration in the effluent from the settling unit.

In a properly operating settling tank X_e is small and so equation (1) can be simplified to

$$\theta_c \approx \frac{V}{Q_w} \quad \dots (2)$$

In most biological treatment processes the cell wastage is accomplished by drawing off the sludge recycle line. The mean cell residence time for this situation can be written as follows:

$$\theta_c = \frac{VX}{Q_w^1 X_r + (Q - Q_w^1) X_e} \quad \dots (3)$$

where X_r = microorganism concentration in return sludge time; and,
 Q_w^1 = cell wastage rate from recycle line.

Assume that X_e approaches zero for a well operating clarifier, then

$$\theta_c \approx \frac{VX}{Q_w^1 X_r} \quad \dots (4)$$

By using a system mass balance approach as in section 3.1.5.4. and considering a finite time basis

$$\frac{1}{\theta_c} = \frac{YkS_1 - b}{K_s + S_1} ; \quad \dots (5)$$

that is, there is a direct relationship between θ_c and Y, b, K_s and k.

A significant point is the difference between the case of no cellular recycle and the case of cellular recycle. The major difference is that with no cellular recycle, θ_c is directly related to the hydraulic retention time, whereas with cellular recycle, θ_c is theoretically independent of the hydraulic retention time. This adds considerably more flexibility to the recycle process. The mean cell residence time is relatively easy to control and so is generally used to control the process. The use of θ_c is based upon the fact that to control the growth rate of microorganisms and hence the degree of waste stabilization, a specified percentage of the cell mass in the system must be wasted each day.

Although theoretically θ_c is independent of θ , operating plants are in fact somewhat dependent on hydraulic retention time due to:

- (a) the oxygen transfer rate in the reactor;
- (b) the proper operation of the settling unit; and
- (c) the settling characteristics of the suspended solids in the mixed liquor.

3.2.4. Kinetic Models Applied to Extended Aeration

Middlebrooks and Garland (47) have developed a mathematical model for an extended aeration plant utilizing the basic biological principles described in previous sections. The complete-mix unit of the cellular recycle is illustrated below as Figure 7.

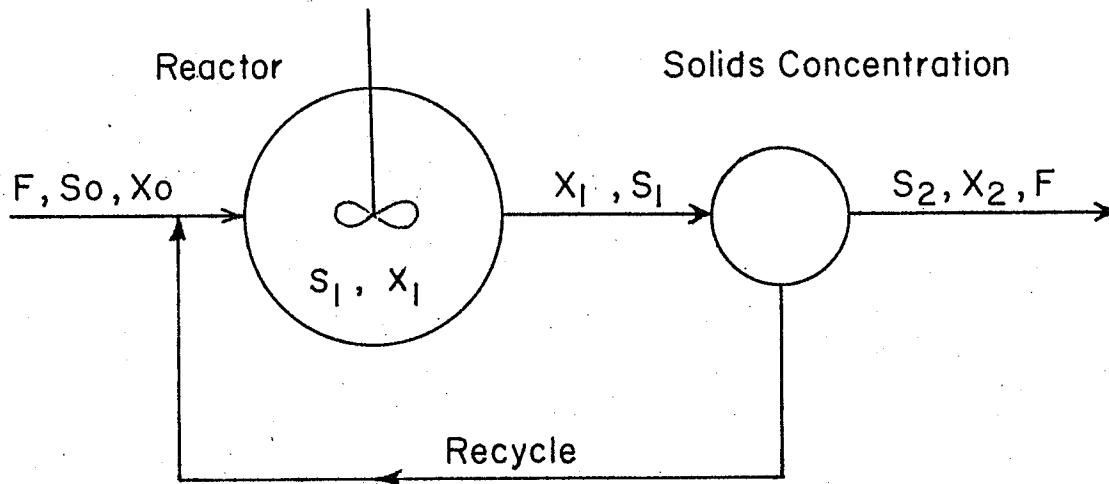


Fig. 7. Typical Extended Aeration Flow Schematic (after Middlebrooks and Garland (47)).

A materials balance for the organisms in the system shown in Figure 7 may be written:

$$\begin{array}{l} \text{Organism} \\ \text{change in} \\ \text{reactor} \end{array} = \begin{array}{l} \text{Organisms} \\ \text{in influent} \end{array} + \begin{array}{l} \text{Growth of} \\ \text{organisms} \end{array} \\ - \begin{array}{l} \text{Loss of organisms} \\ \text{in effluent} \end{array} - \begin{array}{l} \text{Loss due} \\ \text{to decay,} \end{array}$$

or in symbolic form,

$$V(dx_1)_{\text{net}} = X_0 F dt + (dx_1)_g V - k_d X_1 V dt - X_2 F dt; \quad \dots (11)$$

where V = reactor volume;

$(dx_1)_{\text{net}}$ = net change in organism concentration in reactor;

X_0 = organism concentration in the influent;

F = flow rate through the reactor;

$(dx_1)_g = \mu X_1 dt$, where μ = growth rate constant;

k_d = decay rate constant;

X_1 = organism concentration in the reactor;

X_2 = organism concentration in the effluent; and,

t = time.

The hydraulic retention time, θ can be expressed as,

$$\theta = \frac{V}{F}.$$

The concentration of organisms of the influent is insignificant when compared with the microbial concentration in the reactor. Neglecting the organisms in the influent and

dividing equation (1) by V and dt yields

$$\frac{(dX_1)}{dt}_{net} = \mu X_1 - k_d X_1 - \frac{X_2}{\theta} \quad \dots (2)$$

At steady state conditions

$$\frac{(dX_1)}{dt}_{net} = 0$$

Let $\frac{X_1}{X_2} = b^1$ where b^1 is the concentration factor and

represents the ratio of the concentration of organisms in the reactor to the concentration of organisms in the effluent. The ratio b^1 is always greater than 1.

Equation (2) becomes

$$\mu = k_d + \frac{1}{b^1 \theta} \quad \dots (3)$$

Similarly a materials balance may be done on the system substrate.

Substrate change in reactor = Substrate in Influent - Consumption by organisms - Loss of substrate in effluent

which can be written mathematically as

$$V(dS_1)_{net} = S_0 F dt - V(dS_1)_g - S_2 F dt \quad \dots (4)$$

where $(dS_1)_{net}$ = net change in substrate concentration in the reactor;

S_0 = substrate concentration in the influent;

S_1 = substrate concentration in the reactor;

S_2 = substrate concentration in the effluent; and

$(dS_1)_g = \frac{dX}{Y}$ i.e. the change in substrate concentration due to growth;

Since $dX = -\mu X dt$ then

$$(dS_1)_g = \frac{-\mu X_1 dt}{Y} \quad \dots (5)$$

Dividing equation (4) by V and dt and substituting equation (5) gives

$$\frac{(dS_1)_{net}}{dt} = \frac{S_0}{\theta} - \frac{\mu X_1}{Y} - \frac{S_2}{\theta} \quad \dots (6)$$

It is assumed that the substrate in the effluent from the system is the same as that in the substrate in the reactor, i.e. $S_1 = S_2$

At steady state $\frac{(dS_1)_{net}}{dt} = 0$ and

substituting equation (3) into (6)

$$X_1 = \frac{b^1 Y (S_0 - S_1)}{1 + b^1 \theta k_d} \quad \dots (7)$$

From $\mu = \frac{1}{b^1 \theta} + k_d$ and $\mu = \frac{\mu_{\max} S_1}{K_s + S_1}$, we get

$$\frac{\mu_{\max} S_1}{K_s + S_1} = \frac{1 + b^1 \theta k_d}{b^1 \theta} \quad \dots (8)$$

To determine the growth kinetics from equations (7) and (8) it is desirable to rewrite them in a straight line form, i.e. $y = ax + b$ where $a = \text{slope}$, and
 $b = \text{intercept}$.

Rearranging equation (7) into a straight line form

$$X_1 (1 + b^1 \theta k_d) = b^1 Y (S_0 - S_1);$$

$$\frac{1 + b^1 \theta k_d}{Y} = \frac{b^1 (S_0 - S_1)}{X_1};$$

$$\frac{(k_d) b^1 \theta + 1}{Y} = \frac{b^1 (S_0 - S_1)}{X_1} \quad \dots (9)$$

where slope = $\frac{k_d}{Y}$; and,

intercept = $\frac{1}{Y}$.

Rearranging equation (8) into a straight line plot

$$\mu_{\max} S_1 b^1 \theta = (1 + b^1 \theta k_d) (K_s + S);$$

$$\frac{b^1 \theta}{1 + b^1 \theta k_d} = \frac{K_s + S_1}{\mu_{\max} S_1};$$

$$\frac{b^1 \theta}{1 + b^1 \theta k_d} = \frac{K_s}{\mu_{\max} S_1} + \frac{S_1}{\mu_{\max} S_1};$$

$$\frac{b^1 \theta}{1 + b^1 \theta k_d} = \frac{(K_s)}{\mu_{\max}} \left(\frac{1}{S_1}\right) + \frac{1}{\mu_{\max}}; \quad \dots (10)$$

where slope = $\frac{K_s}{\mu_{\max}}$; and,

$$\text{intercept} = \frac{1}{\mu_{\max}}.$$

The development of this mathematical model required the use of microbiological growth fundamentals and is specific for extended aeration where no intentional sludge wasting is practiced. The assumptions made in its development have not allowed for conditions such as varying yield, oxygen levels, turbulence and mixing. It provides a basis on which the performance at an extended aeration plant may be scientifically analyzed.

3.3. Kinetic Model Constants

The previous sections of this literature review has emphasized the importance of the fundamentals in establishing mathematical relationships or models which help to describe and predict the performance of biological treatment processes.

As has been pointed out, the successful use of the process equations is mainly dependent upon the application of constants which are part of the growth relationships. It is important to be aware of the environmental conditions, e.g. temperature substrate concentration, mixing, turbulence, etc., which may change these "constants" and thus have an effect on the efficiency of these equations.

3.3.1. Growth and Substrate Utilization Parameters

The growth parameters for the mathematical models used to describe biological waste treatment are usually considered to be Y , the yield constant and k_d the endogenous respiration rate. As discussed in section 3.2.4. the substrate utilization parameters are k , the substrate utilization rate and K_s the half velocity coefficient. The values for these parameters must be available if the kinetic equations are to be used effectively. Considerable effort has gone into investigation of the growth parameters Y and k_d . For a comprehensive review of the literature on yield coefficients up to 1970 the reader is referred to Friedman (56). Yield coefficients are not to be considered entirely alone since it is thought that the endogenous decay rate is most significant in estimating net sludge production. Ball and Humenick (57) conducted tests on sludge growth constants with air and pure oxygen. The

yield coefficient, Y was found to be 1.0 with pure oxygen and 1.38 with air whereas the endogenous respiration rates were 0.27 and 0.17 per day respectively. According to Middlebrooks and Garland (47) a yield constant greater than one is impossible if the growth rate and yield are dependent on the influent BOD. This high value is probably due to the use of volatile suspended solids as an indicator of cell mass. If the mean cell residence time were low, then it is possible that the measured volatile solids would simply be unmetabolized influent and an unusually high yield would be the result.

Friedman and Schroeder (8) summarized yield coefficient research and concluded that recent design formulations suggest the use of $Y = 0.65$ and 0.50 . Walker (55) discussed the use of growth constants for domestic wastes. He suggested a range of yield values from 0.5 to 0.6 and decay rates of 0.04 to 0.05 as typical averages.

Jenkins (40) used chemical oxygen demand as a measure of the substrate and reported a yield constant range as 0.3 to 0.9 lb. vss produced per lb COD removed at a mean cell residence time of 5 to 15 days. These studies resulted in a decay rate of 0.04.

Eckenfelder (41) concluded that the yield coefficient for domestic wastes can be estimated at 0.4 lb vss per lb COD or 0.55 based on the 5 day BOD. He considered a decay

coefficient of 0.2 to 0.26/day @20°C as being typical for a biological sludge. It was pointed out that when volatile suspended solids are present in the sludge the growth coefficients, Y and k_d will show increased values for yield and decreased values for decay due to the presence of non-biological volatile solids in the sludge mixture.

McKinney (20) stated that with the information to date (1962),

"...the maximum quantity of protoplasm formed in activated sludge systems or in any bacterial system equals 0.47 times the ultimate oxygen demand of the organic matter metabolized."

He claims that this applies to pure cultures as well as mixed cultures. Endogenous respiration is thought to be temperature dependent. Ammonia, a metabolic by-product of endogenous respiration, creates an oxygen demand when nitrifying bacteria are present but may not necessarily result in a significant decrease in biological mass. Decay rate figures are not quoted.

McCarty and Brodersen (58) investigated the theory of extended aeration activated sludge. They considered the yield constant as being comprised of a degradable and a non-degradable fraction. The non-degradable fraction, such as lignin or cellulose, results in an inevitable sludge build-up in extended aeration. They estimate the yield coefficient

for domestic waste to be 0.65 of which 0.53 is degradable and 0.12 is non-degradable.

Eckenfelder (59) determined the yield constant on soluble pharmaceutical wastes. On a 5-day BOD removal basis with a deoxygenation constant of 0.1, Y was found to be 0.57. He quotes Porges et al as giving activated sludge decay rate of 0.3/day at 25°C treating dairy wastes and Wuhrman stating that the decay rate decreases with increasing sludge concentration because of the accumulation of non-biodegradable sludge. Wuhrman is quoted as stating that the average decay rate is 0.24/day at 20°C but may decrease to 0.05/day with a wastewater high in suspended solids such as a pulp and paper waste.

Dryden et al (60) investigated growth parameters on pharmaceutical wastes consisting of antibiotics and vitamins using a 5 litre activated sludge laboratory unit. They found the yield coefficient equal to 0.77 and the decay rate equal to 0.2/day. Industrial wastes were treated via a laboratory complete-mix activated sludge process by Stack and Conway (61). At a 15 hour retention period a net sludge production of 0.4 lb per lb of COD removed was determined.

Kountz and Forney (23) performed tests on total oxidation activated sludge systems. They claimed that when using dry skim milk solids as a source of organic matter 58% of the influent ultimate oxygen demand appeared as new sludge

in a continuous flow system.

The actual endogenous loss was found to be 2% per day of the total weight of activated sludge in the system. Hoover and Porges (26) conducted research on the biochemical oxidation of milk solids by activated sludge and found that on a weight basis the yield of cell material was about 52% of the protein and carbohydrate in the milk solids, for cells containing about 9% ash.

Garrett (62) performed tests on an actual sewage treatment plant in Houston, Texas, for a period of 17 months. With a variation in mean cell residence time of 1 to 2.5 days, plotted data resulted in a yield coefficient of 1.1 based upon volatile suspended matter and 5 day BOD removed.

Reynolds and Yang (45) developed mathematical models for the complete mixed activated sludge process. Using continuous flow laboratory reactors they found a yield constant, Y equal to $0.445 \frac{\text{mg MLSS}}{\text{mg COD removed}}$ and $0.39 \frac{\text{mg MLVSS}}{\text{mg COD removed}}$.

The endogenous respiration rate was found to be 0.16 per day. They also calculated substrate utilization rates to be $\mu_{\text{max}} = 0.80/\text{hr}$ and $K_s = 345 \text{ mg/l}$.

Middlebrooks and Garrland (47) calculated the kinetic constants for actual operating extended aeration plants. These lightly loaded, long retention, complete-mix systems were found to have substrate utilization rates of $\mu_{\text{max}} = 0.00159/\text{hr}$, and $K_s = 12.4 \text{ mg/l}$.

Ghosh et al (63) developed basic kinetic growth relationships based upon enzymatic reactions. They determined the kinetic constants of high retention time cultures with substrate consisting of glucose and galactose. Their results are as shown in Table 1 below.

Substrate	k_d /hr	Y	μ_{max} /hr	K_s (mg/l)
glucose	0.116	.841	.728	1.7
galactose	0.027	.586	.387	3.9

TABLE 1. Kinetic Constants of Glucose and Galactose Grown Cultures (after Ghosh et al).

Servizzi and Bogan (30) summarized the yield constant values as ranging from 0.35 to 1.0. Their work on various organic substrates resulted in an average yield of 0.37 g cells/g COD.

Genetelli and Heukelekian (64) studied the effects of loading and substrate composition on sludge yield of activated sludge. They found that sludge yields were primarily dependent on substrate composition and their results varied from a low yield constant, Y equal to 0.313 for egg albumin to a high of 0.698 for glucose.

Stewart and Ludwig (65) considered a range of loadings in activated sludge and estimated an average yield of 0.56 based on volatile solids and 5 day BOD with a decay constant of about 0.05/day. They concluded that at low loadings as encountered in extended aeration plants the decay constant may be as low as 0.01 per day.

3.3.2. Temperature Coefficient Considerations

Most chemical reactions are temperature dependent. In most instances it is rather difficult to pinpoint exactly the effect temperature has on a given reaction. A commonly used rule of thumb is that the rate doubles for each rise in temperature at 10°C. This can be expressed more precisely by the van't Hoff-Arrhenius equation (5) as

$$\frac{d(\ln k)}{dt} = \frac{E}{RT^2} \quad \dots (1)$$

where k = specific reaction rate constant;

T = temperature (°K);

R = gas constant (1.99 cal per °C); and,

E = activation energy (gram-calories).

Integrating equation (1) between the limits of T_1 and T_2 gives

$$\ln\left(\frac{k_2}{k_1}\right) = \frac{E(T_2 - T_1)}{RT_1 T_2} .$$

Thus the specific rate constant at any temperature can be calculated from a knowledge of E and the rate constant at some one temperature within the range of pertinent activation energies.

Fair, Geyer and Okun (5) give two other methods of expressing the temperature dependency on reaction rates as follows:

$$(a) \quad \frac{k_2}{k_1} = \theta^{T_2 - T_1}$$

where $\theta = \frac{k_2}{k_1}$ for $T_2 - T_1 = 1$; and,

$$(b) \quad Q_{10} = \frac{k_2}{k_1} = \theta^{10} \text{ for } T_2 - T_1 = 10.$$

The temperatures for these relationships may be $^{\circ}\text{C}$ since differences only are used.

Friedman and Schroeder (8) and Friedman (56) have studied in great detail past investigations into temperature effects of biological waste treatment. The most common mathematical form of expressing temperature effects on

reaction rates is $\frac{k_1}{k_2} = \theta^{T_1 - T_2}$ where θ is often called the

temperature coefficient and is assumed to be constant over a given temperature range.

The use of this relationship is not as simple as the equation appears. The magnitude of the temperature effect depends to a large extent on the nature of the process. According to Eckenfelder (59) the oxygen utilization rate which reflects the energy transfer of the process has a first order dependency with temperature.

A main assumption that is made in the use of this basic equation is that, θ is a constant. Fair, Geyer, and Okun (5) state that the use of θ is limited because of its variation with temperature. Eckenfelder (59) has explained the variation of the temperature coefficient, θ by stating that aerobic conditions within the cells depends on diffusion of oxygen from the surface saturated liquid layers. At higher temperatures, oxygen is utilized rapidly and oxygen only penetrates a short distance into the floc. At lower temperatures, deeper penetration is possible and more microorganisms are available for waste assimilation, although at a lower rate. As the loading increases, the utilization rate is increased, penetration is reduced and the temperature coefficient or effect increases. By this reasoning the

temperature effects in a lagoon would be different from an activated sludge unit. The dispersed nature of the biological growth in an aerated lagoon would be expected to yield a temperature coefficient roughly equivalent to the effect on oxygen utilization itself. Conversely, the activated sludge process, operating at a relatively high microbial concentration, is relatively insensitive to temperature fluctuations.

Zanoni (6) listed some of the more important factors as affecting the temperature coefficient, θ as:

- (a) temperature range;
- (b) number of test temperatures;
- (c) type of substrate;
- (d) method of chemical analyses; and,
- (e) procedure for evaluation of rate constants.

The conclusion of his work was that the temperature effect on the 5 day BOD of secondary treatment plant effluents was a temperature coefficient of 1.077 at temperatures from 10°C to 20°C and 1.048 at temperatures from 20°C to 30°C.

Carpenter et al (7) studied operating aerated lagoon treatment of pulp and paper wastes at a temperature range of 2°C to 30°C. The temperature coefficient of the raw and treated wastes deoxygenation rate was found to be 1.016. The 5 day BOD and ultimate BOD temperature coefficient were 1.035 and 1.031 respectively. The overall BOD removal rate for the

aerated lagoon was found to have a θ equal 1.031 to 1.046.

Krenkel et al (66) investigated the effects of temperature on reaction rates of deoxygenation. Their findings indicated that temperature had a significant effect on deoxygenation constants of impounded waters, and that

"...a multitude of different organisms are active in waste assimilation, all with their own characteristics and temperature tolerances. The distribution of organisms may change drastically with shifts in temperature and types of waste each species having a different rate of metabolism. Because the composite metabolism includes many coupled reactions, each with its own characteristics, the composite rate-limiting steps may shift with temperature changes, thus precluding the interpretation of the process as a single reaction."

They go on further to say that because of the variability and complexity of the reactions a semi-empirical reaction at best can be used to describe the temperature effects. Gotaas is quoted as saying that the θ values vary with temperature as follows:

(a) $\theta = 1.109$ at 5°C to 15°C ;

(b) $\theta = 1.042$ at 15°C to 30°C ; and,

(c) $\theta = 0.967$ at 30°C to 40°C .

The authors quote Belehvadek as stating that the deoxygenation rate varies with temperature raised to some power approximately equal to 0.932 in the temperature range of 5°C to 30°C, whereas Stoltenberg and Sobel (67) hypothesized an empirical quadratic relationship between the deoxygenation rate and temperature and found that it was statistically significant when compared with a linear relationship. The authors sum up by stating that

"...the rate of oxidation will vary with the type of waste and the type of biological population which grows in response and oxidizes the waste. Under differing conditions, different biological population will vary in their temperature susceptibility so that temperature coefficients and temperatures at maximum reaction rates will also vary."

Novak (68) decided that the two most important factors in determining θ , were the substrate concentration and the food to microorganism ratio. He concluded that θ is equal to 1.00 at low substrate levels and equal to 1.02 to 1.18 at high substrate levels, the range of values being dependent upon the substrate being metabolized.

