THE ROLE OF SPECIFIC SUBSTRATES IN

EXCESS BIOLOGICAL PHOSPHORUS REMOVAL

by

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ABSTRACT

The principle objectives of this research were to investigate the role of specific carbonaceous substrates in the excess biological phosphorus (P) removal mechanism and to optimize the design of the nutrient removal activated sludge process so as to maximize the availability of these substrates to the micro-organisms involved in the P removal mechanism. The experimental work reported in this thesis was divided into the following four parts: 1) A series of laboratory-scale anaerobic batch tests which were designed to simulate the conditions present in the anaerobic zone of the process. The objectives of the batch tests were to determine which substrates were most effective in inducing anaerobic P release, the fate of the substrate and its role in the P release mechanism, and to quantify the negative effect of the presence of nitrate in the anaerobic zone on the P release mechanism. 2) A series of pilot-scale experiments in which sodium acetate was added to a simplified nutrient removal activated sludge process with the view to determining what concentration of added substrate is required to reliably induce excess biological P removal in the process. 3) A pilot-scale primary sludge fermentation study in which the objective was to determine the nature of, and the quantity of simple carbonaceous substrates that can be produced on-site at an activated sludge treatment plant, and the optimal fermenter operating conditions for such production. 4) A series of pilot-scale experiments in which primary sludge fermentation was incorporated into the design of a simplified nutrient removal activated sludge process

and the UCT process in order to gauge to what extent the P removal characteristics of these processes can be enhanced by such modification.

Results of the batch tests show that the simpler short-chain volatile fatty acids (VFA's) acetate and propionate are the most effective in inducing anaerobic P release in activated sludge. Batch tests in which a range of sodium acetate concentrations was fed into the flasks showed that P release and substrate utilization are integral parts of the same exchange phenomenon with a molar exchange ratio of 1.76 moles of acetate (as HAc) utilized per mole of P released by the micro-organisms. With regard to the detrimental effect of nitrate on the P release mechanism, it appears as if the available substrate required for the excess biological P removal mechanism is utilized in the denitrification reaction at a rate of 3.6 mg COD per mg NO_3-N and is thus rendered unavailable for the anaerobic P release mechanism. The addition of 86 mg/L (as COD) of sodium acetate to a simplified nutrient removal process treating raw sewage resulted in excess biological P removal. However, such removal was achieved by the addition of only 39 mg/L (as COD) when the substrate was added to an unaerated zone that received zero influent nitrate, confirming that the substrates required by the excess biological P removal mechanism are utilized in the denitrification reaction, and the importance of adding any additional substrate into a nitrate-free zone. Operation of the pilot-scale primary sludge fermenter showed that acetate and propionate, the two most important substrates in the excess

biological P removal mechanism, are also the principle products of primary sludge fermentation, making up more than 95% of the total short-chain VFA production. Optimum VFA yields of 0.09 mg of VFA (as HAc) per mg of primary sludge (as COD) were achieved at fermenter sludge ages in the 3.5-5.0 day range. Incorporation of primary sludge fermentation into the design of the simplified nutrient removal process resulted in an improvement of more than 100% in the P removal characteristics of the process. The same modification to a UCT process that was previously exhibiting some degree of excess biological P removal resulted in a further 50% improvement in the P removal characteristics. A proposal for the future design and operation of a primary sludge fermenter for the enhanced P removal activated sludge process that facilitates independent fermenter and process hydraulic and solids detention time control is also outlined.

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ABBREVIATIONS AND TERMINOLOGY

Abb.	Full Name	Units
COD	Chemical Oxygen Demand	mg COD/L
MLSS	Mixed Liquor Suspended Solids	mg/L
ORP	Oxidation Reduction Potential	mV (ref: Ag-AgCl)
SVI	Sludge Volume Index	%/%
RBS	Readily Biodegradable Substrate	mg COD/L
TKN	Total Kjeldahl Nitrogen	mg N/L
VFA	Volatile Fatty Acid	mg HAc/L
ΔP	System Phosphorus Removal	mg P/L
ΔCOD	System COD Removal	mg COD/L

The term "anaerobic" is used somewhat differently in
environmental engineering than in microbiology. Microbiologists
refer to an anaerobic state as being one in which dissolved
oxygen is absent, but various forms of combined oxygen (e.g.
nitrate) may or may not be present. In environmental engineering
the term anaerobic implies the absence of all forms of
bio-available dissolved oxygen, and the term "anoxic" is used to
define a state in which dissolved oxygen is absent, but combined
oxygen is present. In this regard, the terminology adhered to in
this thesis is that used in environmental engineering.

CHAPTER ONE

INTRODUCTION

The removal of phosphorus (P) from domestic and industrial
wastewaters has become increasingly important in recent years,
particularly in the case of process effluent streams being
discharged to inland waters. Phosphorus removal has been
recommended even in cases where limnological studies have shown
nitrogen (N) to be the primary, and P the secondary growth
limiting nutrient in surface water systems. The principle reason
for this is in lakes where P discharge is excessive but N is
limiting, conditions are often ideal for the development of large
populations of certain species of blue-green algae and other
N-fixing organisms. Nitrogen fixation is the process by which
these organisms are able to "fix" atmospheric N, i.e. utilize N
for basic metabolic purposes and incorporate it into their cell
mass. In this way atmospheric N enters the ecosystem in an
uncontrollable manner. The mass of P available to the organisms
of a given ecosystem, however, is solely a function of the mass
of P entering the system via the point and diffuse sources.

The discharge of wastewaters after widely varying degrees of
treatment often represents the major P discharge contribution by
point sources. For this reason, much attention has been focused
on the removal of P in processes treating domestic and industrial
wastewaters. This need has given rise to two basic removal
strategies - chemical and biological P removal.

Chemical P removal is achieved by the addition of precipitating chemicals such as calcium (Ca), aluminum (Al) and iron (Fe) salts. These chemicals are either added at the primary stage, directly into the process or as a tertiary stage of treatment. Chemical treatment, although generally successful in terms of achieving low effluent P concentrations, suffers from a number of drawbacks. For example, it creates a significantly larger solids handling problem in that in many cases large amounts of chemical sludge are produced that require disposal. The major disadvantage of chemical removal, however, is the cost of the precipitating chemicals. Rising chemical costs, coupled with the fact that chemical requirements tend to increase dramatically for P removals to concentrations below about 1.5 mg/L, have made the cost of chemical P removal prohibitive for many communities.

The problems associated with chemical P removal, as mentioned above, motivated research workers in many parts of the world to investigate biological P removal with special reference to excess biological P removal. Excess biological P removal is the phenomenon whereby a significant fraction of the organisms present in the activated sludge process remove a larger mass of P from the incoming wastewater than that which they require for basic metabolic purposes. This excess P is generally thought to be stored by the organisms in the form of long chains of inorganic polyphosphate known as volutin granules. It is generally assumed that P makes up approximately 1.5 percent of the dry weight of cells that have no stored polyphosphate (Hoffmann and Marais, 1977). Excess P removal is assumed to

be occurring when the measured dry weight percent P in the waste activated sludge from a process exceeds this amount. Excess P is removed from such a process by the sludge wasted each day. A comprehensive history of excess biological P removal and its effect on the development of the enhanced P removal activated sludge process is presented in Chapter Two.

Ever since 1959, when excess P removal was first reported by Srinath, Sastry and Pillai in India, there has been considerable debate as to whether the phenomenon is a purely biological one or a bacterially mediated chemical precipitation. However, the vast majority of research has indicated that excess P removal in the activated sludge process is primarily biological in nature, although removal by chemical precipitation does occur to a lesser extent. Reasons cited for this are that the pH range present in most processes and the presence of large amounts of organic material are unfavourable for the rapid precipitation of calcium phosphate.

Excess P removal was reported in the mid 1960's in plug flow high rate activated sludge processes (Scalf et al., 1969) and later in the 1970's in single sludge nitrification-denitrification processes (Barnard, 1974, 1976). Furthermore, it was noted that in all cases where the phenomenon occurred, the sludge was subjected to anaerobic conditions of such an intensity that P release from the sludge into the supernatent occurred. Upon entering a zone where aerobic conditions existed, the released as well as excess concentrations of P were taken up,

resulting in system excess P removal. It was hypothesized that the excess biological P removal was a consequence of P release having taken place and that the release was the result of the sludge being subjected to a certain degree of anaerobic stress.