Benedict (69) discovered that θ was equal to 1.078 at 4°C and was independent of loading when the loading rate did not exceed 0.53 lbs BOD per day per lb mixed liquor suspended solids.

Research conducted by Pohl (70) indicated that the temperature coefficient value decreased as the temperature was lowered. Usually θ values are thought to increase as temperature is decreased. The explanation for this is the theory put forth by Ingraham who reported that psychrophilic bacteria have a lower energy of activation than mesophiles. Since it is thought that the energy of activation is related to θ , θ should be lower at lower temperatures if psychrophiles are present.

3.3.3. Effects of Temperature on Activated Sludge Performance

The kinetics of aerobic BOD removal is related to the physical characteristics of the waste. Suspended organic matter, for example, is removed by flocculation and enmeshment with the biological floc.

Colloidal material is thought to be adsorbed on the biological surfaces. According to Eckenfelder (59) levels of removal are a function of time and are related to the sludge growth. He states that

"...there is evidence that each of the constituents of the waste is removed at a linear rate, i.e. zero order, and that the total removal at any time is the sum of the individual components. At high concentrations when all the components are present, the overall rate is

zero order and the total removal for unit sludge solids is constant. As the more readily removable components are removed the overall rate will decrease..."

and this can be estimated by a first or second order equation. According to Eckenfelder (41) this process is temperature dependent, and the magnitude of the temperature effect depends to a large extent of the nature of the process. His work indicated that at activated sludge food to microorganism loading of less than 0.5 no temperature effects were noted at temperatures less than 95°F. Increasing the process loading increased the temperature effects. An explanation of this was given by claiming that at low loading the reduced respiration rate permits greater diffusion into the floc and hence a larger mass of organisms are active in colder weather. At high loads with dispersed growth all the organisms are equally affected by temperature and so decreased efficiency is noted at colder temperatures. A common example of this is a lagoon operating at low temperatures.

Carpenter et al (7) studied aerated lagoons treating pulp and paper wastes at temperatures ranging from 2°C to 30°C. Their findings showed that a temperature reduction from 20°C to 2°C resulted in a BOD removal efficiency drop of 74% to 56% at 2.5 day retention, from 82% to 70% at 5 days

and from 87% to 79% at 10 days. The temperature effects appeared to be minimized by prolonged aeration.

Keefer (71) studied 20 years of records of the Back River wastewater treatment plant serving the major portion of Baltimore, Maryland.

The results showed:

(a) BOD removal efficiencies of

(i) 85% at 12.2°C;

(ii) 90% at 18.9°C; and,

(iii) 91% at 23.4°C;

(b) Increase in suspended solids removal at higher sewage temperatures; and,

(c) Increased % removal of BOD as the BOD load per day increased.

Banerji (72) studied the removal kinetics of potato starch at 5°C, 20°C and 30°C using a batch flow activated sludge system. The starch removal followed zero order kinetics based upon COD. The effects of temperature were such that a temperature increase increased the starch removal.

Ludzack (73) studied the behaviour of a bench scale extended aeration plant at different temperatures and observed that:

(a) rapid temperature fluctuations usually resulted in temporary plant upset;

(b) the effluent quality was not greatly impaired with steady state operation at 5°C as compared to operation at 20°C;

(c) sludge accumulations became greater at lower temperatures; and,

(d) the sludge produced at lower temperatures was of a lower density and tended to become gelatinous with poor settling characteristics.

Earlier Ludzack et al (74) discovered that temperature appeared to have a significant effect on the variety and motility of the predator population, i.e. more rotifers were noted at 30°C than at 5°C. The predator acclimation to different test feeds was 5 times as long at 5°C as at 30°C. It was also observed that the filamentous bacteria Sphaerotilus showed a predominance at 5°C.

Schroepfer et al (75) investigated the temperature effects on trickling filter performance. The percent BOD removal efficiency was plotted versus temperature (50°F to 75°F). A straight line equation was developed using a least squares procedure.

Hunter et al (76) conducted batch tests on the activated sludge process at temperatures ranging from 4°C to 55°C. The BOD removal was 79.1% at 4°C and 92.3% at 20°C.

The effluent suspended solids removal was 81.5% at 4°C versus 91.8% at 20°C. Increasing the temperature increased the protozoan and rotifer activities. The lower temperatures appeared to favour increased filamentous growth as well as increased cellulose content.

Reynolds and Yang (45) developed mathematical models to describe the complete-mix activated sludge process. Their results indicated that

"...a variation in temperature in the mesophilic range does not appreciably affect the sludge yield coefficient, however, it is known to affect the endogenous respiration rate."

Vela (77) noted that the total oxidative ability of soil bacteria was greater than expected from classical microbiological theory. Tests showed that psychrophiles represented only 1.65% of the entire soil bacterial population. It was concluded that the major portion of the oxidative capacity of the soil microflora resides on the metabolic activities of the mesophilic bacteria operating within the temperature range of 10°C to 25°C even though conditions may be out of the mesophilic range.

Benedict and Carlson (78) conducted tests on temperature acclimation in aerobic bio-oxidation systems. The experiments indicated that acclimation time was related to the amount

and rate of temperature change.

Acclimation tests involved changing the temperature from 19°C down to 4°C and from 19°C to 32°C. The 4°C temperature occurred within about two weeks. Acclimation at 32°C had not occurred after 26 days. The study indicated that low temperature effects were less than high temperature effects when compared to a 19°C culture.

4. EXPERIMENTAL PROCEDURE

The experimental procedures used for this investigation are discussed in several sections. These include:

- (a) special apparatus;
- (b) feed source;
- (c) laboratory extended aeration plant operation; and,
- (d) tests and analytical procedures.

4.1. SPECIAL APPARATUS

This investigation was dependent upon precise temperature control of the model extended aeration waste treatment plant. A controlled-temperature environment was maintained within a walk-in environmental chamber. The chamber used for this investigation was an Enconaire unit model number ER608LT, manufactured by Enconaire Systems Ltd. of Winnipeg, Manitoba, Canada. This unit was capable of maintaining a temperature setting within $\pm 0.5^{\circ}\text{C}$ for the 0°C to 20°C temperature test range used. Photographs of this unit are shown as Figure 8.

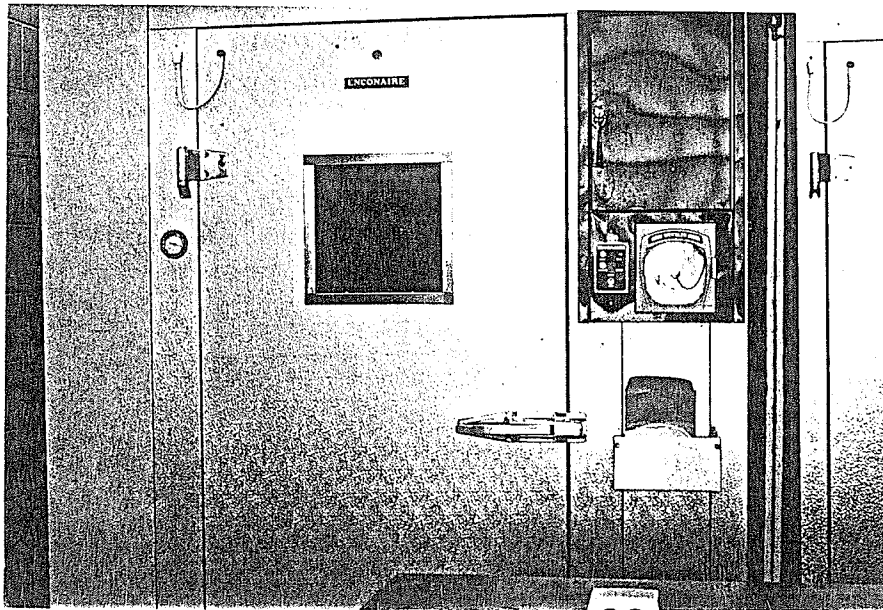
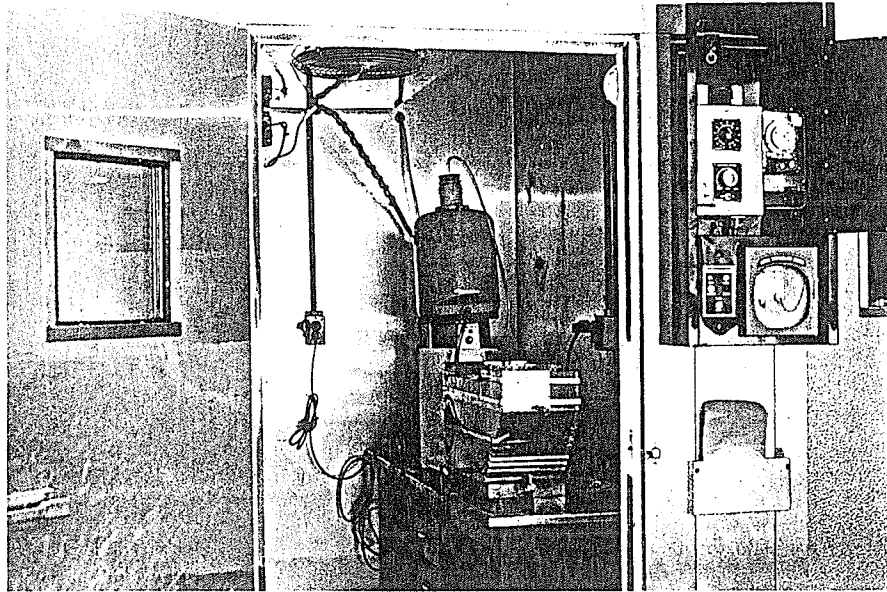


Fig. 8. Enconaire Environmental Chamber.

The extended aeration unit was designed to treat approximately 19 litres of typical raw domestic sewage per day in accordance with the design criteria set out in Ten State Standards (15). The design calculations are given in Appendix 1. The unit was constructed of $\frac{1}{4}$ inch acrylic plastic sheet according to the dimensions in Figure 30 of Appendix 1. The two baffles which made up the energy dissipation section were constructed as follows:

(a) Baffle number 1 indicated in Figure 30 of Appendix 1 is shown below as Figure 9.

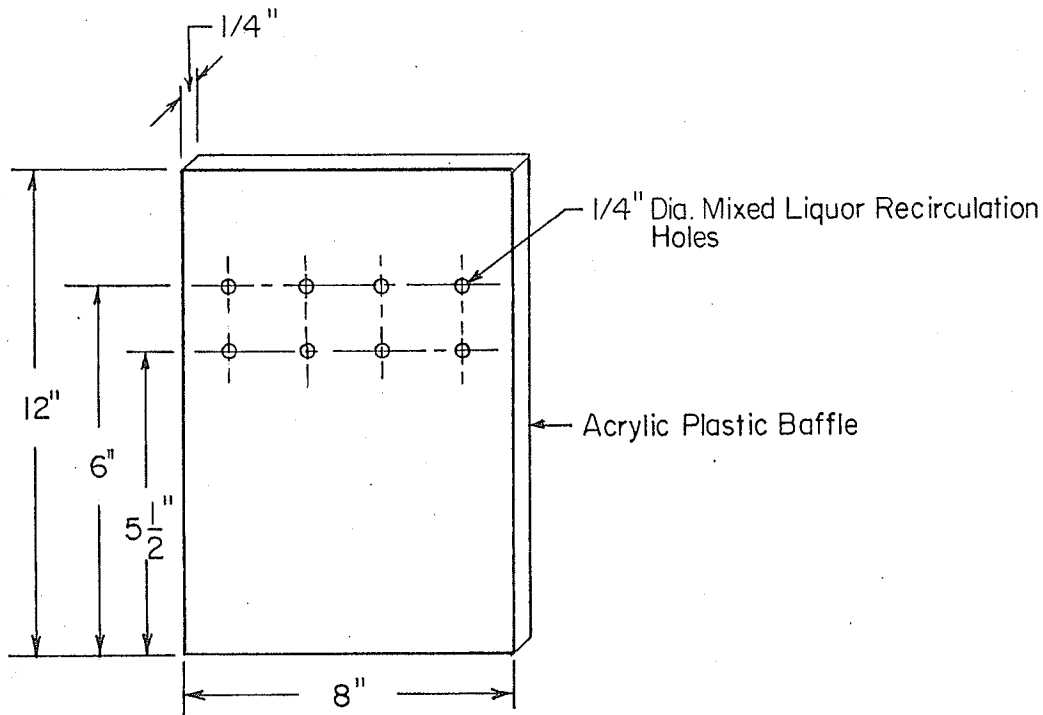
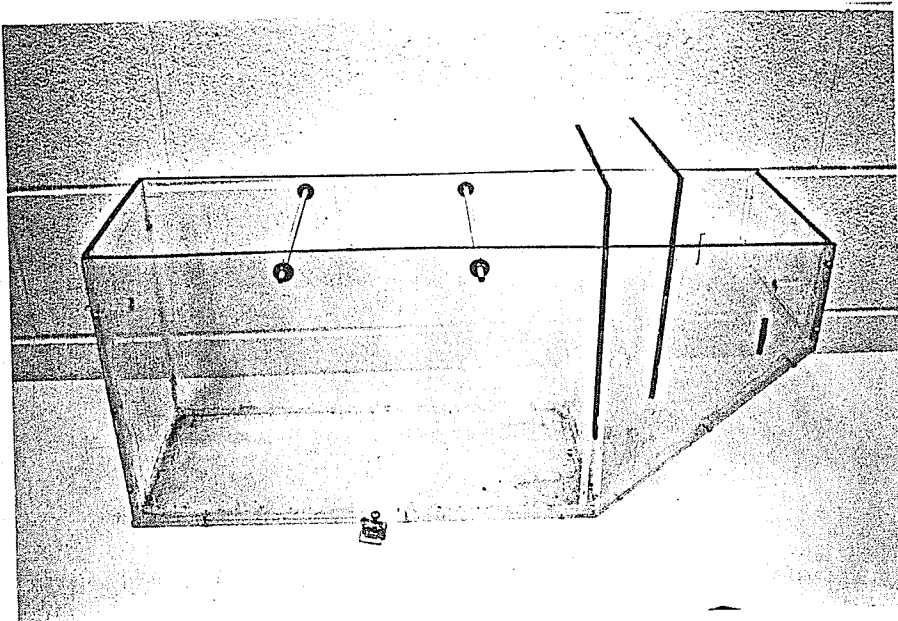


Fig. 9. Baffle Number One of Laboratory Extended Aeration Plant.

This baffle was adjusted in the vertical plane to ensure that adequate sludge return would occur. The $\frac{1}{4}$ inch diameter holes were located about $\frac{1}{2}$ inch below the aeration basin surface so that the flow of mixed liquor into the energy dissipation area would not create excessive turbulence.

(b) Baffle number 2 indicated in Figure 30 of Appendix 1 acted as the impingement surface for the mixed liquor as it passed through the holes in baffle number 1. It also deflected the mixed liquor downward to ensure mixed liquor recirculation back to the aeration chamber. Photographs of the laboratory unit are shown as Figure 10.



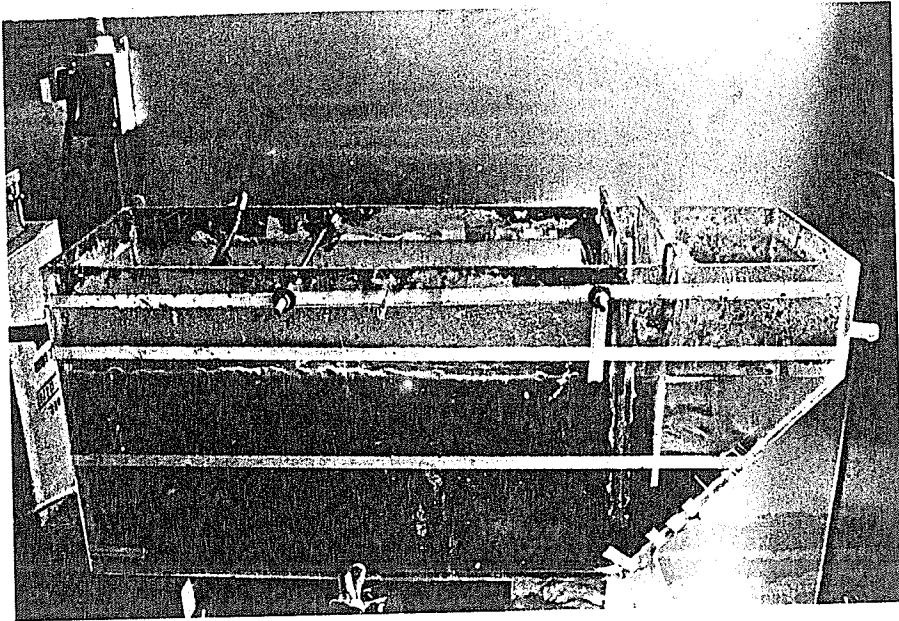
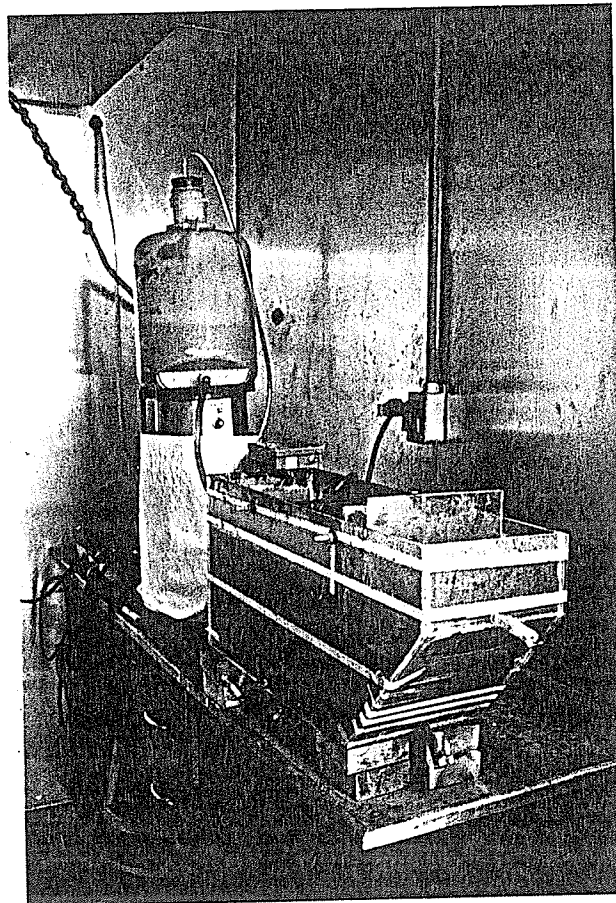


Fig. 10. Laboratory Extended Aeration Plant.

The apparatus which was used to feed the extended aeration plant is shown in Figure 11.



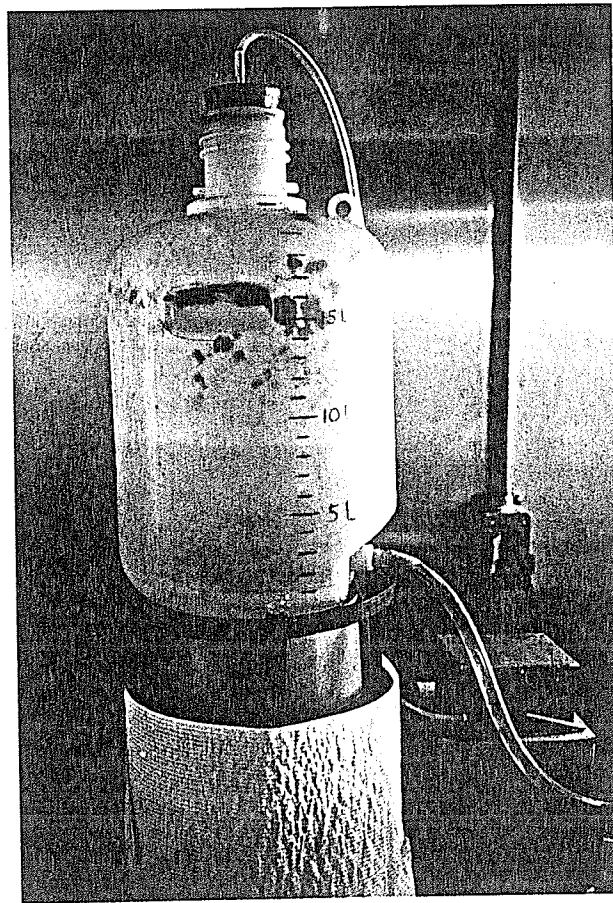


Fig. 11. Laboratory Plant Feed Apparatus.

Although continuous feeding in the true sense was not achieved, it was closely approximated by using a timer to activate a solenoid to intermittently allow air to enter the reservoir and allow feed to flow into the aeration basin. The rate of flow for each feed cycle was held constant by immersing the air inlet tube to within one inch of the reservoir bottom. The timer on-off cycle was adjustable which allowed a steady 24-hour feed distribution. An on cycle of 5 seconds every 10 minutes resulted in the desired food flow rate of 19 litres every 24 hours.

The potential problem of solids settling in the storage reservoir and clogging the feed line was minimized by stirring the reservoir contents every 5 minutes for 30 seconds using a magnetic stirring device. Elevating the reservoir and operating the one-half inch diameter feed line full at all times provided a positive syphoning action with every on-cycle.

4.2. FEED SOURCE

The wastewater that was used as feed consisted of screened raw domestic sewage, obtained from the City of Winnipeg's South End Water Pollution Center. The samples were taken from the degritting chambers once a week. The sampling schedule was arranged so that the wastewater was obtained at approximately 11:00 a.m. every Friday. Thirty-five gallons of wastewater were taken to the laboratory where it was stored in the environmental chamber.

4.3. LABORATORY EXTENDED AERATION PLANT OPERATION

The operation of the laboratory extended aeration plant consisted of maintaining a system that most closely simulated an actual field operating plant. Continuous flow was necessary to simulate field conditions which required daily filling of the feed reservoir. The effluent was collected in a 25 litre plastic container and removed daily for analysis and disposal.

The laboratory test unit was started by filling the aeration basin with activated sludge obtained from the City of Winnipeg's North End Water Pollution Control Center which operates a conventional activated sludge process.

The testing program began at 20°C. The test temperatures used and the duration of each test is shown below as Table 2. As the temperature was decreased to 0°C, the duration of each test was chosen to ensure steady state operation.

The duration time of each test as the temperature was increased from 0°C to 10°C was governed by nitrification studies as well as steady state operations.

Test Temperature (°C)	Duration (days)
20	34
15	49
10	19
7	14
5	19
3	17
1	24
0	54
1	7
3	6
5	4
7	8
10	6

TABLE 2. Temperature and Duration of Laboratory Tests.

Sludge wasting was not intentionally done on a regular basis. When operating conditions were such that sludge build-up was noted in the clarifier, some sludge was wasted to ensure proper clarifier operation.

The air supplied to the unit was monitored via a rotometer. The air flow was adjusted to ensure adequate mixing in the aeration basin. The amount of air that was needed was governed by the mixing requirement.

4.4. TESTS AND ANALYTICAL PROCEDURES

Tests were performed on a daily and intermittent basis throughout the investigation period. All analytical tests were carried out in accordance with the procedure outlined in Standard Methods of the Examination of Water and Wastewater (79) hereafter referred to as Standard Methods.

Daily tests in accordance with the appropriate sections of Standard Methods (79) are given below:

- (a) Biochemical Oxygen Demand on feed and effluent as given in Sections 218B and 219.

Incubation of replicate samples was done at extended aeration operating temperatures;

- (b) Suspended solids and volatile suspended solids (non-filterable residue) on the aeration basin mixed liquor and effluent as given in Section 224C;

(c) Turbidity tests on the effluent using a Hellige turbidimeter;

(d) Sludge volume index on aerator mixed liquor as given in Section 210C;

(e) The amount of daily flow by recording the time and amount of feed required to fill the reservoir;

(f) A Radiometer pH meter was used to measure the pH of the feed and aerator mixed liquor; and,

(g) The environmental chamber air temperature was monitored continuously by a recording chart on the environmental chamber. A laboratory thermometer was used to verify the recorder as well as measure the temperature of the mixed liquor.

Tests that were performed intermittently in accordance with Standard Methods where applicable include:

(a) Chemical oxygen demand on feed and effluent as given in Section 220;

(b) Ammonia nitrogen, organic nitrogen and nitrate nitrogen on feed and effluent as given in Sections 132A and B, 135 and 213C respectively;

(c) Total phosphate on feed, aerator mixed liquor and effluent as given in Sections 223 III and 223E; and,

(d) Oxygen uptake tests were conducted using a YSI oxygen meter with a BOD bottle stirring accessory. Tests were performed by drawing a sample of mixed liquor from the aeration basin, transferring the sample to a BOD bottle, and observing the oxygen depletion with time.

5. EXPERIMENTAL RESULTS

The experimental results obtained from this investigation will be presented as:

- (a) analytical test results, which will include the data obtained using the analytical procedures described in section 4.4.; and,
- (b) biokinetic interpretation, which will use the basic data to calculate the parameters used to describe the Monod growth and substrate utilization parameters of extended aeration.

5.1. ANALYTICAL TEST RESULTS

5.1.1. Biochemical Oxygen Demand Tests

The biochemical oxygen demand tests were performed daily on the feed and the treated effluent. The average feed and effluent BOD_5 including percent removal for a given temperature is shown below in Table 3 and Figure 12.

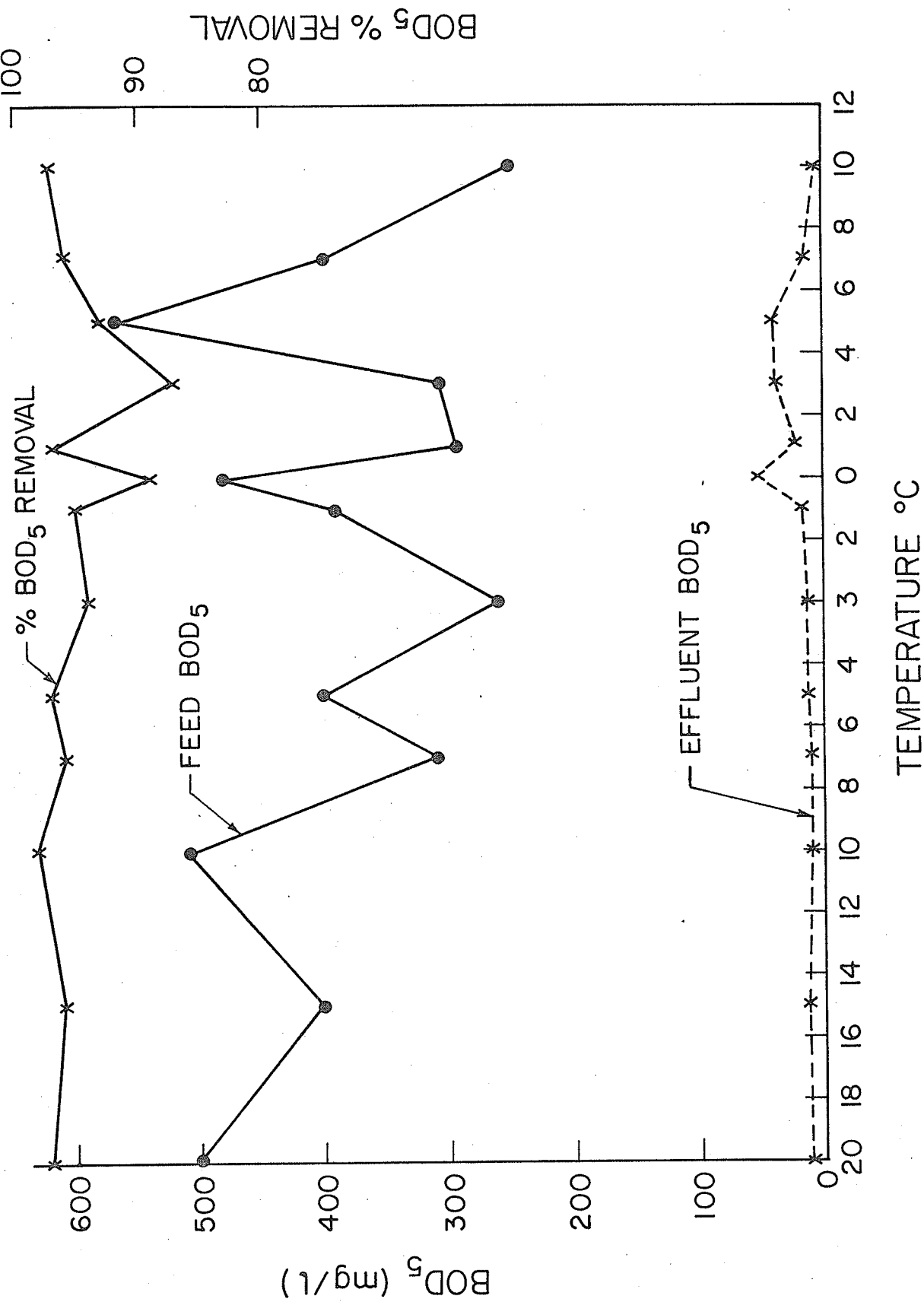


Fig.12.Average Feed and Effluent BOD₅ and Percent BOD₅ Removal

Temperature °C	Average Feed BOD ₅ (mg/l)	Average Effluent BOD ₅ (mg/l)	% BOD removed
20	500	12	97
15	400	15	96
10	510	10	98
7	310	12	96
5	400	13	97
3	260	15	94
1	390	19	95
0	480	53	89
1	295	23	92
3	307	40	87
5	568	41	93
7	395	16	96
10	250	8	97

TABLE 3. Temperature Effect on BOD₅ Removal Efficiency.

5.1.2. Chemical Oxygen Demand Tests

The chemical oxygen demand tests were done at least once a week on the feed and effluent. The average COD for the feed and effluent and the percent COD removal is shown below as Table 4 and Figure 13.

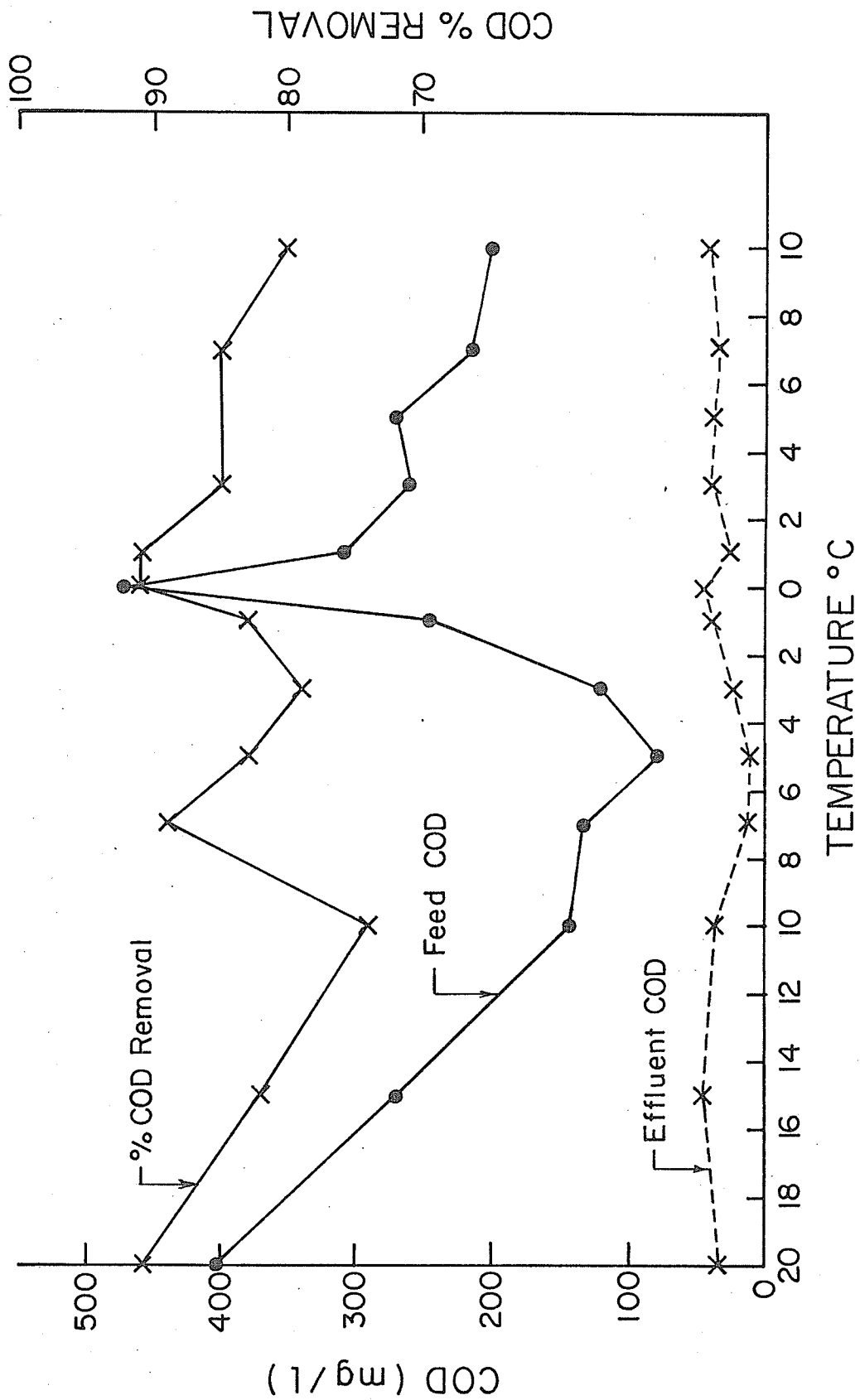


Fig. 13. Average Feed and Effluent COD and Percent COD Removal

Temperature °C	Average Feed COD (mg/l)	Average Effluent COD (mg/l)	% COD removed
20	405	35	91
15	270	49	82
10	145	38	74
7	135	15	89
5	80	14	83
3	122	26	79
1	246	41	83
0	473	45	91
1	310	27	91
3	260	41	85
5	250	38	85
7	214	33	85
10	200	40	80

TABLE 4. Temperature Effect on COD Removal Efficiency.

5.1.3. Solids Test

5.1.3.1. Feed Solids Tests

Suspended and volatile suspended solids tests were done daily on the aeration basin mixed liquor and the effluent. Intermittent suspended and volatile suspended solids tests were done on the feed. The average suspended, volatile suspended and percent volatile suspended solids of the feed for a given temperature are shown below as Table 5 and Figure 14.

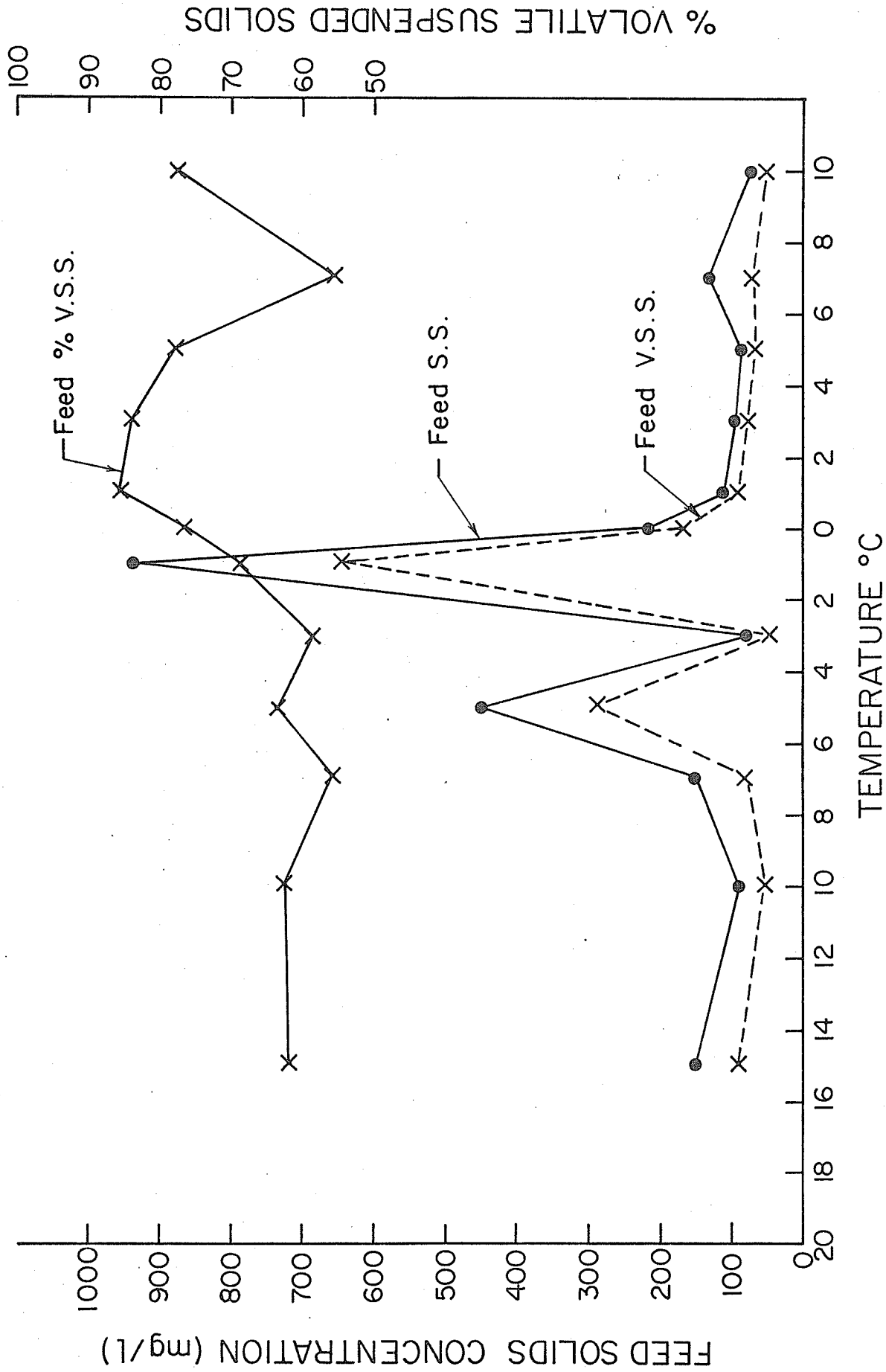


Fig. 14. Average Feed Suspended, Volatile Suspended and Percent Volatile Suspended Solids

Temperature °C	Average Feed Suspended Solids (mg/l)	Average Feed Volatile Suspended Solids (mg/l)	% Volatile Suspended Solids
20	no test	---	---
15	158	98	62
10	90	57	63
7	155	87	56
5	450	287	64
3	80	47	59
1	940	650	69
0	220	170	77
1	114	98	86
3	95	80	84
5	90	70	78
7	135	75	56
10	73	57	78

TABLE 5. Feed Suspended, Volatile Suspended and Percent Volatile Suspended Solids.

5.1.3.2. Mixed Liquor Solids Test

The average suspended, volatile suspended and percent volatile suspended solids of the mixed liquor for a given temperature are given below as Table 6 and Figure 15.

Temperature °C	Average Mixed Liquor Suspended Solids (mg/l)	Average Mixed Volatile Suspended Solids (mg/l)	% Volatile Suspended Solids
20	4725	2450	52
15	7400	3590	51
10	6520	3260	50
7	6770	3380	50
5	8255	4160	50
3	7950	4100	51
1	8100	4525	56
0	7200	4300	60
1	6400	4355	68
3	5630	3900	69
5	5450	3850	70
7	6000	4225	70
10	6500	4150	64

TABLE 6. Mixed Liquor Suspended, Volatile Suspended and Percent Volatile Suspended Solids.

5.1.3.3. Effluent Solids Tests

The average suspended, volatile suspended and percent volatile suspended solids of the effluent for a given temperature are given below as Table 7 and Figure 16.

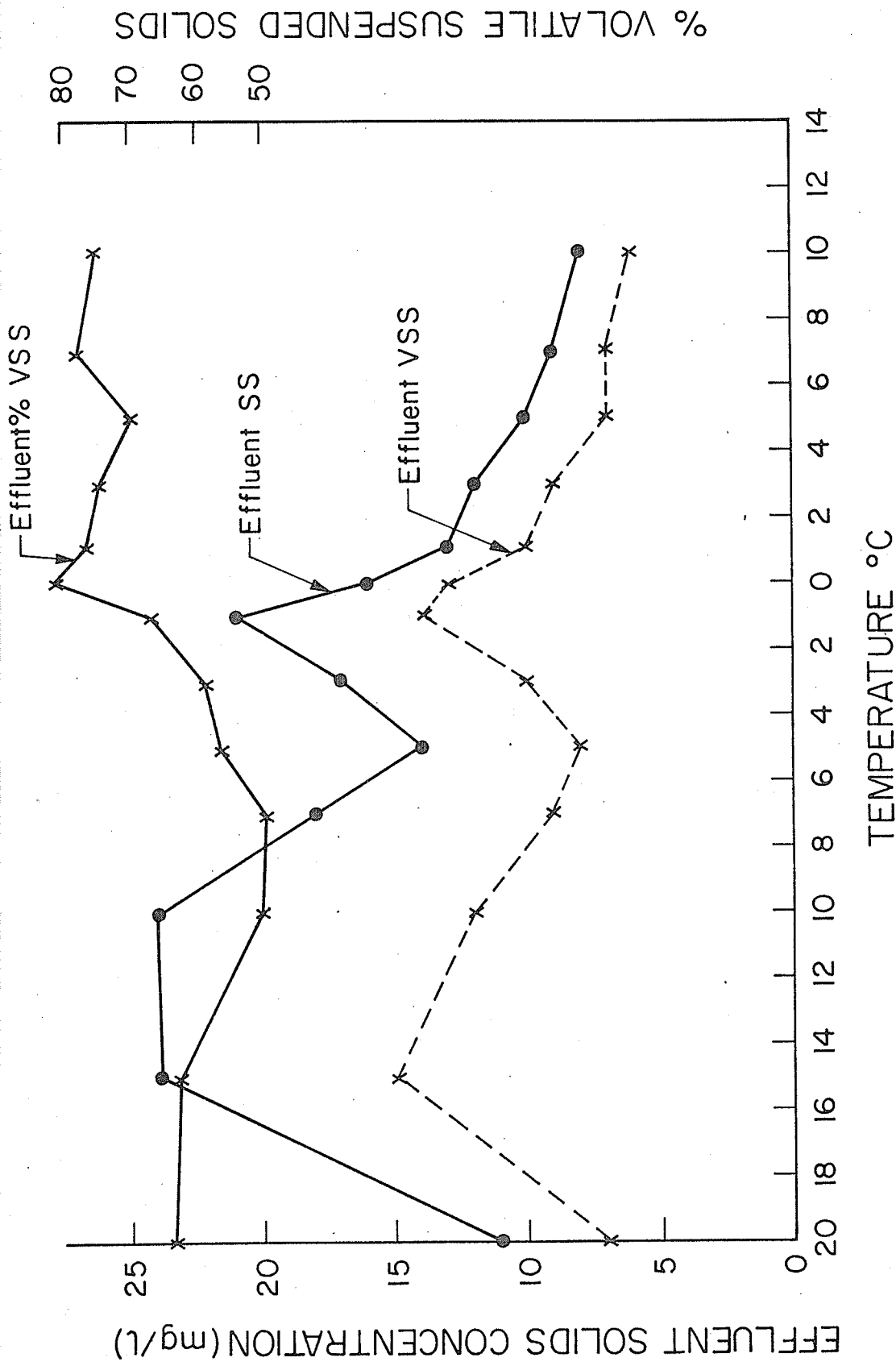


Fig. 16. Effluent Suspended, Volatile Suspended, and Percent Volatile Suspended Solids

Temperature °C	Average Effluent Suspended Solids (mg/l)	Average Effluent Volatile Suspended Solids (mg/l)	% Volatile Suspended Solids
20	11	7	64
15	24	15	63
10	24	12	50
7	18	9	50
5	14	8	57
3	17	10	59
1	21	14	67
0	16	13	82
1	13	10	77
3	12	9	75
5	10	7	70
7	9	7	78
10	8	6	75

TABLE 7. Effluent Suspended, Volatile Suspended and Percent Volatile Suspended Solids.

5.1.4. Sludge Volume Index

The average sludge volume index of the mixed liquor is given below in Table 8 and Figure 17. Table 8 and Figure 18 show the settled volume of the mixed liquor, i.e. the sludge-liquid interface in a 1000 ml graduated cylinder after settling for one-half hour.