Research work was then focused on attempting to quantify the degree of stress required to reliably induce excess biological P removal. The measurement of oxidation-reduction potential (ORP), which is based on the ratio of the sums of the total oxidized and reduced species present in a given solution, was chosen to quantify the degree of anaerobiosis required. Although ORP measurement is relatively simple in pure solutions, it was thought to be unreliable in activated sludge mixed liquor. In addition, it was found that it was not possible to accurately define the requirements for excess biological P removal in terms of anaerobic stress alone. In the early 1980's, it was found that P release was more closely related to the concentration and nature of substrate available to the micro-organisms under anaerobic conditions, than to a degree of stress obtained in the anaerobic zone.

This shift in emphasis from anaerobic stress quantification to the nature of the substrate available to the micro-organisms under anaerobic conditions gave rise to the concept of specific substrate induced excess biological P removal. This thesis outlines the investigation of the use of various substrates in an attempt to determine which substrates are most effective in inducing P release and what concentrations of the substrates are

required. Evidence will be presented that clearly demonstrates short chain volatile fatty acids (VFA's) and their salt forms to be preferred substrates. The development of a simplified nitrification-denitrification process configuration, that requires a minimum addition of the preferred substrates in order to reliably induce excess biological P removal, is described. An additional part of this research involves the on-site production of short chain VFA's by the fermentation of primary sewage sludge to be used in the subsequent process. In this manner, a form of treatment was developed in which excess P removal is achieved in processes treating low organic strength wastewaters without the addition of either additional substrate or precipitating chemicals.

CHAPTER TWO

LITERATURE REVIEW

This research deals with two previously unrelated areas of wastewater treatment, excess biological phosphorus removal and primary sludge fermentation with the objective of maximizing volatile fatty acid production. A brief overview of the available literature covering these two areas will be presented in Sections 2.1 and 2.2, respectively.

2.1. Excess Biological P Removal

Comprehensive literature reviews of excess biological P removal have been presented by Siebritz et al. (1983), Comeau (1984) and others. As such, only a brief review of the principle research will be presented in order to place it in an historical perspective with recent research being described in greater detail.

Excess biological P removal was first reported by Srinath et al. (1959) in India, and by Alarcon (1961) who conducted aerobic batch tests on mixed liquor taken from activated sludge processes exhibiting excess P removal. In both cases the mixed liquor was combined with raw sewage and very rapid initial P uptake was observed. Their results are presented in Figs. 2.1 and 2.2, respectively. Furthermore, Alarcon (1961) noted that if aeration was allowed to continue for a number of hours, a release of P from the sludge back into the supernatant occurred. Srinath et al. noted that the degree of P uptake appeared to be related to
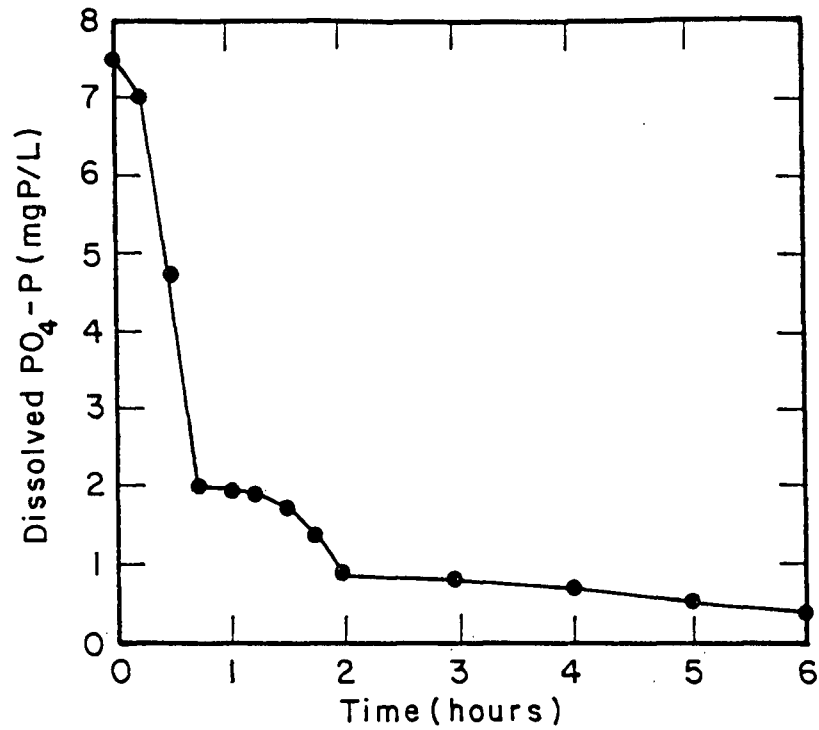
Fig. 2.1. Aerobic P uptake in a batch test using mixed liquor from a process exhibiting excess biological P removal. Data reported by Srinath et al. (1959).
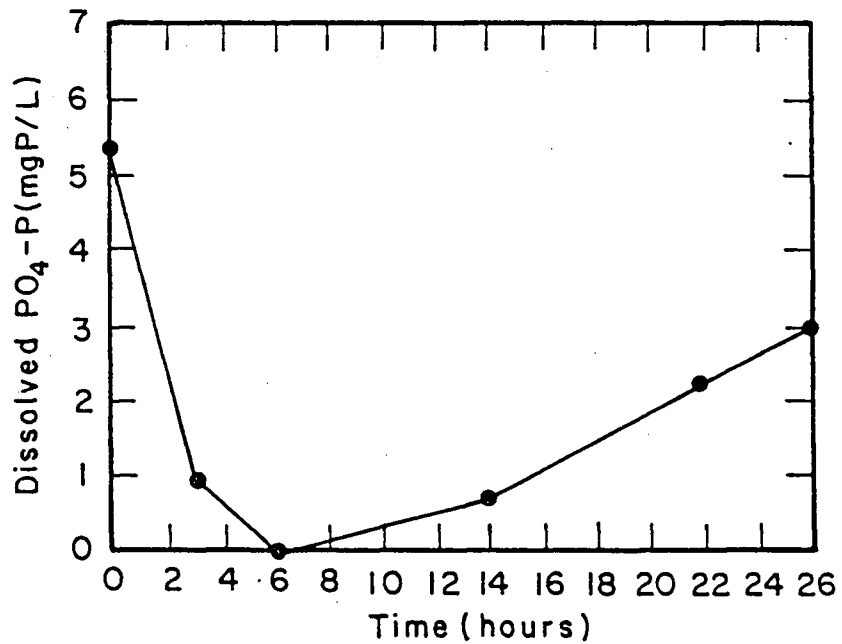
Fig. 2.2. Aerobic batch test on mixed liquor from a process exhibiting excess biological P removal showing both P uptake and release. Data reported by Alarcon (1961).

the sludge concentration while Alarcon reported that the degree
of uptake was a function of the intensity of aeration. Neither
Srinath et al. nor Alarcon offered any explanation for why excess
P uptake occurred or why certain treatment facilities exhibited
excess P removal while others did not.

The first attempt to establish which process operational
parameters enhance removal was carried out by Feng (1962) in a
series of batch tests on sludge obtained from the Nine Springs
Sewage Treatment Plant in Madison, Wisconsin. He concluded that
the conditions favouring excess P removal were a process
temperature around $25^{o}C$, adequate aeration and a correct food/
micro-organism (F/M) ratio. It should be noted, however, that
the above conditions are often found in processes not exhibiting
excess P removal. Of greater significance was Feng's finding
that inadequate aeration adversely affected P uptake and, in some
cases, even resulted in the release of P back into the
supernatant.

The first researchers to propose a biochemical basis for excess P
removal were Levin and Shapiro (1965). They noted the important
role of P in the aerobic utilization of carbohydrates, and that
the literature had indicated the ability of certain fungi, algae
and bacteria to store P in long chains of inorganic poly-P called
volutin granules. Harold (1966) presents an excellent review of
literature available on polyphosphate accumulation. He suggests
that, from an evolutionary point of view, polyphosphate may
predate adenosine triphosphate (ATP) as the principle energy

carrier of the cell. He pointed out that the ability to store excess P gives the organisms an advantage over organisms not having this ability but that there was no clarity as to whether poly-P accumulation serves as an energy storage, or simply a P storage function.