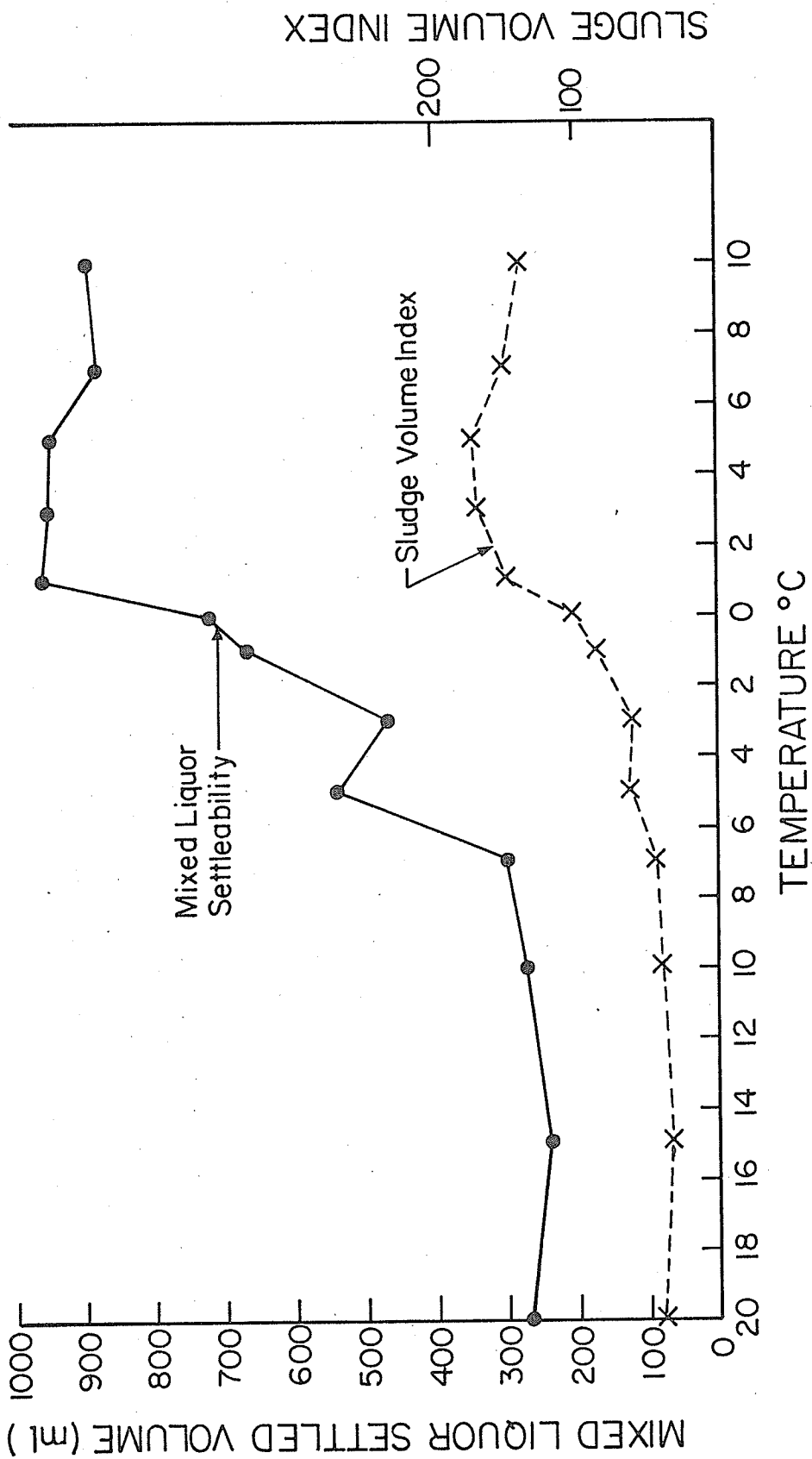


Fig. 17. Mixed Liquor Settled Volume and Sludge Volume Index

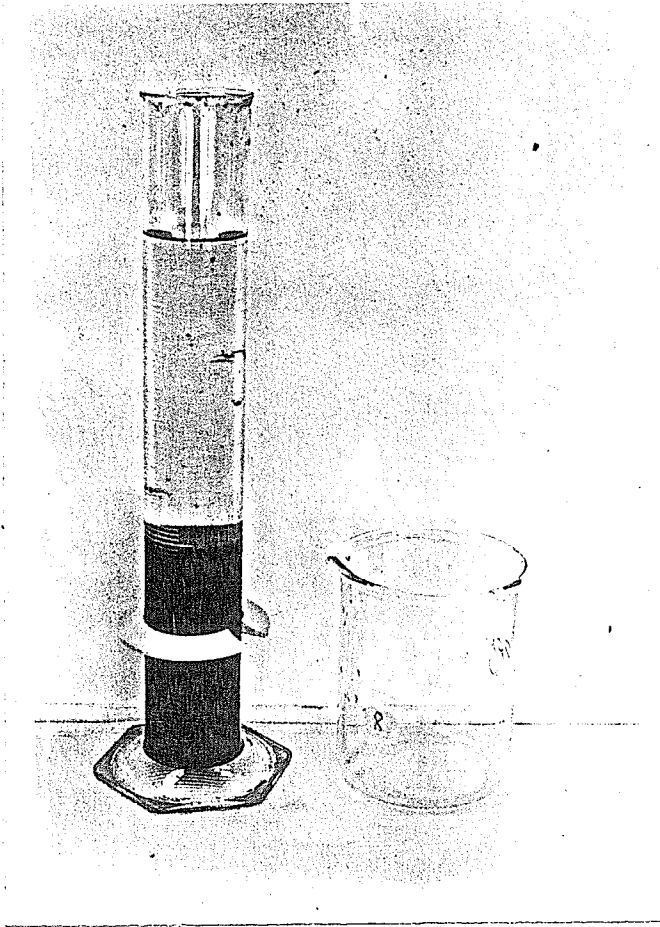


Fig. 18. Settled Mixed Liquor in 1000 ml Graduated Cylinder after Settling for ½ hour.

Temperature °C	Average Settled Volume (ml)	Average SVI
20	269	40
15	240	35
10	275	42
7	300	44
5	540	63
3	470	60
1	670	87
0	720	102
1	960	150
3	950	170
5	945	175
7	880	150
10	895	140

TABLE 8. Mixed Liquor Settled Volume and Sludge Volume Index.

5.1.5. Turbidity and pH Tests

The average mixed liquor pH and effluent turbidity at a given temperature is given below in Table 9 and Figure 19.

Temperature °C	Average pH	Average Turbidity (J.T.U.)
20	7.7	no tests
15	7.7	19
10	8.1	12
7	8.3	8
5	8.0	3
3	8.1	5
1	8.0	7
0	8.2	9
1	8.2	5
3	8.0	5
5	7.9	4
7	7.4	4
10	7.2	4

TABLE 9. Mixed Liquor pH and Effluent Turbidity.

5.1.6. Oxygen Uptake Rate Tests

The average oxygen uptake rate of the mixed liquor for a given temperature is given below as Table 10 and Figure 20.

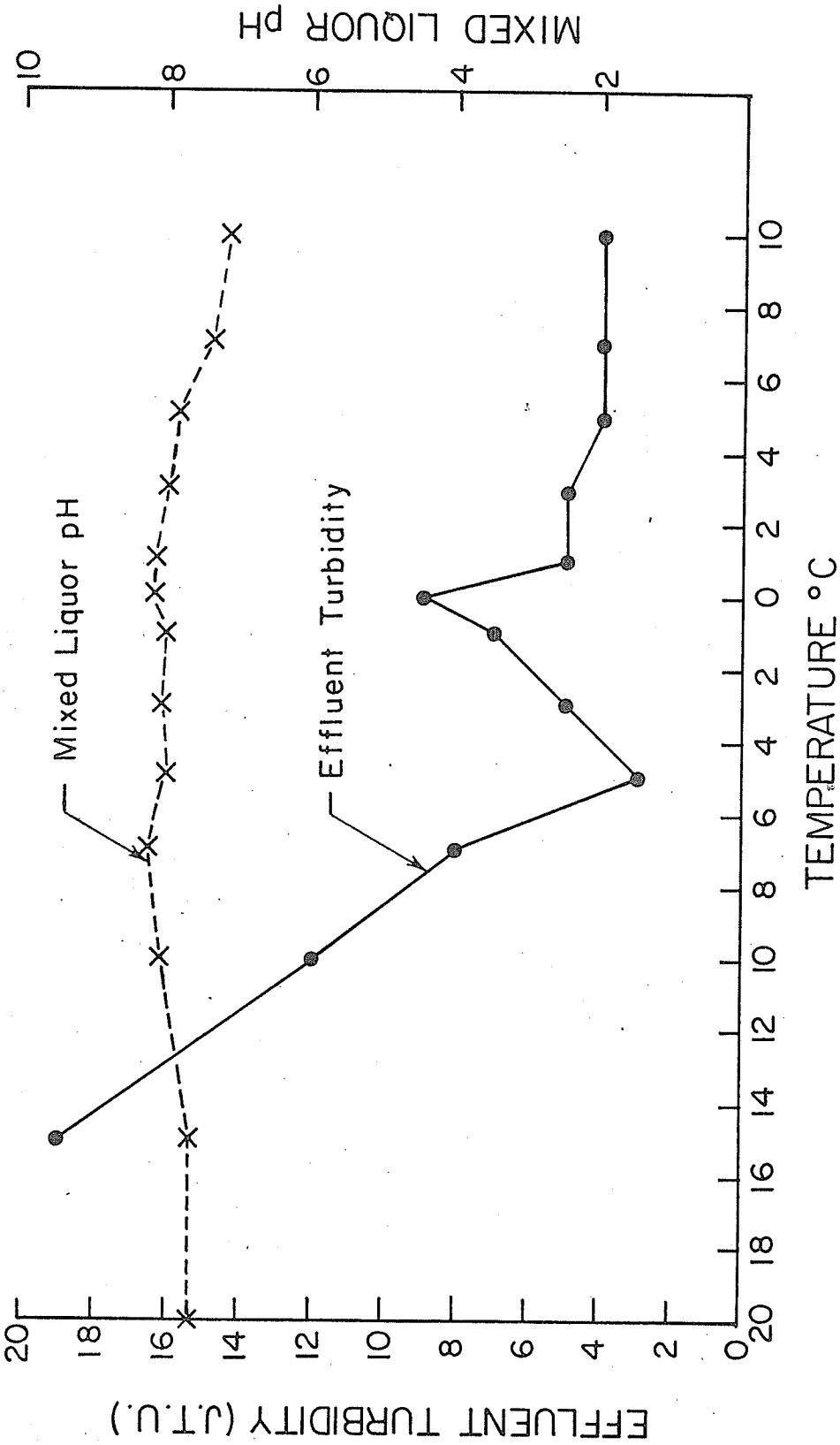


Fig. 19. Mixed Liquor pH and Effluent Turbidity

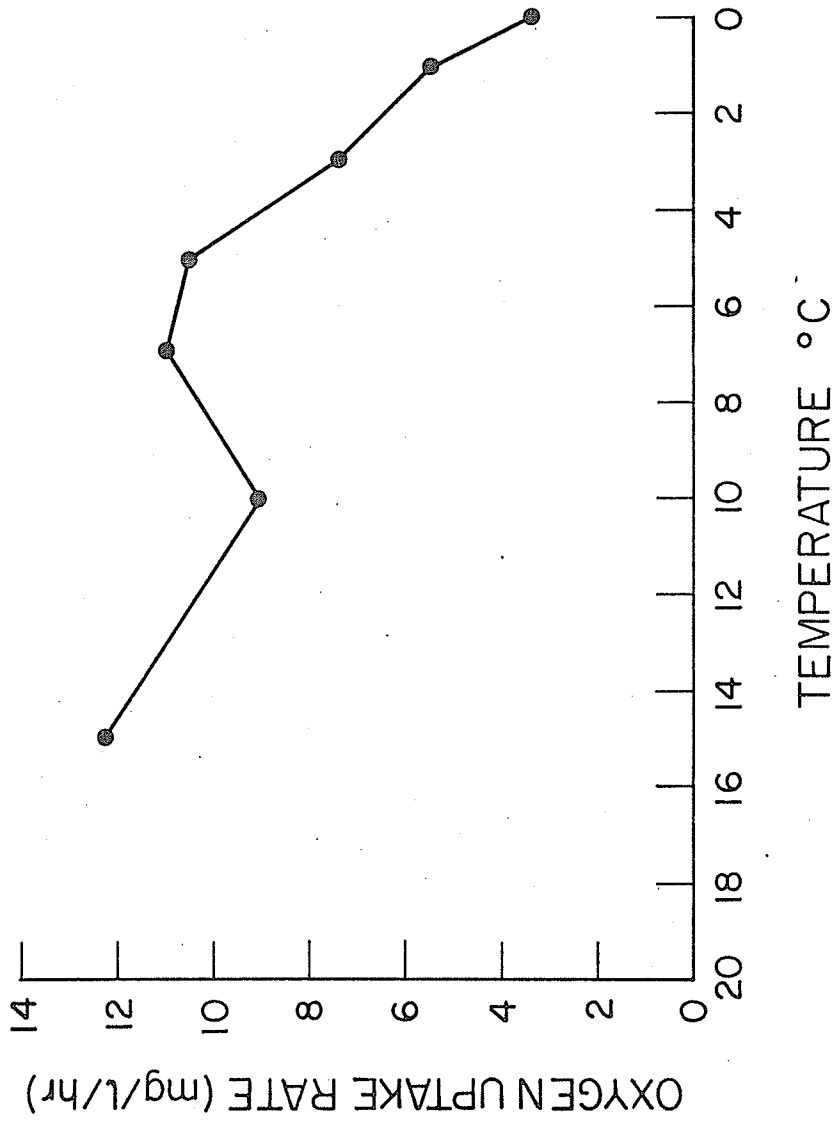


Fig. 20. Mixed Liquor Oxygen Uptake Rate

Temperature °C	Average Oxygen Uptake Rate (mg/l/hr)
15	12.4
10	9.2
7	11.1
5	10.4
3	7.4
1	5.7
0	3.5

TABLE 10. Mixed Liquor Oxygen Uptake Rate.

5.1.7. Nitrogen Tests

The nitrogen test results of the feed and treatment plant effluent are shown below in Table 11 and Figure 21.

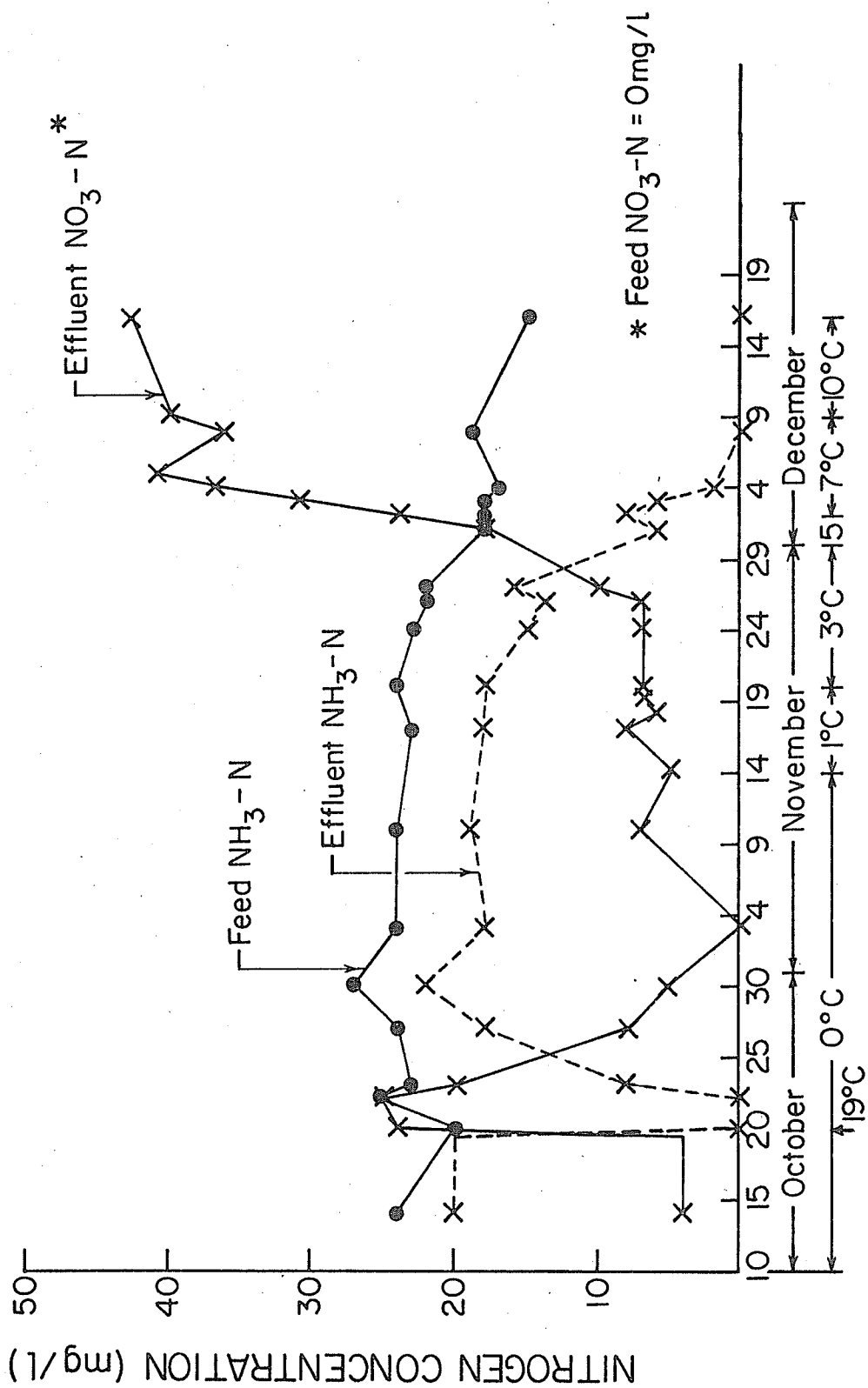


Fig. 21. Nitrogen Tests on Treatment Plant Feed and Effluent

Date	Temperature °C	NH ₃ -N (mg/l)		NO ₃ -N (mg/l)	
		Feed	Effluent	Feed	Effluent
Oct. 14	0	24	20	0	4.4
20	19	20	0	0	24
22	0	25	0	0	25
23	0	23	8	0	20
27	0	24	18	0	7.7
30	0	27	22	0	5.2
Nov. 3	0	24	18	0	0
10	0	24	19	0	7
14	0	--	--	0	5
17	1	23	18	0	8
18	1	--	--	0	6
19	1	--	--	0	7
20	1	24	18	0	7
24	3	23	15	0	7
26	3	22	14	0	7
27	3	22	16	0	10
Dec. 1	5	18	6	0	18
2	5	18	8	0	24
3	7	18	6	0	31
4	7	17	2	0	37
5	7	--	--	0	41
8	7	19	0	0	36
9	10	15	0	0	43

-- Indicates no test

TABLE 11. Ammonia and Nitrate Nitrogen of Treatment Plant Feed and Effluent.

Organic nitrogen tests on the feed and effluent never indicated more than a "trace" concentration.

5.1.8. Total Phosphorus Tests

The total phosphorus test results of the feed, reactor mixed liquor and effluent are given below in Table 12 and Figure 22.

Date	Temperature °C	Total PO ₄ -P (mg/l)			
		Feed	Effluent	% Removal	Mixed Liquor
Oct. 14	0	10.0	6.0	40	--
20	19	5.7	3.4	40	--
27	0	6.0	3.7	38	--
Nov. 3	0	14.0	6.2	56	--
6	0	16.0	6.0	63	--
10	0	9.8	5.0	49	--
12	0	15.0	4.7	69	123.0
13	0	11.6	3.3	71	116.0
17	1	7.4	4.3	42	135.5
24	3	9.0	4.3	51	125.0
Dec. 1	5	6.6	3.7	44	120.0
8	7	8.0	4.0	50	116.0
16	10	8.0	5.3	34	110.0

TABLE 12. Total Phosphorus of Feed, Aerator Mixed Liquor and Effluent.

5.2. BIOKINETIC INTERPRETATION

5.2.1. Kinetic Growth and Substrate Utilization Rate Constants

The kinetic growth and substrate utilization rate constants were calculated according to the kinetic theory

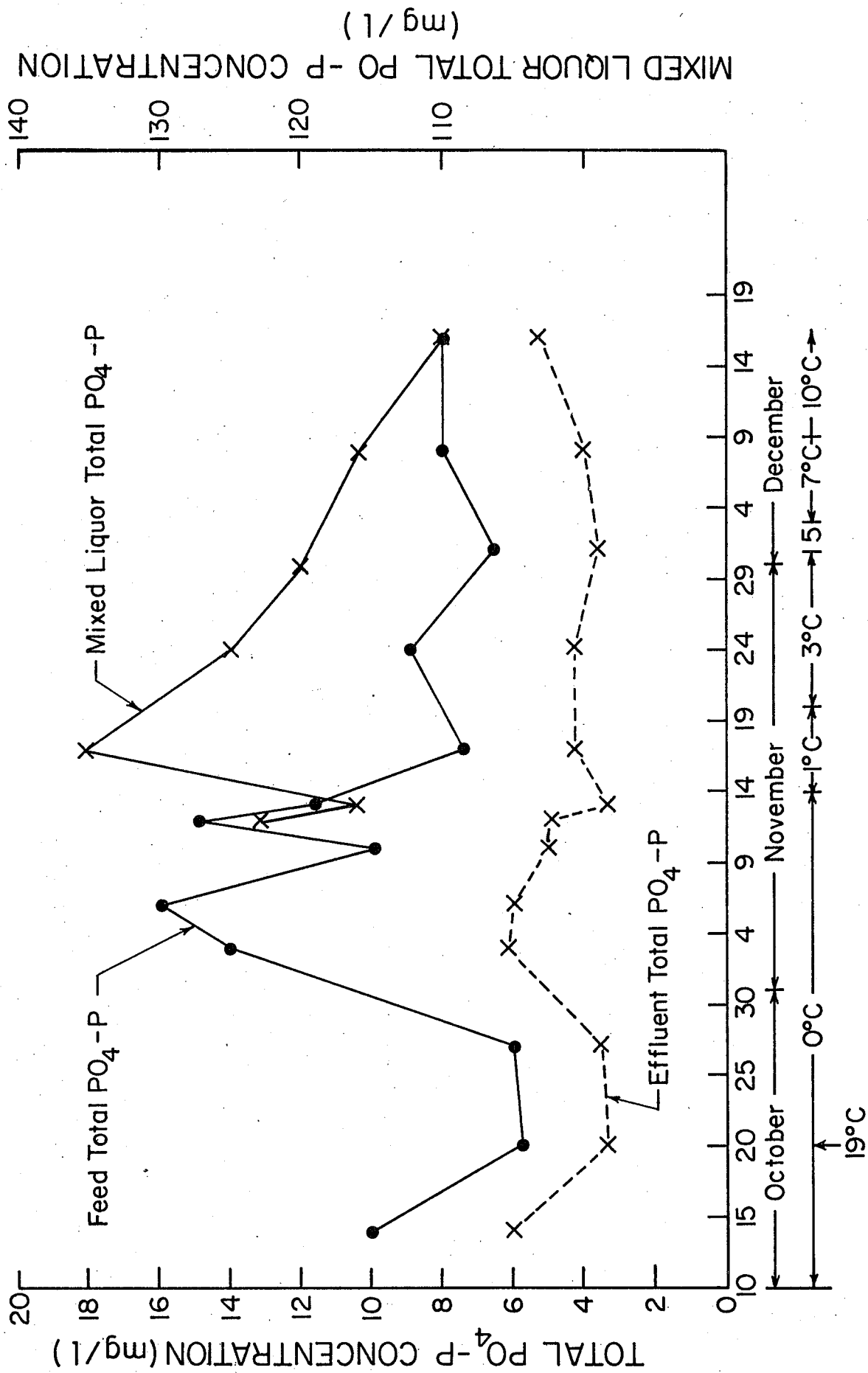


Fig. 22. Total Phosphorus of Feed, Mixed Liquor and Effluent

outlined in Section 3.2.4. An example calculation for an average feed BOD₅ concentration of 346 mg/l at 0°C is given in Appendix 2. A plot of the calculated data in Appendix 2 is shown as Figures 31 and 32 .

All the kinetic constants were evaluated as in the example in Appendix 2. A summary of the results is given below as Table 13.

5.2.2. Temperature Effects on Extended Aeration Based Upon BOD₅ Removal

Eckenfelder (41) developed a simplified substrate removal equation in a completely mixed activated sludge system as follows:

$$K_a S = \frac{S_r}{X_v t}$$

where

K_a = BOD removal rate constant;

S = effluent BOD;

S_r = BOD removed;

X_v = average mixed liquor volatile suspended solids; and,

t = retention time.

The BOD rate removal constant is obtained by plotting the mg of BOD₅ removed per day per mg mixed liquor volatile suspended solids versus the effluent BOD₅ using

TEMPERATURE = 20°C

N	Avg. BOD ₅ * mg/l	Y Wt O ₂ -Eq Wt BOD ₅	k _d x 10 ³ /hr	r	t	K _s mg/l	μ _{max} x 10 ³ /hr	r	t
7	244	0.107	0.066	0.53	13.17	112	1.72	0.86	3.8
4	515	1.00	4.22	0.79	4.58	0	4.35	--	--
4	658	0.166	0.638	0.97	12.48	0	0.69	--	--
Avg. COD									
8	406	1.00	3.27	0.83	3.65	0	3.37	--	--

TEMPERATURE = 15°C

6	240	0.520	0.470	0.69	1.93	14	1.14	0.75	2.27
9	403	1.00	2.25	0.96	9.07	0	2.30	--	--
4	585	0.197	0.518	0.80	1.89	4.5	0.85	0.85	0.82

TEMPERATURE = 10°C

6	252	0.073	0.045	0.73	3.13	0	0.098	--	--
5	366	0.096	0.138	0.84	2.68	0	0.210	--	--
6	430	0.235	0.100	0.99	14.04	0	1.130	--	--
4	543	0.136	0.343	0.97	5.64	0.25	0.500	0.28	0.41

* All values BOD₅ except where indicated as COD

.... con't

con't

TEMPERATURE = 10°C

N	Avg. COD mg/l	Y Wt O ₂ -Eq Wt BOD ₅	k _d x 10 ³ /hr	r	t	K _s mg/l	μ _{max} x 10 ³ /hr	r	t
4	159	1.00	0.69	0.98	6.96	9	0.952	0.33	0.49

TEMPERATURE = 7°C

10	220	1.00	1.19	0.86	4.77	0	1.250	--	--
7	314	1.00	1.69	0.98	11.0	0	1.750	--	--
8	486	0.221	0.569	0.93	6.20	0	0.617	--	--

Avg. BOD₅
mg/l

TEMPERATURE = 5°C

6	195	1.00	0.800	0.68	1.85	0	0.850	--	--
5	412	0.055	0.025	0.64	1.44	0	0.203	--	--
5	544	1.00	3.120	0.83	2.58	0	3.20	--	--
6	653	0.018	0	0.63	1.62	6	0.075	0.69	1.91

-106-

TEMPERATURE = 3°C

7	160	0.172	0.067	0.23	0.53	0	0.143	--	--
12	260	0.088	0.050	0.50	1.63	0	0.133	--	--
6	338	0.080	0.077	0.70	1.96	0	0.156	--	--

Avg. COD
mg/l

4	160	1.00	0.596	0.49	0.79	0	0.667	--	--
---	-----	------	-------	------	------	---	-------	----	----

.... con't

con't N	Avg. BOD ₅ mg/l	TEMPERATURE = 1°C				K _s mg/l	μ _{max} x10 ³ /hr	r	t
		Y Wt O ₂ -Eq Wt BOD ₅	k _d x10 ³ /hr	r	t				
6	228	1.00	1.19	0.61	1.54	2.61	1.51	0.79	2.58
5	384	0.163	0.235	0.95	5.27	0	0.326	--	--
8	529	0.103	0.176	0.91	5.38	0	0.270	--	--
Avg. COD									
4	268	0.252	0.186	0.31	0.46	1.0	0.356	0.60	1.06
TEMPERATURE = 0°C									
4	258	0.128	0.026	0.08	0.11	0	0.152	--	--
7	346	1.00	1.58	0.75	4.09	0	1.67	--	--
6	443	1.00	2.50	0.65	1.71	0	2.63	--	--
4	545	1.00	2.62	0.73	1.51	0	2.70	--	--
5	640	0.105	0.26	0.99	22.3	0	3.37	--	--
4	710	1.00	3.67	0.85	2.28	0	3.85	--	--
5	868	1.00	4.57	0.94	4.77	0	4.76	--	--
8	398	0.060	0.015	0.14	0.34	40	0.190	0.73	2.62
Avg. COD									
mg/l									

-- Indicates not applicable

TABLE 13. Kinetic Growth Constants, Substrate Utilization Constants, Correlation and Student's t Values Based Upon BOD₅ and COD Parameters.

Cartesian co-ordinates. The slope of the line passing through the point (0, 0) is K_a , the BOD removal rate constant.

The average \bar{X} , \bar{Y} co-ordinates and resulting straight line plot used to establish K_a are shown below as Table 14 and Figures 23 and 24.

Temperature °C	Effluent BOD ₅ \bar{X}	mg BOD ₅ removal/day mg MLVSS, \bar{Y}	K_a
20	11.2	0.111	0.0099
15	14.7	0.072	0.0049
10	9.96	0.081	0.0081
7	13.4	0.063	0.0047
5	17.9	0.072	0.0040
3	21.0	0.047	0.0022
1	22.0	0.064	0.0029
0	55.0	0.077	0.0014

TABLE 14. Temperature Effects on the BOD₅ Removal Rate.

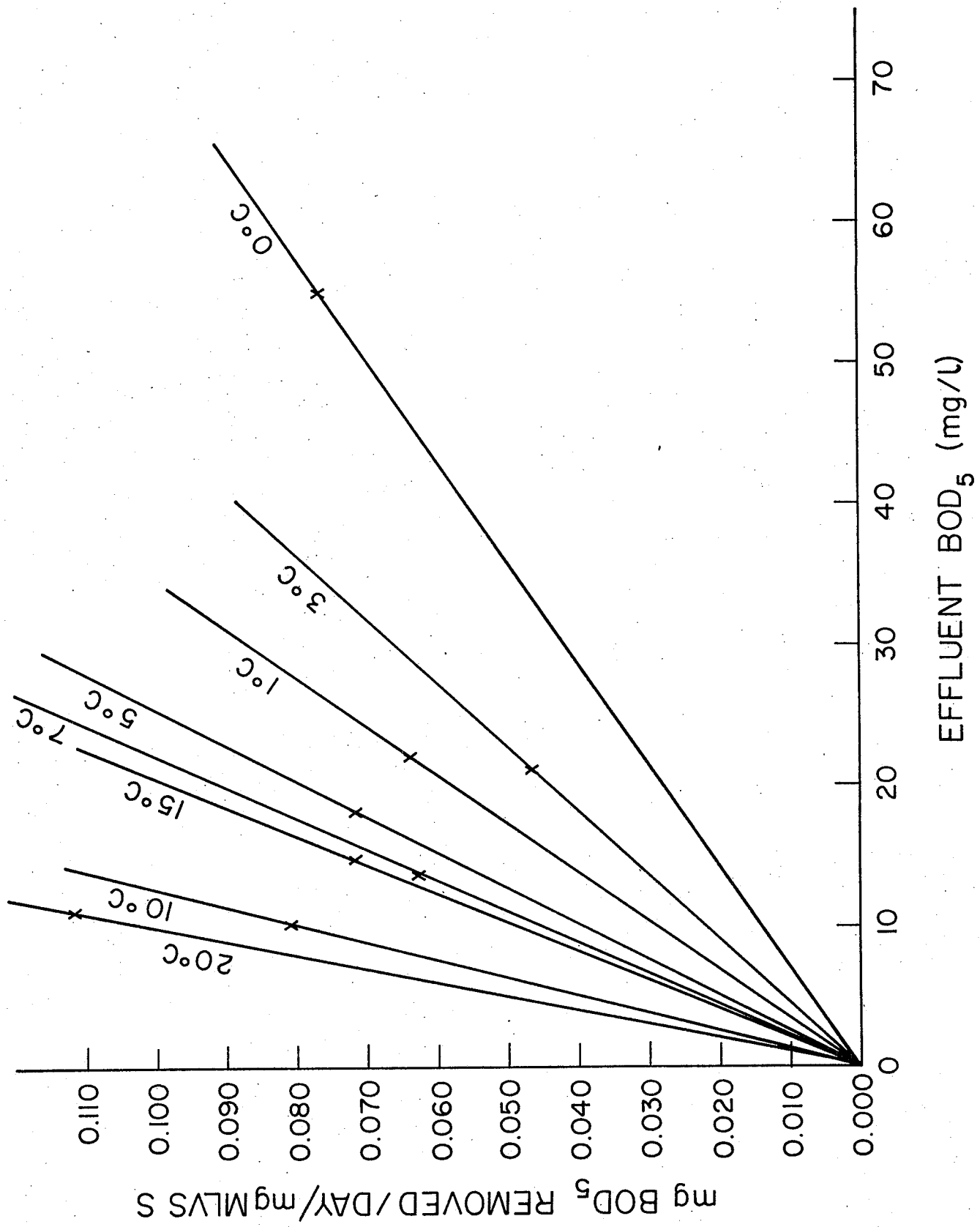


Fig. 23. Temperature Effects on BOD₅ Removal

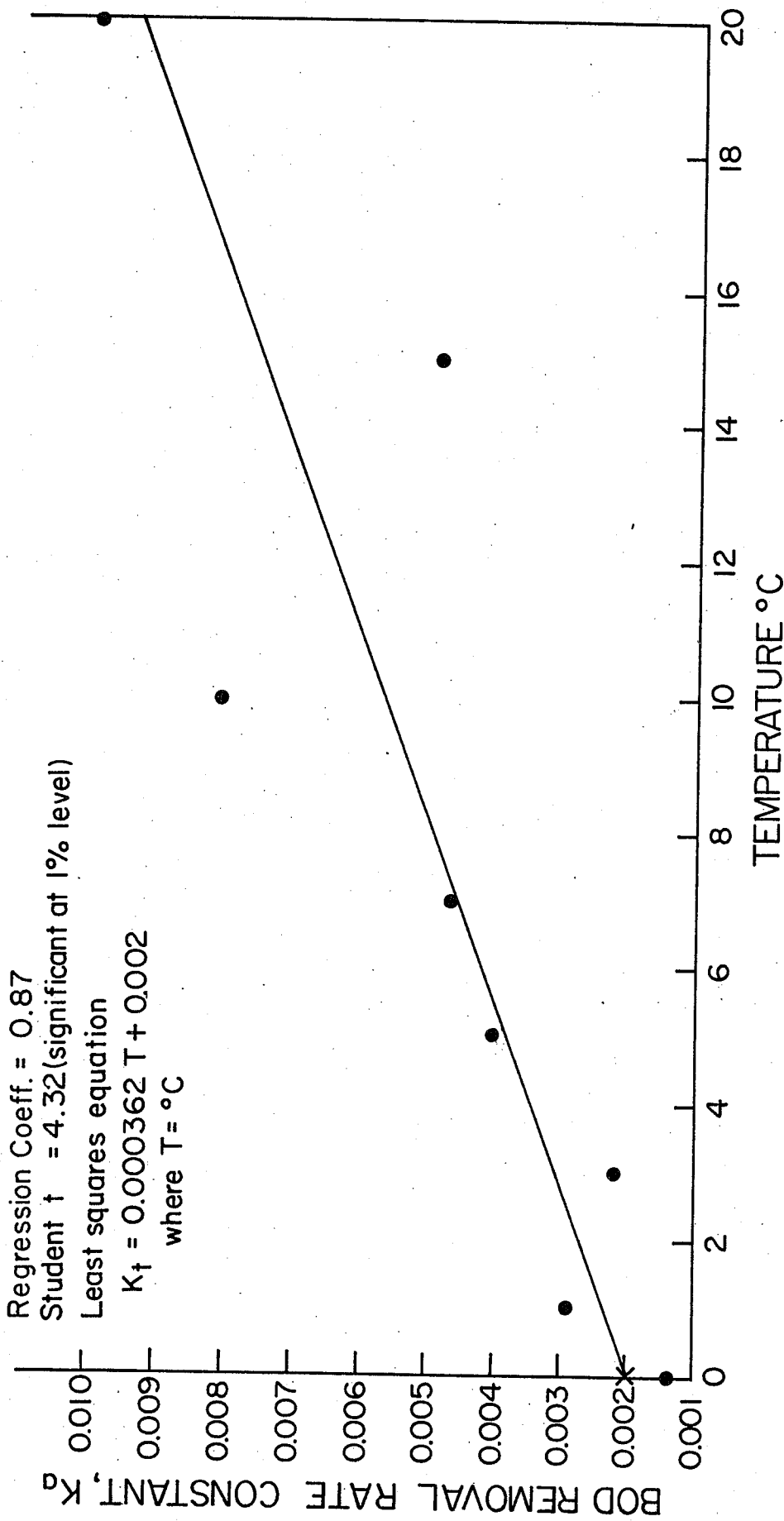


Fig. 24. Temperature Effects on BOD₅ Removal Rate Constant

5.2.3. Temperature Effects on BOD₅ Results

Replicate BOD's were run at the operating temperature of the laboratory extended aeration plant. The average BOD_T/BOD₂₀ ratio for the feed and effluent at different temperatures are shown below in Table 15

Figure 25.

Temperature °C	BOD _T /BOD ₂₀	
	Feed	Effluent
20	1	1
15	0.81	0.76
10	0.63	0.69
7	0.60	0.44
5	0.40	0.39
3	0.32	0.26
1	0.20	0.22
0	0.14	0.10

TABLE 15. Temperature Effects on the BOD_T/BOD₂₀ Ratio.

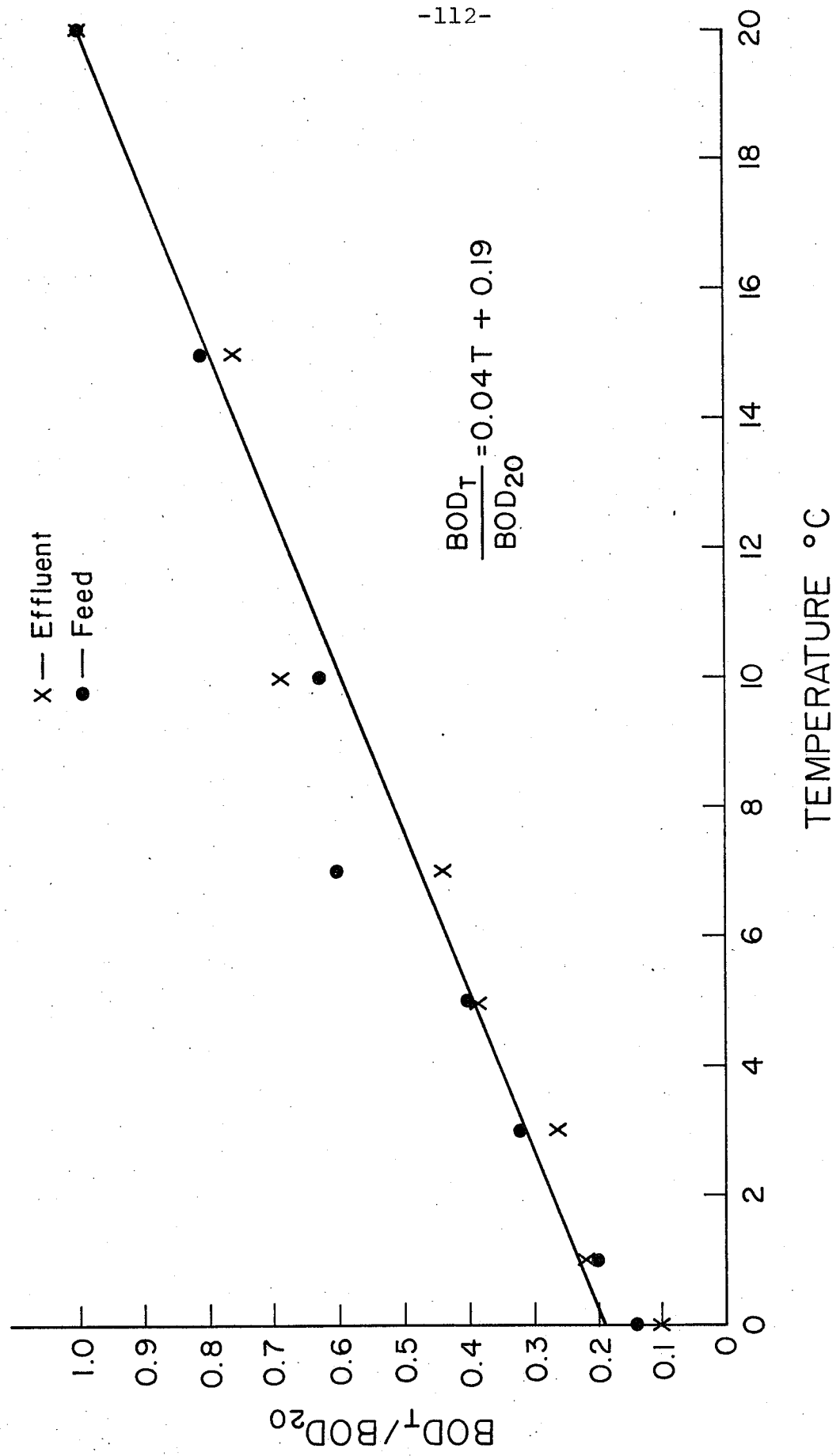


Fig. 25. Temperature Effects on the BOD_T / BOD₂₀ Ratio

6. DISCUSSION OF RESULTS

6.1. ANALYTICAL TEST RESULTS

6.1.1. Biochemical Oxygen Demand Results

The results of the biochemical oxygen demand tests are shown in Table 3 and Figure 12. The BOD₅ values of the feed ranged from 260 mg/l to 568 mg/l with an average value of 390 mg/l. The number of samples represented by each value is given in section 4.3. A seven-day feed supply was taken each Friday morning at about 11:00 a.m.. The reported 24 hour average BOD₅ for the raw sewage entering the South End Water Pollution Control Center is about 250 mg/l. The overall BOD₅ value of 390 mg/l for the entire test period is higher than the 24 hour average because 11:00 a.m. is considered a peak loading time in a typical diurnal flow pattern for domestic sewage plant (2).

The effluent BOD₅ was calculated similarly as the feed BOD₅ for each given temperature. The effluent BOD₅ values ranged from a low of 8 mg/l to a high of 53 mg/l with an average value of 21 mg/l. The plant effluent was consistently low at about 12 mg/l BOD₅ regardless of the feed BOD₅ until 1°C when a slight increase to 19 mg/l was noticed. Decreased BOD₅ removal efficiency was more pronounced at 0°C than at higher temperatures. The average effluent BOD₅ after 34 days at 0°C plant operation was 53 mg/l. This represents an 89%

BOD₅ removal efficiency. This may be contrasted by work done by Hunter et al (76) where 79% BOD was removed at 4°C using batch reactors and artificial substrate. Recovery from this cold temperature operation was relatively slow. Typical low effluent BOD₅ values of 12 mg/l were not obtained until the temperature was increased to 7°C over a 17 day period.

The BOD₅ removal efficiency ranged from a low of 87% at 3°C (increased from 0°C) to a high of 98% at 10°C (decreased from 20°C). The overall removal averaged 94%. This compares with the 75 to 95% BOD removal efficiency reported by Metcalf and Eddy (2) for extended aeration.

4.1.2. Chemical Oxygen Demand Results

The chemical oxygen demand results are shown in Table 4 and Figure 13. These results are based on weekly samples. The COD of the feed ranged from a low of 80 mg/l to a high of 473 mg/l with an average value of 240 mg/l.