Levin and Shapiro conducted a series of batch experiments on sludge obtained from the District of Columbia Sewage Treatment Plant, a short sludge age or "high rate" process. In one experiment samples were aerated with and without the addition of various substrates (glucose, succinate, etc.). It was found that although P uptake was obtained in both cases, the magnitude and rate of P uptake was greater in reactors receiving substrate addition than in the control reactors, i.e. that the presence of carbonaceous substrate promoted the uptake of P. They confirmed the findings of Alarcon (1961), that in the samples kept under aerobic conditions for long periods of time, P release took place. This phenomenon indicated to them that P storage takes place aerobically by the active fraction of the sludge and that with time, the reduction of the active mass by cell lysis and endogenous respiration results in the release of the associated stored P.

In another experiment designed to demonstrate the role of the sludge active fraction in P uptake, Levin and Shapiro aerated a series of reactors with varying degrees of intensity, one reactor remaining anaerobic. From their results, presented in Fig. 2.3, it can be seen that P uptake was observed in the aerobic reactors
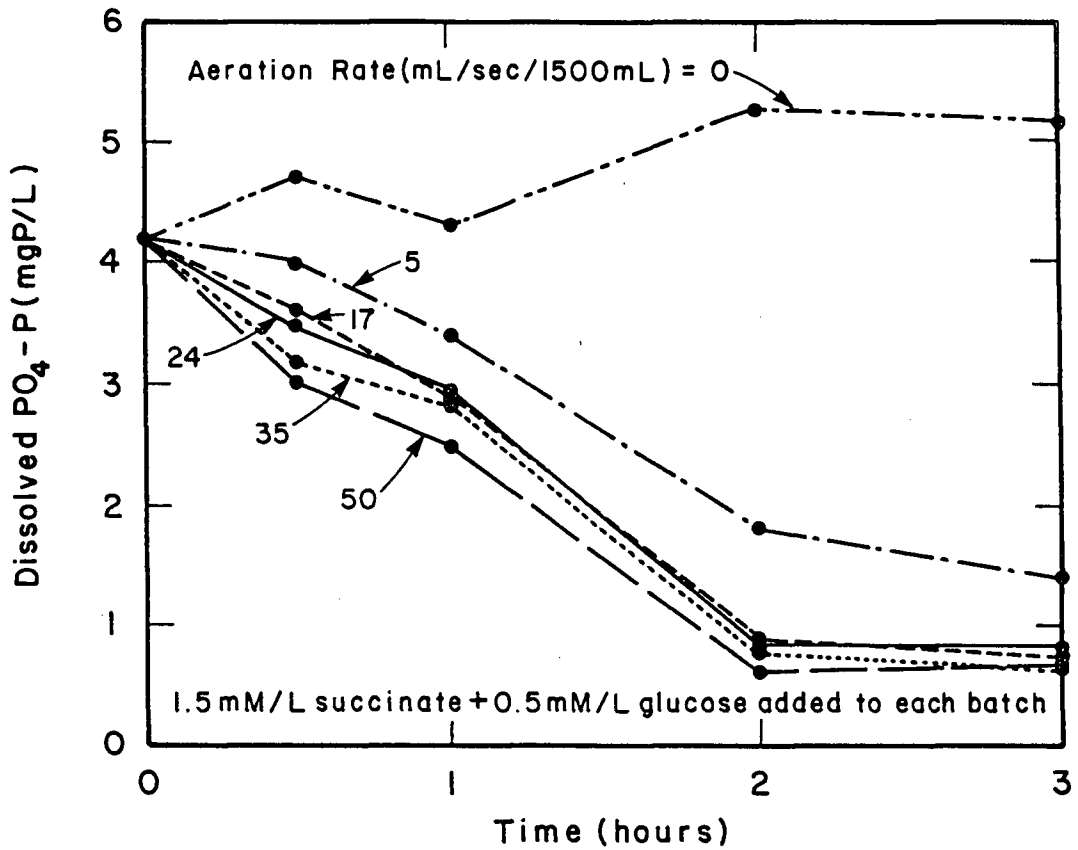
Fig. 2.3. Aerobic batch test sequence showing the
effect of aeration intensity on P uptake.
Data reported by Levin and Shapiro (1965).
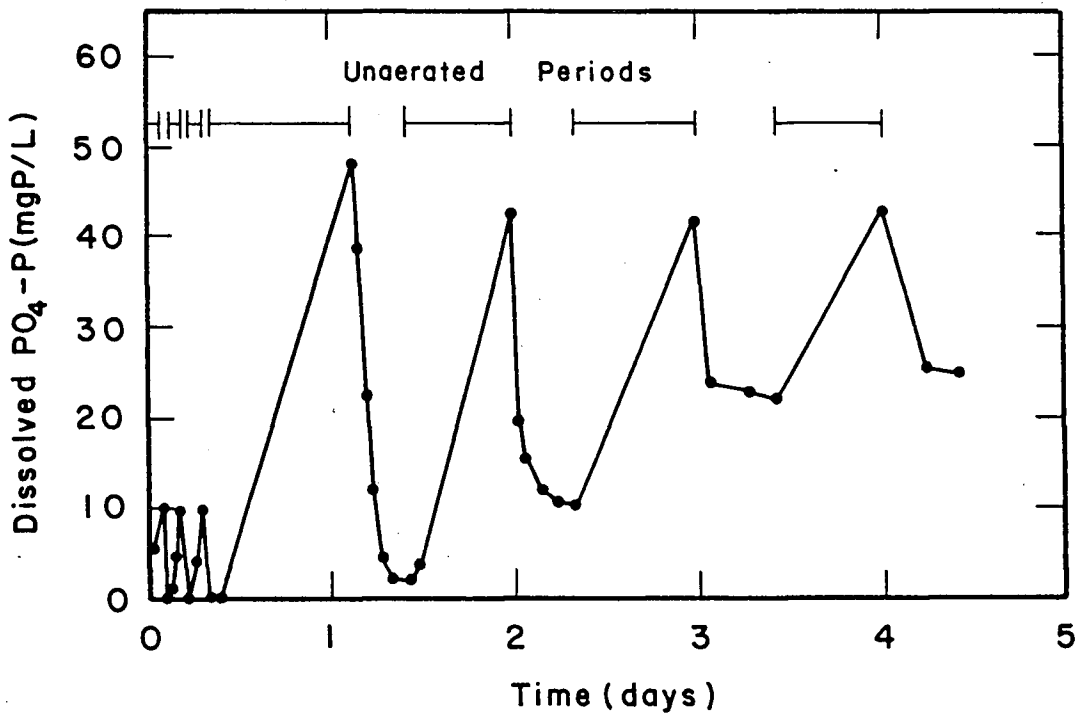
Fig. 2.4. A batch test done under sequential aerated and
unaerated conditions showing P uptake and
release. Data reported by Wells (1969).