The effluent COD ranged from a low of 14 mg/l to a high of 45 mg/l with an average value of 34 mg/l. The apparent decrease in COD removal efficiency, i.e. increased effluent COD, was first noted at 1°C. The highest effluent COD of 45 mg/l was recorded at 0°C. This is similar to the increase of BOD₅ at 1°C to 0°C. It is of significance to note

that the increase in effluent COD at 1°C and 2°C does not coincide with a decrease in removal efficiency. Actually the highest COD removal efficiency was noted at 0°C, i.e. 91%. No definite change in COD removal efficiency due to temperature change was observed.

The COD test normally yields results higher than the BOD₅ test for the same sample. This is primarily due to the fact that the chemical oxidation process often is able to oxidize materials which are not readily biodegradable, e.g. cellulose. On the other hand, the COD test is unable to record an oxygen demand for ammonia. If nitrifying bacteria are present in the BOD incubation bottle, the nitrogenous demand for sewage samples containing ammonia can be significant. This was indicated in the COD/BOD₅ tests done on identical feed samples. The average BOD₅/COD ratio was 1.62, indicating that ammonia exerted an additional oxygen demand not measured by the COD test. The effluent with little or no ammonia nitrogen depending on temperature (see Table 11 and Figure 21) resulted in an average BOD₅/COD ratio at 0.62. This BOD₅/COD analysis significantly pointed out the limitation of using the COD test as a parameter for measuring feed strength for aerobic biological waste systems.

6.1.3. Solids Test Results

6.1.3.1. Feed Solids Test Results

The average feed suspended and volatile suspended solids results are shown in Table 5 and Figure 14. The feed suspended solids ranged from a low of 73 mg/l to a high of 940 mg/l with an average value of 200 mg/l. The feed volatile suspended solids ranged from a low 47 mg/l to a high of 650 mg/l with an average value of 136 mg/l. The average volatile suspended solids for the feed was 68 percent. The feed solids composition is considered to be typical of a medium strength domestic sewage (2).

6.1.3.2. Mixed Liquor Solids Test Results

The mixed liquor suspended and volatile suspended solids results are shown in Table 6 and Figure 15. The laboratory unit was operated without intentional sludge wasting. Sludge was wasted only when carry-over was imminent or had occurred. Figure 15 shows how the mixed liquor concentration build-up did occur. The peak mixed liquor concentration occurred when operating at 5°C, 68 days after the experiments had started. At this time the mixed liquor concentration was approximately 10,000 mg/l and sludge was carried over in the effluent. One litre of mixed liquor was wasted each day for 6 days to stabilize

the plant operation. Stable steady state operation was achieved at the average mixed liquor concentration of 8255 mg/l as shown in Table 6.

The capability of the clarifier to separate the solids from the mixed liquor determined the maximum aeration basin mixed liquor concentration. The maximum concentration was exceeded a second time and resulted in another carry over. This occurred after 100 days of operation and at a temperature of 1°C. Five litres of mixed liquor was then wasted and steady state operation was achieved at a mixed liquor concentration of about 8100 mg/l.

Mixed liquor carry over was controlled during the remaining test period by maintaining a mixed liquor suspended solids average lower than 8000 mg/l as shown in Table 6.

The mixed liquor volatile suspended solids, commonly used as an index of viable cell mass, showed a definite trend as the operating temperature was varied. As the temperature was decreased from 20°C the volatile suspended solids began to increase. This is in agreement with work done by others (71)(73). The volatile suspended solids increased from about 50% of total suspended solids at 20°C to 60% at 0°C and continued to increase to 70% at 5°C (increased from 0°C). The increase in volatile matter was likely due to cellular activity since the external addition of volatile solids via feed in fact decreased at the point

where the mixed liquor volatile percentage was at its greatest.

Although tests were not specifically conducted to prove the following, it was considered likely that as the temperature decreased the volatile solids increase was due to cellular food storage rather than cellular replication. A further explanation of this phenomenon is that although the bacterial metabolism slows down which precludes the probability of increased production as the temperature decreases, cells still maintain the capability of adsorbing or absorbing the substrate. This might be considered analagous to the storage of "fat" in higher life forms. Considering the effects of temperature on this food storage, the increase of temperature and hence metabolic rate would cause this "excess" food (fat) to be consumed for energy at higher temperatures.

6.1.3.3. Effluent Solids Test Results

The results of the effluent suspended and volatile suspended solids are shown in Table 7 and Figure 16. The average effluent suspended solids did not vary significantly with temperature. The values ranged from a low of 8 mg/l to a high of 24 mg/l with an average value of 15 mg/l. The effluent volatile suspended solids followed a similar trend.

The effluent solids data helps to confirm the hypothesis put forth in section 6.1.3.2. An expected low metabolism rate at low temperatures should be reflected in a poorer effluent, i.e. high BOD₅, COD, suspended solids, etc. The apparent low values for these parameters may be explained by the "food storage" luxury uptake phenomenon as a result of substrate adsorption.

6.1.4. Sludge Volume Index Results

The results of the sludge volume index at a given temperature are shown in Table 8 and Figure 17. The SVI remained below 100 until 0°C. Relatively poorer settling characteristics were exhibited by the mixed liquor when the temperature was again increased from 0°C. This can be explained again by the luxury uptake theory of Section 6.1.3.2. This "fat" microbial culture tended to be bulky which prevented rapid compact settling.

A noteworthy point is the relationship between the settled sludge volume and the SVI test. It is commonly accepted that a SVI test with a result of about 100 indicates a good settling sludge. This test parameter has serious field limitations in that a suspended solids test must be done to arrive at the SVI result. A "rule of thumb" in actual practice is that if a sample of aeration basin mixed liquor is allowed to settle quiescently for 30 minutes in

any container such as a quart glass jar, and if the settled sludge volume is between 30% and 70% of the total jar volume, the mixed liquor sludge exhibits good settling characteristics.

This "rule of thumb" was shown to be reasonable as is shown in Figure 26 where 30% and 70% of the total volume was equivalent to an SVI of 44 and 114, respectively.

6.1.5. Turbidity and pH Test Results

The turbidity and pH test results are shown in Table 9 and Figure 19. The pH increased as the temperature decreased to 0°C and then decreased as the temperature was increased to 10°C. The reason for the pH fluctuation from a low of 7.2 to high of 8.0 was attributed to the turbulence of the aeration basin. Excessive quantities of air, (approximately 8 l/min) were required to maintain a complete mix condition. This flow of air tended to strip the carbon dioxide produced by microbial respiration from solution. This tended to raise the pH. Although the turbulence was maintained throughout the test, the pH decreased as the temperature was increased from 0°C. This was attributed to a mixed liquor which was gelatinous and bulky and thus rendered the air stripping process less effective. The higher carbon dioxide concentration in solution depressed the pH.

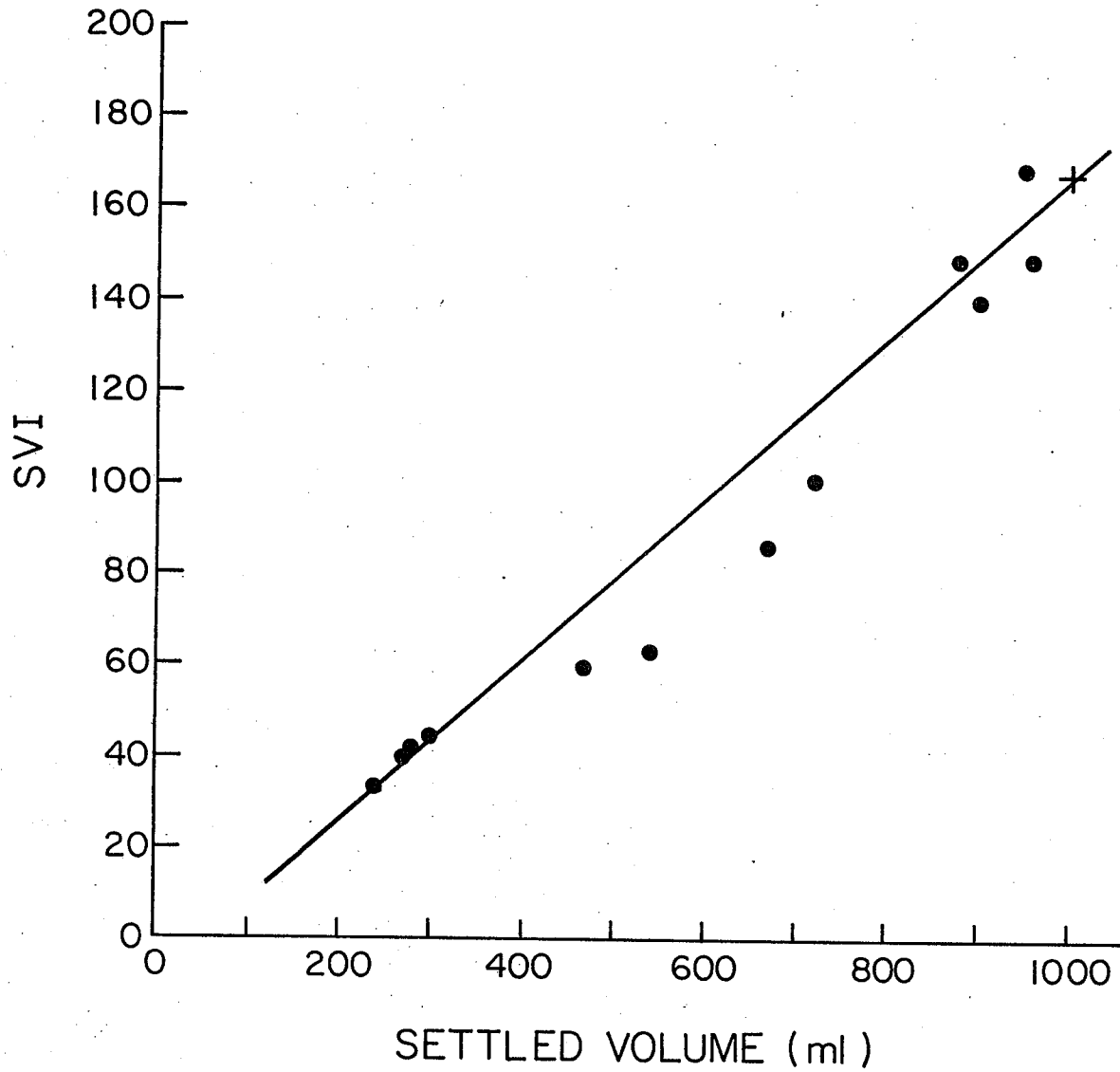


Fig.26. Relationship Between Sludge Volume After 1/2 Hour Settling in a 1000 ml Graduated Cylinder and SVI

The effluent turbidity was relatively low throughout the test ranging from a low of 4 J.T.U. to a high of 19 J.T.U. The temperature effects on turbidity were not significant. The results shown in Table 9 do not include the periods of sludge carry over. Carry over resulted in an extremely turbid effluent, but since this was not a steady state condition it was not considered in the turbidity calculations.

6.1.6. Oxygen Uptake Rate Test Results

The results of the oxygen uptake tests are shown in Table 10 and Figure 20. These results are an index of the relative metabolic activity of the mixed liquor at different temperatures. Although extensive testing was not done on this aspect of the investigation, it does show a general trend of decreasing metabolic activity, i.e. a decrease in oxygen utilization, with decreasing temperatures. Figure 20 shows the rapid decrease of oxygen utilization as the temperature decreased. This data suggested a power law relationship. A plot of the oxygen uptake rate data on log-log scale is shown in Figure 27. The straight line was drawn with an estimated best "fit". The data points lie reasonably near this line which can be described mathematically as

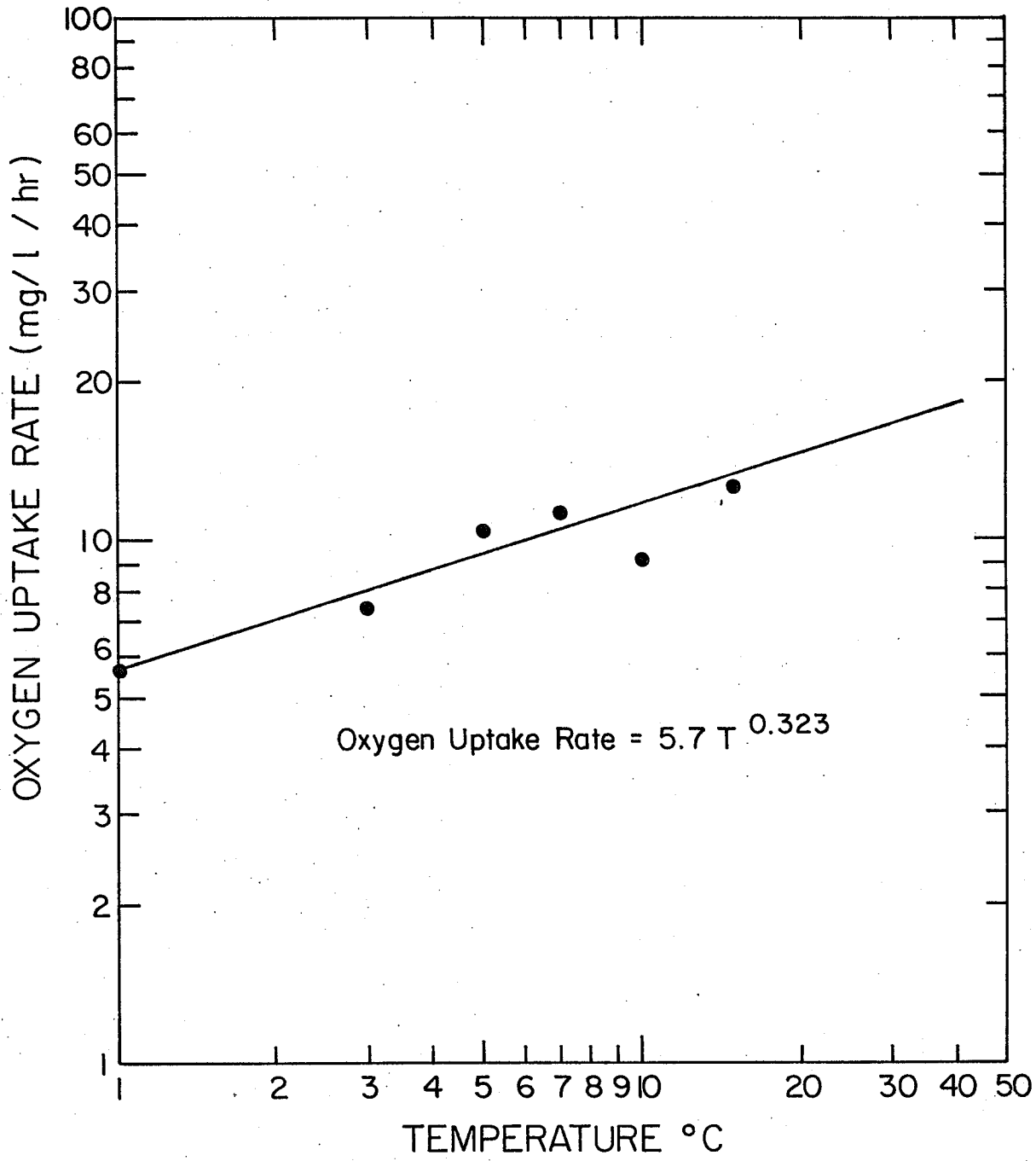


Fig. 27. Oxygen Uptake Rate vs Temperature

$$\text{Oxygen Uptake Rate} = 5.7T^{0.323}$$

where $T = ^\circ\text{C}$ and

oxygen uptake rate is in mg/l/hr.

6.1.7. Nitrogen Test Results

This study is one of the first attempts to identify nitrification behaviour in an extended aeration plant at temperatures down to 0°C . The nitrogen test results are shown in Table 11 and Figure 21. Tests were performed at temperatures increasing from 0°C to 10°C . The feed that was used had an average ammonia concentration of 22 mg/l as N and a nitrate concentration of 0 mg/l as N. At steady state conditions on October 14 at an operating temperature at 0°C the effluent had a $\text{NH}_3\text{-N}$ concentration of 20 mg/l and a $\text{NO}_3\text{-N}$ concentration of 4.4 mg/l. Nitrification is generally thought to be absent of temperatures below 5°C (2) (3). The long retention of the extended aeration process resulted in approximately 18% oxidation of the total feed ammonia nitrogen to nitrate.

On October 20 a malfunction of the environmental chamber resulted in an overnight temperature increase to 19°C . The effluent tested 24 hours later showed an ammonia concentration of 0 mg/l and a nitrate concentration of 24 mg/l. This indicated that within 24 hours of the temperature increase, the nitrifying bacteria population was capable of completely converting the ammonia to nitrate. This implies that, if a nitrifying microbial population is

present, continued low temperature operation will not inactivate them, but rather place them in a dormant stage to react immediately when exposed to a more favourable temperature.

The environmental chamber temperature was then returned to 0°C (within 15 minutes) and after basin flow-through, the effluent was tested. The results showed that it took 9 days to return the nitrifying bacteria to their dormant stage (18 to 20% nitrification of feed ammonia). This is significant in that nitrifying bacteria are quickly activated with temperature increases, but respond much more slowly when the temperature is decreased.

Increasing the temperature gradually from 0°C gradually increased the amount of ammonia that was oxidized.

This rate of ammonia nitrification with respect to temperature is shown in Figure 21.

In the nitrification process the conversion of ammonia to nitrate obviously means that an increase of nitrate should result in a decrease of ammonia, the total nitrogen remaining relatively constant. In this series of tests this was generally true when the temperature was decreased from 20°C to 0°C. When the temperature was then increased from 0°C to 7°C the nitrate concentration began to increase at an unusually high rate, e.g. on December 5 the nitrate concentration of the effluent was 41 mg/l as N

resulting from an ammonia feed on December 4 of 17 mg/l as N. This difference in total N (41 mg/l vs 17 mg/l) could not be accounted for by considering organic nitrogen in the feed since only a trace was detected at any time. The high effluent nitrate concentrations were measured until the termination of the experiments on December 16.

The following two possible explanations may be given to explain this phenomenon.

(1) The temperature increase increased the endogenous respiration rate whereby the lysing of microbial cell protein resulted in the release of ammonia which was rapidly oxidized to nitrate; or,

(2) The luxury uptake theory indicated that at low temperatures the microbial populations in extended aerations tended to take up substrate into reserve. This reserve has been shown by McKinney (20) to be primarily of a carbohydrate nature. This does not preclude the fact that nitrogenous material is also taken up which will ultimately be oxidized to nitrate as the temperature increases.

6.1.8. Total Phosphorus Test Results

The total phosphorus concentrations of the feed, effluent and mixed liquor are shown in Table 12 and Figure 22. Temperature effects on phosphorus removal efficiency are not readily apparent. An average removal efficiency of 50%

was noted, which is considered typical for the activated sludge process (2). The oxygen uptake rate discussed earlier indicated that at lower temperatures metabolic activity as measured by oxygen uptake decreased. This would imply that the phosphorus removal efficiency should decrease as the temperature decreased. This was not indicated in these experiments. It was noted however, that from 0°C to 1°C the total phosphorous concentration of the mixed liquor began to increase, while as the temperature increased to 3°C and higher, the mixed liquor total P began to decrease. This behaviour reinforces the luxury uptake theory for the apparent no-temperature effect on phosphorous removal efficiencies at low temperatures. To the writer's knowledge phosphorus removal efficiencies in extended aeration at temperatures down to 0°C have not been documented. The results from this study will aid in quantifying the removal at this nutrient for cold weather extended aeration applications.

6.2. BIOKINETIC INTERPRETATION RESULTS

6.2.1. Kinetic Growth and Substrate Utilization Rate Constant Results

The kinetic growth and substrate utilization constants using BOD₅ and COD as a measure of substrate strength are shown in Table 13.

6.2.1.1. Kinetic Growth Constants

The yield constants shown in Table 13 are expressed in terms of weight of oxygen equivalent cell mass per weight of BOD₅ utilized. In order to express the yield

constant as weight of volatile suspended solids per weight of BOD₅, it was necessary to multiply the former by 0.704 i.e. 1 pound of cells = 1.42 lbs oxygen equivalent. Based on BOD₅ results, the yield coefficient Y, and the decay coefficient k_d do not show an apparent temperature dependency. The yield coefficient varied from a low of 0.055 to a high of 1.00 $\frac{\text{wt O}_2\text{-Eq.}}{\text{wt BOD}_5}$. The decay coefficient varied from a low of 0.025 x 10⁻³/hr to a high of 4.57 x 10⁻³/hr.

The yield and decay coefficients did not show a specific trend with respect to substrate concentration. The average results of the yield and decay coefficients are shown below as Table 16 and Figure 28.

Temperature °C	Substrate (mg/l)		Y		k _d x 10 ³ /hr
	BOD ₅	COD	$\frac{\text{Wt O}_2\text{-Eq}}{\text{Wt Substrate}}$	$\frac{\text{Wt VSS}}{\text{Wt Substrate}}$	
20	470		0.42	0.30	1.64
		406	1.00	0.70	3.27
15	410		0.57	0.40	1.07
10	400		0.14	0.10	0.16
		159	1.00	0.70	0.69
7	340		0.74	0.52	1.15
5	450		0.52	0.37	0.99
3	255		0.11	0.08	0.07
		160	1.00	0.70	0.60
1	380		0.42	0.30	0.53
		268	0.25	0.18	0.19
0	545		0.75	0.53	2.18
		398	0.06	0.04	0.015

TABLE 16. Average Yield and Decay Coefficients for Extended Aeration.

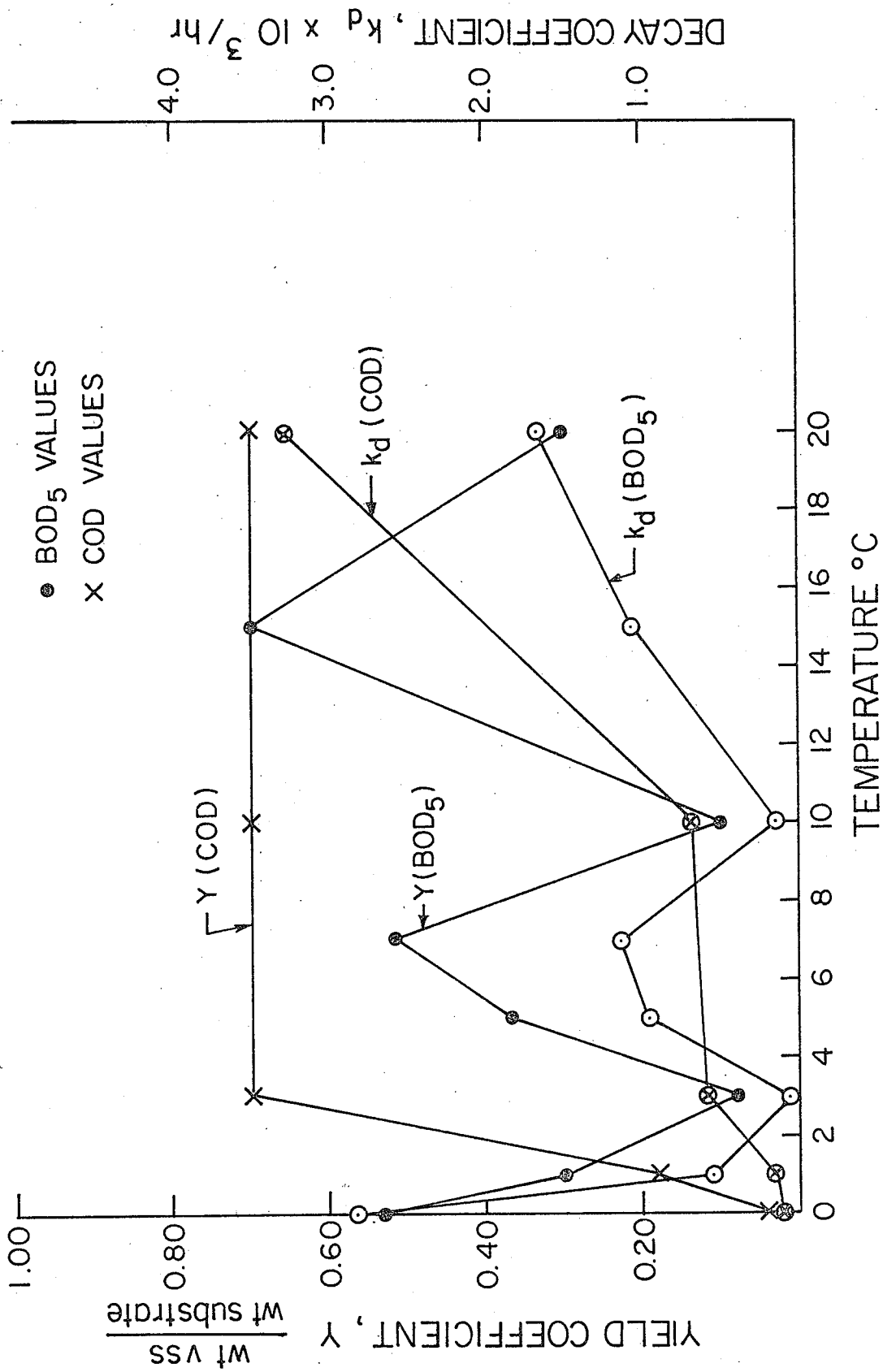


Fig. 28. Average Yield and Decay Coefficients vs Temperature

The BOD₅ results of Table 16 and Figure 28 show a random pattern of growth constants with respect to temperature. The growth yield coefficient Y, at 0°C was 0.53 while at 20°C it was 0.30 $\frac{\text{wt VSS}}{\text{wt. BOD}_5}$. These tests show no apparent temperature effects on the growth constants. Although the COD results show an apparent trend with temperature in Figure 28, the low r-values of the data at 0°C and 1°C, and the greater than 1.0 yield coefficient, based on oxygen equivalent, as shown in Table 13, precludes a definitive statement about the temperature effects on these biokinetic parameters.

The yield coefficient of 1.00 was obtained by forcing the straight line to intersect the y-axis at 1.0. This was done for cases where the y-intercept was less than 1.0 or negative. An example is shown as Figure 31. It is physically impossible to have a Y value greater than one since this would imply that the food conversion efficiency was greater than 100% which is impossible. Similarly a negative yield constant is not possible. The reason that Y values greater than one or negative values were obtained was due to the lack of data near the origin. Data near the origin could have been obtained by decreasing the basin retention time, decreasing the mixed liquor concentration or allowing the effluent suspended solids to increase.

The average yield coefficient was found to be 0.33 $\frac{\text{wt. VSS}}{\text{wt BOD}_5}$ and 0.46 $\frac{\text{wt VSS}}{\text{wt COD}}$. The average decay coefficient

was found to be 0.97×10^{-3} /hr based on BOD_5 and 0.98×10^{-3} /hr based on COD. Middlebrooks and Garland (47) found that actual field operating extended aeration plants at 20°C had a yield coefficient of $0.54 \frac{\text{lb VSS}}{\text{lb BOD}}$ and a decay rate of 0.58×10^{-3} /hr.

The apparent absence of temperature effects on the growth and substrate utilization constants at low temperatures is contrary to McKinney (20) who stated that,

"it appears that between 5°C and 35°C the rate of biological reactions double with each 10°C increase", with the result that the rate of synthesis, i.e. growth, is temperature dependent and that the endogenous respiration constant i.e. decay is "definitely temperature dependent".

Young (81), describing a design model for complete-mix activated sludge, makes no mention of temperature effects on the yield coefficient, but states that,

"the decay coefficient would be expected to be temperature dependent... since decay represents the respiration of bacteria, it would not be unreasonable to expect the same temperature effect as in the BOD test where much of the oxygen uptake is due to bacterial respiration ..."

Young claims that the substrate utilization constants are only of "academic interest" when dealing with aerobic systems.

Eckenfelder (41) has stated that temperatures below 95°F have no effect on biological waste treatment performance as long as the food to microorganism ratio was maintained below 0.5. This is a rather general statement, since no mention was made of the temperature effects on the growth or substrate utilization constants.

Metcalf and Eddy (2) have mentioned the lack of information with regard to the availability of growth and substrate utilization constants. The experiments for this investigation were specifically set up to investigate in detail the effects that temperature has on the growth and substrate utilization constants for extended aeration. This work is the first attempt to the writer's knowledge of a determination of biokinetic constants for extended aeration at low temperatures.

6.2.1.2. Substrate Utilization Constants

The substrate utilization constants are defined by K_s and μ_{max} . The average values are shown in Table 17 and Figure 29. This data shows that K_s , i.e. the substrate concentration when the rate of substrate utilization is $\frac{1}{2}$ that of the maximum, is zero, or approaches zero. This implied that a low substrate concentration was able to activate a microbial culture in this extended aeration study.

Temperature °C	Substrate BOD ₅	mg/l COD	K _S mg/l	μ _{max} × 10 ³ /hr
20	470		27	2.25
		406	0	3.37
15	410		6	1.43
10	400		0	0.49
		159	9	0.95
7	340		0	1.21
5	450		0	1.08
3	255		0	0.14
		160	0	0.67
1	380		1	0.70
		268	1	0.36
0	545		0	2.73
		398	40	0.19

TABLE 17. Average Substrate Utilization Coefficients for Extended Aeration.

The μ_{\max} values for BOD₅ plotted in Figure 29 do not in fact show significant variations due to temperature changes. The plot in Figure 29 appears to show a trend for the COD values. This trend, is not significant since the expanded scale of μ_{\max} in Figure 29 tends to exaggerate the variation with temperature. The correlation of the data is shown in Table 13. The correlation r , ranged from 0.28 to 0.86. In most cases a correlation could not be found because the straight line developed was parallel to the x-axis i.e. x and y were independent of each other. An example is shown in Figure 32. When a correlation could

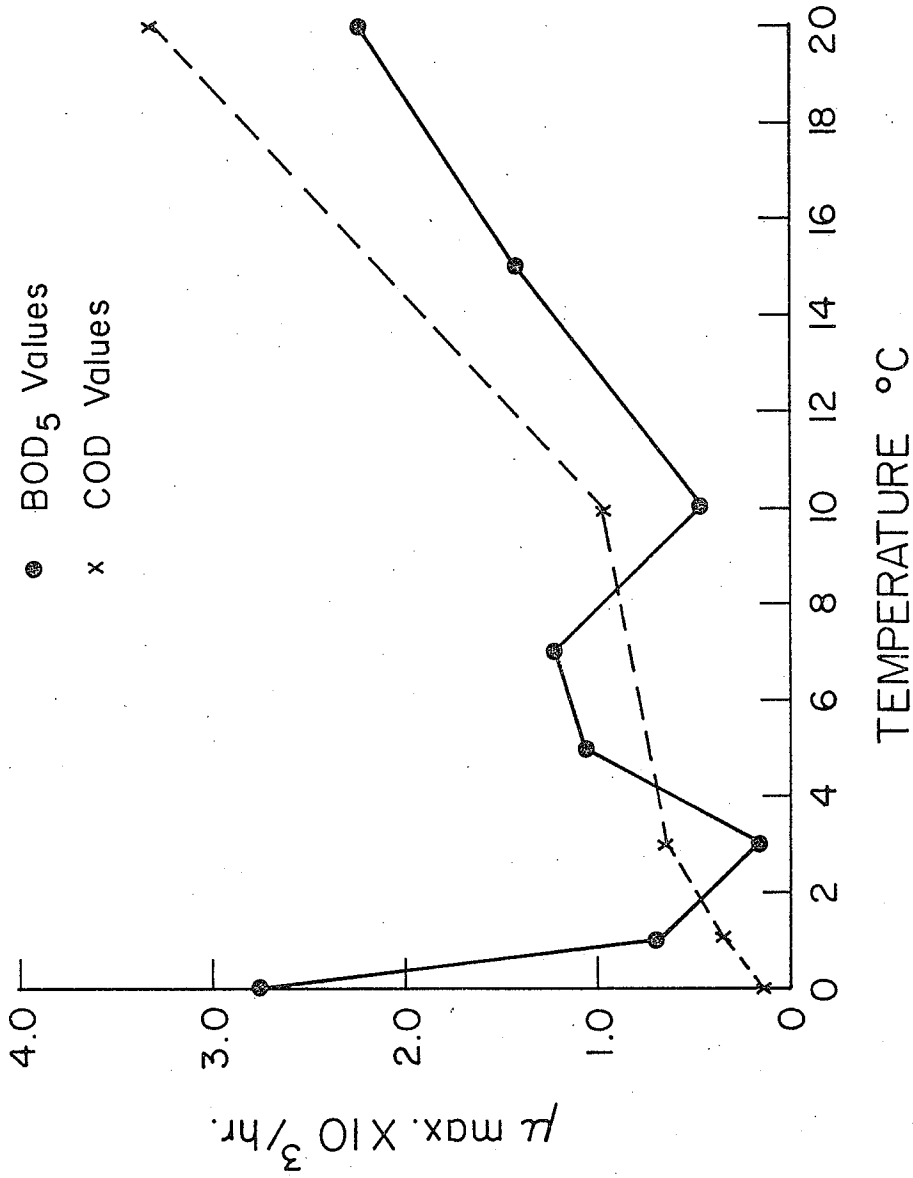


Fig.29. Average Maximum Substrate Utilization Rate Constant Versus Temperature.

be found, the level of significance of this data is shown in Table 13.

The overall average K_s value based on BOD_5 was 6 mg/l and 10 mg/l based on COD. The overall average μ_{max} based on BOD_5 was 1.27×10^{-3} /hr and 1.11×10^{-3} /hr based on COD. Middlebrooks and Garland (47) found that actual field operating extended aeration plants at $20^\circ C$ had a K_s value of 12.4 mg/l and a maximum growth rate, $\mu_{max} = 1.59 \times 10^{-3}$ /hr.

6.2.2. Results of Temperature Effects Upon BOD_5 Removal

The results of temperature effects on overall BOD_5 removal for extended aeration are shown in Table 14 and Figures 23 and 24. From Figure 23 it can be seen that generally a steeper line, i.e. a high rate of removal is noted at the higher temperatures. Exceptions to this are the $10^\circ C$ and $1^\circ C$ lines which show a higher slope than $15^\circ C$ and $3^\circ C$ lines respectively.

The slopes or BOD removal rate constants K_a , are plotted versus temperature on Figure 24. The plotted line shows a zero order relationship with temperature. The regression coefficient of this straight line equation was 0.87 which indicated a good measure of "fit" of the data points to the line. A Student's t analysis indicated that the data was significant at the 1% level.

The straight line equation may be written as:

$$K_t = 0.000362T + .002,$$

where K_t = BOD removal rate at a given temperature; and,
 $T = ^\circ\text{C}.$

Exceptions to the first order rule have been put forth by Stoltenberg and Sobel (67) by stating that in their evaluation of the temperature effects on a polluted estuary ..

"the Van't Hoff-Arrhenius formulation was not used primarily because it apparently would not yield a direct continuous relationship between the reaction rate and temperature over a sufficiently wide range. In addition, estimates of θ (a factor which itself varies with temperature) would not have been comparable to those at previous investigators, since the method of estimating θ would not necessarily have been identical".

The zero order kinetics of BOD_5 removal has been shown to apply to at least one other complete mix activated sludge treatment plant (82). Temperature dependency on activated sludge BOD removal efficiency has generally been described by the classical first order equation. The convincing zero order behaviour at low temperatures for this study is significant in that it definitely contradicts the commonly accepted first order temperature dependency.

6.2.3. Temperature Effects Upon BOD₅ Incubation

The fact that temperature does affect microbial activity was demonstrated by the BOD₅ results at different incubation temperatures. The $\frac{BOD_t}{BOD_{20}}$ ratios are shown in

Table 15 and Figure 25. The temperature effects were similar for the BOD's done on the feed as well as the effluent. A zero-order relationship was noted. The effect of incubation temperature on the five day BOD was found to follow the relationship

$$BOD_T = BOD_{20} (0.04T + 0.19)$$

where $BOD_T = BOD_5$ at a given temperature and

T is in °C.

This equation may be compared to the variation with temperature of the ultimate BOD given by Babbitt and Bauman (80) as

$$(L_a)_T = (L_a)_{20} (0.02T + 0.6)$$

where L_{aT} = ultimate demand at a given temperature and

T is in °C.

This zero-order relationship emphasizes the significance of not blindly accepting a first-order relationship for temperature effects on biological reactions.

7. CONCLUSIONS

The following are the conclusions of this temperature controlled laboratory extended aeration study in which the aeration basin dissolved oxygen concentration was at near saturation.

(1) The growth and substrate utilization constants calculated from basic Monod kinetics did not show any apparent temperature effects as the temperature was varied from 20°C to 0°C. From the data as presented the average results were as shown below in Table 18.

Substrate (mg/l)	Y wt VSS wt substrate	$k_d \times 10^3$ /hr	$\mu_{max} \times 10^3$ /hr	K_s mg/l
BOD ₅	0.33	0.97	1.27	6
COD	0.46	0.98	1.11	10

TABLE 18. Summary of Growth and Substrate Utilization Constants for Laboratory Extended Aeration Plant.

(2) The temperature effects on the overall BOD₅ removal rate using Eckenfelder's simplified substrate removal equation were found to follow a zero order relationship written as:

$$K_t = 3.62 \times 10^{-4}T + 2 \times 10^{-3},$$

where K_t = BOD₅ removal rate (day⁻¹); and,

T = temperature °C (0°C to 20°C).

(3) The effects of temperature on BOD₅ incubation was found to follow a zero-order relationship shown below as:

$$\text{BOD}_{5T} = \text{BOD}_5 (0.04T + 0.19),$$

where BOD_{5T} is the 5-day BOD at a given temperature between 0°C and 20°C; and,

T is the temperature in °C.

(4) The mixed liquor suspended solids concentration that could be maintained in the aeration basin before sludge carry over occurred was found to be 8255 mg/l or less. This contradicts the commonly accepted maximum MLSS concentration of 6000 mg/l given for proper extended aeration operation (2)(11). Mixed liquor volatile suspended solids were found to increase as the temperature decreased and was considered to be caused by substrate storage rather than cell replication.

Effluent suspended and volatile suspended solids did not vary significantly with changes in temperature.

(5) Nitrification was found to occur at all temperatures including 0°C. At 0°C 18% of the feed NH₃-N concentration was oxidized to NO₃-N after 35 hours retention time. Temperature increases from 0°C to 20°C within several hours resulted in 100% ammonia oxidation to nitrate nitrogen.

Temperature decreases from 20°C to 0°C within several hours required 9 days to return the nitrifying bacterial population to its steady state 0°C performance (about 18% ammonia oxidation).

Gradual temperature increases from 0°C to 20°C resulted in a gradual increase in percentage of ammonia oxidized.

Unexpected high concentrations of nitrate nitrogen were found to occur when the temperature was gradually increased from 0°C to 10°C. This was considered to be due to the utilization of cellular nitrogenous substrate storage stimulated by increased temperatures.

(6) The average total phosphorous removal was found to be 50% and was found to be unaffected by changes in operating temperature between 20°C and 0°C. The mixed liquor total phosphorus concentration was found to decrease as the temperature increased from 0°C to 10°C.

(7) The microbial oxygen uptake rate was found to vary with temperature according to the following equation:

$$\text{Oxygen Uptake Rate} = 5.7T^{0.323}$$

where oxygen uptake rate is in mg/l/hr, and

T is the temperature in 0°C.

This decrease in oxygen utilization at low temperatures is significant in that it shows that while microbial activity decreases with temperature the performance parameters, such as BOD₅ removal, remained at high efficiencies.

(8) The average BOD₅ removal efficiency was found to be 94%. A 5% decrease in BOD₅ removal efficiency (89%) was found to occur at 0°C.

(9) The COD removal efficiency was not significantly affected by varying the temperature between 20°C and 0°C. The highest COD removal efficiency (91%) was found to occur at 0°C.

(10) The BOD₅/COD ratios for the feed and effluent were found to be 1.62 and 0.62 respectively. The use of COD as a measure of substrate strength for the design of biological aerobic waste treatment systems has been recommended by Jenkins et al (40). The inability of the COD test to measure the oxygen demand of ammonia limits its usefulness as a measure of substrate strength in biological waste treatment systems.

(11) The sludge volume index was found to remain below 100 with temperature decreasing to 0°C. With temperatures increasing from 0°C to 10°C the sludge volume index began to increase which resulted in poorer settling characteristics.

(12) The pH of the mixed liquor increased slightly from 7.7 to 8.0 as the temperature was decreased from 20°C to 0°C, and decreased to 7.2 as the temperature was increased from 0°C to 10°C.

(13) The effluent turbidity at steady state operation was unaffected by temperature and resulted in an overall average of 7 J.T.U.

8. FUTURE WORK

As a result of this investigation future work is suggested in the following area:

(1) Additional tests should be conducted at shorter retention times using the laboratory extended aeration plant to establish data points nearer the origin for the kinetic constants equations.

(2) Parameters other than volatile suspended solids should be investigated to determine the viable cell mass in an activated sludge plant. The kinetic constants at low temperatures should be determined using mixed liquor organic nitrogen, DNA or ATP as an indicator of active microbial growth.

(3) The apparent uptake and storage of substrate by microbial organisms should be more thoroughly investigated to determine the mechanism and composition of the apparent substrate "luxury uptake".

(4) The kinetic growth constants at low temperatures should be investigated in the laboratory using modifications of the activated sludge process other than extended aeration.

(5) A continuing test program over a period of several years should be conducted on operating field activated sludge units to verify simulated laboratory studies. Field and laboratory results should be analyzed for correlation

and significance so that kinetic behaviour can be applied to rational design for low temperature operation.

(6) Investigations into final clarifier operation at low temperatures with special emphasis on sludge return should be made for the activated sludge process.

(7) Minimum mixing and oxygen requirements at low temperatures should be determined as part of an investigation into heat loss from exposed activated sludge aeration basins.

(8) An investigation should be made to determine kinetics of nitrification in activated sludge systems at low temperatures at various retention times.

ACKNOWLEDGMENT

The author wishes to acknowledge with gratitude financial assistance that was made available for this project by the National Research Council via NRC Grant No. A8955.

The technical and moral support offered by my principal advisor Dr. A.B. Sparling is sincerely appreciated.

The assistance of the technical staff of Civil Engineering and in particular Mr. Harry Brooks is gratefully acknowledged.

Thanks are extended to Mrs. Arleen Sinclair for typing the manuscript.

A special thanks is extended to my wife Ellie for her patience and understanding throughout the course of this study.

APPENDICES

APPENDIX 1

DESIGN CALCULATIONS FOR A LABORATORY
EXTENDED AERATION WASTE TREATMENT UNIT

PROBLEM:

Design an extended aeration activated sludge plant in accordance with the design criteria given in Ten State Standards given the following conditions:

- (a) Wastewater to consist of typical domestic wastes only;
- (b) Average BOD₅ of influent = 250 mg/l;
- (c) Average influent suspended solids = 200 mg/l; and,
- (d) Sewage flow rate = 5 gallons per day.

SOLUTION:

Design criteria for extended aeration as per Ten State Standards, p. 78 includes:

- (a) Aeration retention (based on design flow) = 24 hrs;
- (b) Aerator loading = 12.5 lbs BOD₅/1000 ft³;
- (c) Aerator loading = 10-25 lbs BOD₅/1000 ft³ (Metcalf & Eddy);
- (d) MLSS/lb BOD₅ = 10/1 to 20/1;
- (e) Clarifier retention = 4.0 hours; and,
- (f) Clarifier surface overflow rate = 300 gal/day/ft².

A. Consider Organic Loading

$$\begin{aligned} & 5 \text{ gallons sewage per day} \\ & = 5 \text{ gal} \times 4.54 \frac{\text{litres}}{\text{gal}} = 22.7 \text{ l/day} \\ & = 23 \text{ l/day.} \end{aligned}$$

$$\begin{aligned} & \text{Average BOD}_5 \text{ loading per day} \\ & = 23 \frac{\text{litre}}{\text{day}} \times 250 \frac{\text{mg BOD}_5}{\text{litre}} = 5.74 \times 10^3 \frac{\text{mg BOD}_5}{\text{day}} \end{aligned}$$

To calculate required aerator volume to handle the organic load:

$$\text{Design organic load} = 12.5 \frac{\text{lb BOD}_5}{1000 \text{ ft}^3}$$

$$1 \text{ lb} = 4.54 \times 10^5 \text{ mg}$$

$$\text{Therefore } 5.74 \times 10^3 \text{ mg BOD}_5 = \frac{5.74 \times 10^3}{4.54 \times 10^5} =$$

$$1.27 \times 10^{-2} \frac{\text{lbs BOD}_5}{\text{day}}$$

$$\text{Design loading} = 1.25 \times 10^{-2} \frac{\text{lb BOD}_5}{\text{ft}^3}$$

$$\text{Therefore require } \frac{1.27 \times 10^{-2}}{1.25 \times 10^{-2}} = 1.01 \text{ ft}^3$$

say 1.0 ft³ of aerator volume.

$$1.0 \text{ ft}^3 = 28.32 \text{ l.}$$

Therefore for organic load require aeration basin size = 28.32 litres.

B. Consider Hydraulic Loading

Ten State Standards requires 24 hour retention.

Therefore, require 23 litres aeration basin volume to meet hydraulic loading requirements.

C. Consider Clarifier Requirements

Design clarifier retention = 4.0 hrs.

Therefore at design flow require $\frac{23}{6.0} = 3.8$ litre capacity.

Design O/F rate = 300 gal/ft²/day require surface area =

$$\frac{5}{300} = .0167 \text{ ft}^2 = 2.4 \text{ in}^2 \text{ (not practical).}$$

Therefore at 5 gallons per day total laboratory unit must be 26.8 litres for hydraulic considerations or 32.1 l for organic considerations.

Since the organic loading governs designs volume for unit should be about 32 litres.

D. Extended Aeration Unit Sizing

Practicalities of the laboratory continuous flow feed system is that a maximum of 19 litres per day can be fed.

Thus organic loading becomes

$$1.27 \times 10^{-2} \times \frac{19}{23} = 1.05 \times 10^{-2} \frac{\text{lb BOD}_5}{\text{day}}$$

Required aerator design volume

$$= \frac{1.05 \times 10^{-2}}{1.25 \times 10^{-2}} = 0.84 \text{ ft}^3$$

which is about 24 litres.

Hydraulic retention time:

$$\text{Clarifier capacity} = \frac{19}{6} = 3.2 \text{ litres.}$$

To meet the required design conditions construct a reactor with the following dimensions.

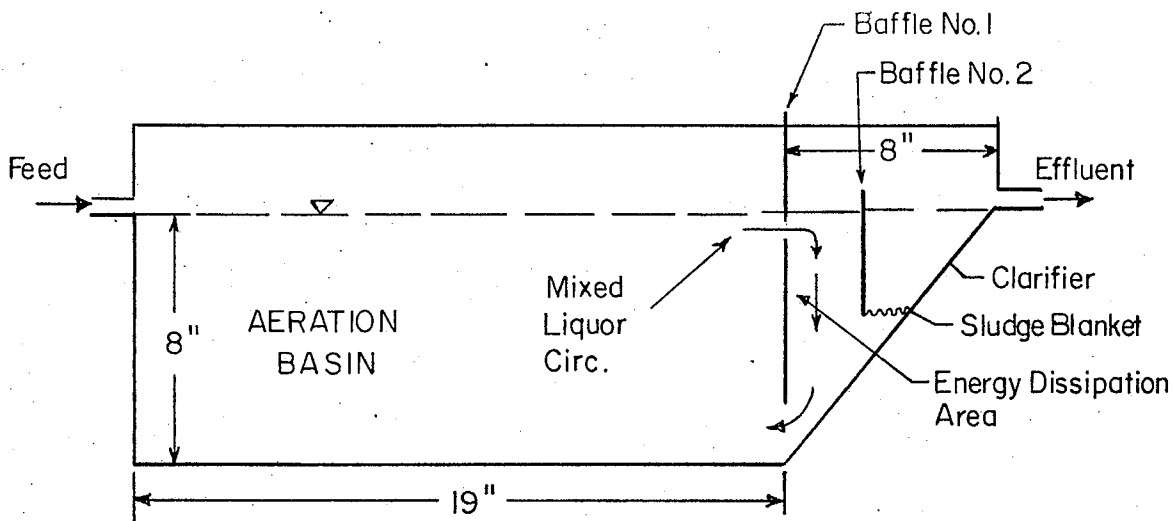


Fig. 30 Laboratory Extended Aeration Basin Dimensions.

Actual Active Aeration Volume

= 21.8 litres

Actual Clarifier Volumes

= 3.2 litres

Thus the laboratory model as constructed meets the design criteria entirely for hydraulic loading. The organic loading is somewhat less than "design" except that according to Metcalf and Eddy (2) the allowable organic loading range lies somewhere between 10 and 25 lbs BOD₅/1000 ft³ of aeration volume. Therefore sizing is acceptable.

APPENDIX 2

EXAMPLE CALCULATIONS OF

- (a) Growth yield constant, Y;
- (b) Endogenous decay rate constant, k_d
- (c) Maximum substrate utilization rate, μ_{max} ;
- (d) Half-velocity coefficient, K_s ; and,
- (e) Correlation coefficient, r and Student's t level of significance.

From Section 3.2.4. it was shown that the following straight-line equations could be used to determine the growth and substrate utilization constants for the steady-state extended aeration process.

$$\frac{(k_d)}{Y} b'\theta + \frac{1}{Y} = b' \frac{(S_0 - S_1)}{X_1} \dots\dots\dots (1)$$

where: $\frac{k_d}{Y}$ = slope; and,
 $\frac{1}{Y}$ = intercept.

$$\frac{b'\theta}{1+b'\theta k_d} = \frac{(K_s)}{\mu_{max}} \frac{(1)}{S_1} + \frac{1}{\mu_{max}} \dots\dots\dots (2)$$

where: $\frac{K_s}{\mu_{max}}$ = slope; and,
 $\frac{1}{\mu_{max}}$ = intercept.

From inspection of these equations it is obvious that the decay constant k_d , must be determined first, since k_d is used in equation (2).

The kinetic growth and substrate utilization constants were determined using the data where the BOD_5 of the feed, S_o was 0-99 mg/l, 100-199 mg/l, 200-299 mg/l etc.

Consider the calculations of a series of tests conducted at 0°C. The BOD_5 (S_o) range was 300 to 399 mg/l. The BOD_5 for the feed and effluent and the VSS for the mixed liquor and effluent are shown below in Table 19.

Date	Feed BOD_5 (S_o) mg/l	Effluent BOD_5 (S_1) mg/l	MLVSS		EffvVSS	
			(X_1) * O_2 -Eq mg/l	O_2 -Eq	(X_1) * O_2 -Eq mg/l	O_2 -Eq
Sept. 25	300	45	5150	7310	15	21.3
Oct. 6	380	29	4330	6160	12	17.1
7	390	73	3970	5640	14	19.9
13	340	128	4020	5710	14	19.9
15	380	26	3780	5360	12	17.1
22	320	16	3710	5260	6	8.5
24	300	54	4280	6080	13	18.5
25	370	92	3600	5110	12	17.1
29	370	23	4430	6300	12	17.1
30	390	42	4140	5890	9	12.8
31	370	53	3700	5260	8	11.4
Nov. 8	310	75	4630	6570	17	24.2
11	320	83	4480	6370	15	21.3
14	300	36	4620	6560	16	22.7
15	310	43	4360	6200	10	14.2

* Oxygen equivalent = MLVSS x 1.42

TABLE 19. BOD and Volatile Suspended Solids Data for Kinetic Constants.

The parameters required for plotting the kinetic growth equation are shown below in Table 20.

Date	b' (X ₁ /X ₂)	θ (hrs) (V/F)	b'θx10 ³ (hrs)	X mg/l	S ₀ -S ₁ (mg/l)	b'(S ₀ -S ₁) X
Sept. 25	344	28	9.63	7310	255	12.0
Oct. 6	361	34	12.27	6160	351	20.6
7	283	59	16.70	5640	317	15.9
13	287	27	7.75	5710	212	10.7
15	314	29	9.11	5360	354	20.7
22	619	28	17.33	5260	304	35.8
24	328	28	9.18	6080	246	13.3
25	299	28	8.37	5110	278	16.3
29	368	28	10.30	6300	347	20.3
30	459	28	12.85	5890	348	27.1
31	462	28	12.94	5260	317	27.8
Nov. 8	272	28	7.62	6570	235	9.7
11	298	27	8.05	6370	237	11.1
14	289	28	8.09	6560	264	11.6
15	436	28	12.21	6200	267	18.8

TABLE 20. Kinetic Growth Equation Data.

The plotted growth equation (1) is shown in Figure 31. The straight line growth equation was plotted according the procedure of least squares where

$$\text{slope} = \frac{\Sigma XY - \frac{(\Sigma X)(\Sigma Y)}{N}}{\Sigma X^2 - \frac{(\Sigma X)^2}{N}} \dots\dots\dots (3)$$

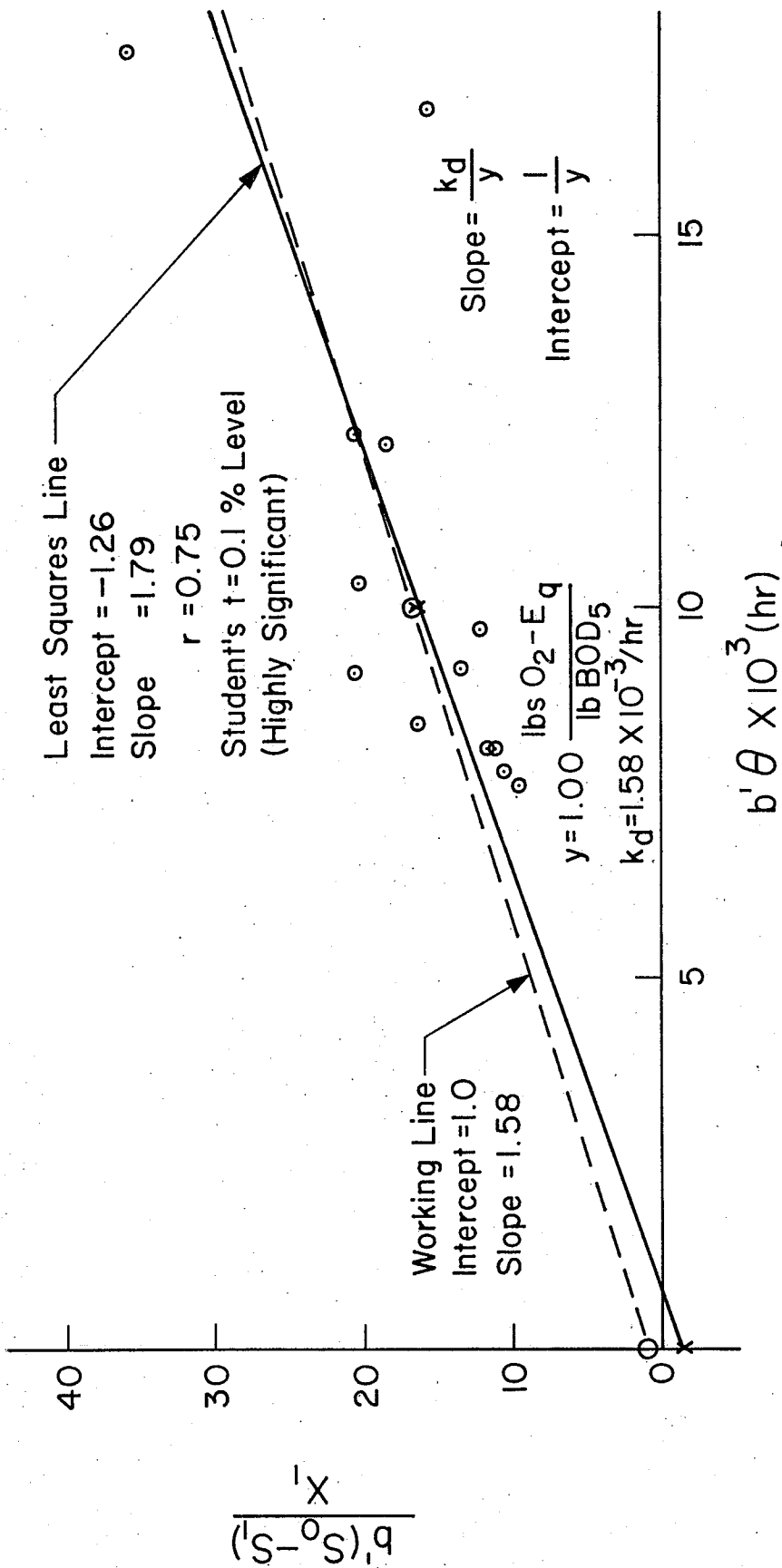


Fig. 31. Kinetic Growth Equation Curve for Average Feed BOD_5 (S_0) = 346 mg/L at 0°C

$$\text{Intercept} = \frac{(\Sigma x)(\Sigma xy) - (\Sigma y)(\Sigma x^2)}{(\Sigma x)^2 - N(\Sigma x^2)} \dots\dots\dots (4)$$

where N = number of data points;
 x = abscissa axis values; and,
 y = ordinate axis values.

The data necessary for calculation of the least squares slope and intercept are shown below in Table 21.

Date	$b' \theta x 10^3$ (x-axis)	$\frac{b'(S_0 - S_1)}{X_1}$ (y-axis)	xy	x^2	y^2
Sept. 25	9.63	12.0	115.56	92.74	144.00
Oct. 6	12.27	20.6	252.39	150.55	423.12
7	16.70	15.9	265.70	278.89	253.13
13	7.75	10.7	82.62	60.06	113.64
15	9.11	20.7	188.94	82.99	430.15
22	17.33	35.8	619.89	300.33	1279.49
24	9.18	13.3	121.82	84.27	176.09
25	8.37	16.3	136.18	70.06	264.71
29	10.30	20.3	208.78	106.09	410.87
30	12.85	27.1	348.49	165.12	735.49
31	12.94	27.8	360.25	167.44	775.07
Nov. 8	7.62	9.7	74.14	58.06	94.67
11	8.05	11.1	89.27	64.80	122.99
14	8.09	11.6	94.09	65.45	135.26
15	12.21	18.8	229.30	149.08	352.69

N = 15; $\Sigma x = 162.40$; $\Sigma y = 271.65$; $\Sigma xy = 3187.42$; $\Sigma x^2 = 1895.93$;
 $\Sigma y^2 = 5711.37$; Average $x = \bar{x} = 10.82$; Average $y = \bar{y} = 18.11$

TABLE 21. Data for Least Squares Analysis of Growth Constants Curve.

Substituting the values of Table 21 into equation (3) and (4) gives

$$\text{slope} = 1.79; \text{ and,}$$

$$\text{intercept} = -1.26.$$

This defines the straight line shown as a solid line in Figure 31.

From equation (1):

$$\text{Intercept} = \frac{1}{\bar{y}} = -1.26 = \frac{1}{\bar{y}}; \text{ and,}$$

$$\text{slope} = \frac{k_d}{\bar{y}}$$

A negative intercept suggests a "negative" yield which is impossible, as is a yield coefficient greater than one (47). The growth curve, therefore "approaches" an intercept of one and the growth curve is plotted using \bar{y} and \bar{x} to define the slope. This "working" or practical curve is shown as a dotted line in Figure 31.

$$\text{From the "working" line: } \bar{y} = \frac{1}{\bar{y}} = 1; \text{ and,}$$

$$k_d = 1 \times \text{slope} = 1.58 \times 10^{-3} / \text{hr.}$$

The calculations of the substrate utilization curve given as equation (2) are based on the data from Tables 19 and 20 and are shown below in Table 22 and Figure 32.

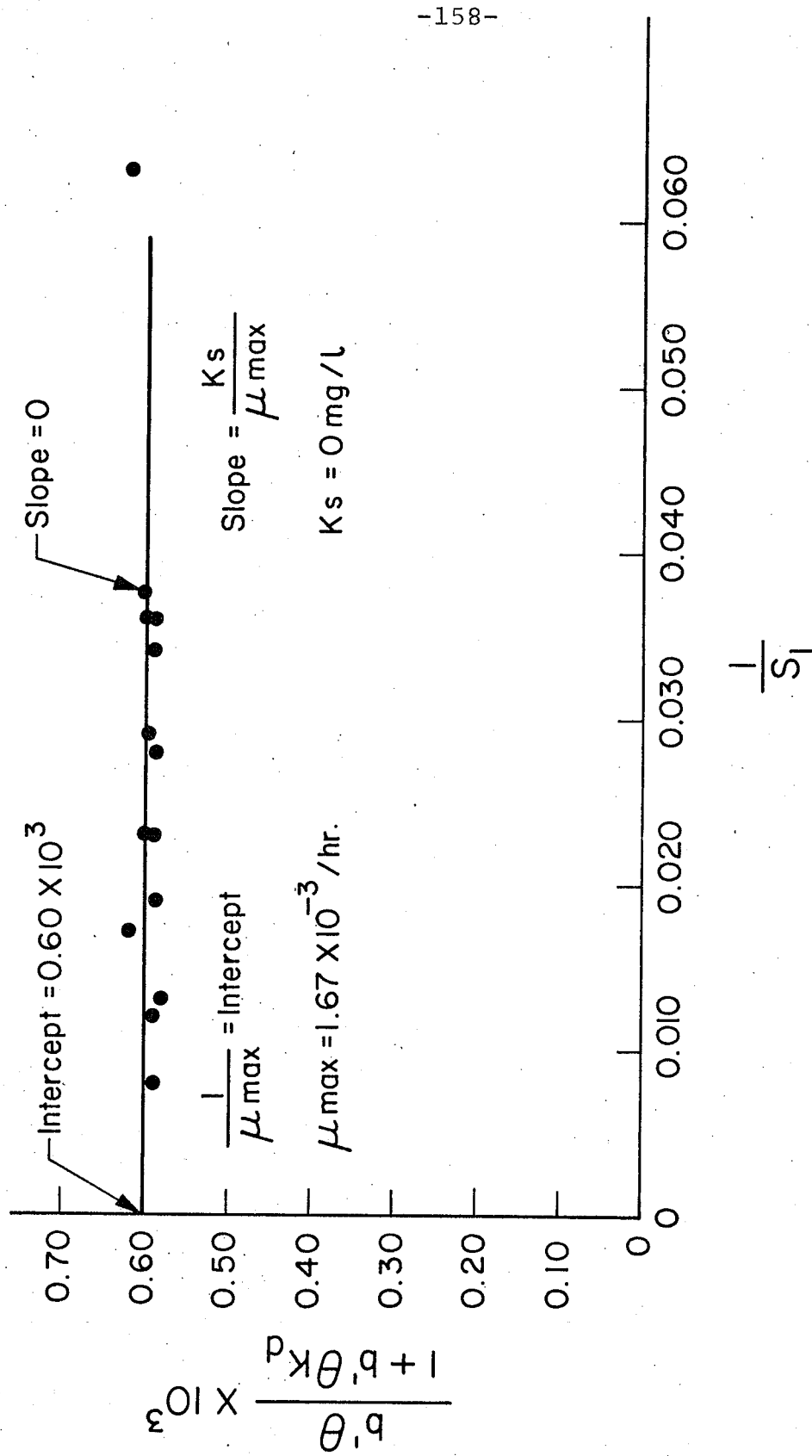


Fig. 32. Kinetic Substrate Utilization Curve for Average Feed BOD₅ (S_0) = 346 mg/L @ 0°C

Date	$b'\theta \times 10^3$ (hrs)	$k_d \times 10^3$ /hr	$b'\theta k_d$	$\frac{b'\theta}{1+b'\theta k_d} \times 10^3$	S_1	$\frac{1}{S_1}$
Sept. 25	9.63	1.58	15.22	0.59	45	0.022
Oct. 6	12.27	1.58	19.39	0.60	34	0.029
7	16.70	1.58	26.39	0.61	59	0.017
13	7.75	1.58	12.25	0.59	128	0.008
15	9.11	1.58	14.39	0.59	29	0.034
22	17.33	1.58	27.38	0.61	16	0.063
24	9.18	1.58	14.50	0.59	54	0.019
25	8.37	1.58	13.22	0.59	28	0.036
29	10.30	1.58	16.27	0.60	28	0.036
30	12.85	1.58	20.30	0.60	28	0.036
31	12.94	1.58	20.44	0.60	28	0.036
Nov. 8	7.62	1.58	12.04	0.58	75	0.013
11	8.05	1.58	12.72	0.59	83	0.012
14	8.09	1.58	12.78	0.59	36	0.028
15	12.21	1.58	19.29	0.60	43	0.023

TABLE 22. Substrate Utilization Rate Equation Data.

From equation (2) the abscissa is $\frac{1}{S_1}$ and the ordinate is $\frac{b'\theta \times 10^3}{1+b'\theta k_d}$ and is shown in Figure 32.

It becomes apparent that the "y-axis" values are essentially identical and independent of the "x-axis" i.e. parallel to the "x-axis". This precludes the use of a least squares procedure for establishing the "best-fit" straight line.

The intercept therefore is an average of the "y-axis" values equal to 0.600 and the slope = 0.

From equation 2

$$\begin{aligned} \mu_{\max} &= \frac{1}{\text{intercept}} = \frac{1}{.600 \times 10^3} = 1.67 \times 10^{-3} / \text{hr}; \text{ and,} \\ K_s &= \mu_{\max} \cdot \text{slope}; \\ &= 0 \text{ mg/l.} \end{aligned}$$

The calculation of the correlation coefficient and the Student's t level of significance was calculated and evaluated according to the formulae given by Moroney (83) where

(a) Correlation coefficient, r

$$r = \frac{(\frac{1}{N} \sum xy) - \bar{x} \bar{y}}{\delta x \delta y}$$

$$\text{where } \sigma_x = \sqrt{(\frac{1}{N} \sum x^2) - (\bar{x})^2};$$

$$\sigma_y = \sqrt{(\frac{1}{N} \sum y^2) - (\bar{y})^2};$$

N = number of samples; and,

(b) Student's t

$$t = \frac{r \sqrt{N-2}}{\sqrt{1-r^2}}$$

For this example the information required for the use of these equations is given in Tables 21 and 22. Substitution into the above equations for the growth rate least squares straight line results in

$r = 0.75$; and,

$t = 4.09$ which indicates a highly significant
group of samples (approximately 0.1% level).

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