but in the anaerobic reactor P release occurred. Furthermore, the uptake of P was related to the intensity of aeration at the lower aeration rates but for higher rates of aeration no improvement in P uptake was observed. They hypothesized that P uptake was the result of ATP formation by oxidative phosphorylation in the aerobic Krebs cycle with oxygen serving as the final electron acceptor. However, no explanation was offered for the release of P under anaerobic conditions and whether this was in any way related to excess P uptake. Shapiro, Levin and Zea (1967) later demonstrated the reversibility of the uptake phenomenon, i.e. that a sample left under anaerobic conditions would release P, but the released P would subsequently disappear from solution upon aeration of the sample. Levin and Shapiro (1965) also demonstrated that the principle excess P removal mechanism is of a biological nature rather than a chemical precipitation. This was done by having a series of batch reactors maintained in the pH range where calcium phosphate occurs with no improvement in P uptake over an uncontrolled reactor, and by demonstrating the inhibitory effects of 2,4-dinitrophenol on aerobic uptake. Dinitrophenol is known to inhibit ATP production and the transport mechanism of the cell, indicating the biological nature of the removal mechanism.

Wells (1969) confirmed the findings of Levin and Shapiro regarding the reversibility of the uptake mechanism by using sludge from the San-Antonio plant, which was exhibiting excess P removal at the time. A batch that was sequentially aerated during the day and left unaerated at night released P under

anaerobic conditions and took up P under aerobic conditions (See Fig. 2.4). Note that both the uptake and release decreased progressively with each of the cycles, probably due to the decrease of the active fraction of the sludge with time. It was Wells who first reported the significance of the sludge characteristics by noting that the sludges from different plants behaved in an entirely different manner when subjected to identical batch test conditions.

Milbury et al. (1970) confirmed the findings of Levin and Shapiro with regard to the significance of intensity of aeration in a batch study. They found that high P uptake could be achieved provided that the dissolved oxygen was kept at a minimum concentration of 0.2 mg/L. Increasing the dissolved oxygen concentration beyond this level did not significantly improve the P uptake.

During the period from the late 1960's till the mid-1970's, research into the P removal mechanism concentrated on establishing whether removal was principly biological or chemical in nature. Proponents of the the chemical removal hypothesis reasoned that the resultant increase in pH due to CO_2 stripping by aeration would create conditions favourable for calcium phosphate precipitation. Vacker et al. (1967) and Yall et al. (1970) found that inorganic precipitation played a minor role in P removal by monitoring Ca^{++}, Mg^{++} and P concentration changes in their experiments. Bargman et al. (1970) conducted aerobic batch tests on sludge from different plants, spiked with

settled sewage, at an elevated pH of about 7.9. The sludge used in these batch tests came from two different plants, only one of which was exhibiting excess P removal. In spite of the fact that the pH in both reactors was kept at around the same value, significant P uptake was only observed in the reactor having sludge from the plant exhibiting excess P removal. From their experiment, Bargman et al. concluded that the excess uptake was a function of some characteristic of the sludge and not due to inorganic precipitation. Hoffmann and Marais (1977) attempted to separate out the mass of P taken up biologically and by calcium phosphate precipitation in two laboratory scale activated sludge processes. The processes were two-stage and identical except that one was purely aerobic and the other anoxic-aerobic, i.e. the "anoxic" reactor had nitrate but no dissolved oxygen present. Batch tests were conducted on sludge from the two processes and the mass of P removed by inorganic precipitation estimated by measuring the changes in Ca^{++} concentration upon raising the pH. They found that all the P removed by the aerobic process could be explained by that required by the organisms for basic metabolic purposes, plus that removed by calcium phosphate precipitation. However, the anoxic-aerobic process consistently removed significantly more P irrespective of whether the processes were run at pH 7.3 or 6.0. From their results, Hoffmann and Marais concluded that chemical precipitation accounted for a relatively small fraction of P removal, a maximum of about 1.5 mg/L. Furthermore, it appeared as if the presence of the anoxic reactor stimulated excess biological P removal in some unexplained

manner. In addition to laboratory scale and batch studies during this period, a number of relevant studies of full scale plant operation were also reported which laid the groundwork for understanding the prerequisites for excess biological P removal.

A number of research workers reported data from full-scale processes exhibiting excess P removal. All of these processes were high rate, plug flow processes with short sludge ages in the 1.5-6 day range. Underflow sludge recycle rates were in the 0.25:1-0.5:1 range with respect to the influent flow. Typical results of the period were presented by Scalf et al. (1969), who were investigating the effects of aeration intensity at the Black River Sewage Works in Baltimore. Dissolved oxygen and phosphorus concentration profiles for the two semi-plug flow aeration tanks are presented in Fig. 2.5. Aeration Tank 2 was aerated at a lower rate than Aeration Tank 1 such that it had oxygen limiting conditions as far as the halfway point along the tank length. The significant difference between the P profiles is that Aeration Tank 2 exhibited P release in the first quarter. Phosphorus uptake occurred in the middle half of the process at a slightly higher rate than in Aeration Tank 1 so that all the P was removed at approximately the three-quarter point. This release was thought to be the result of the reduced aeration intensity but Scalf et al. also came to the erroneous conclusion that the reduced aeration also led to a reduction in the P uptake rate. It was recommended that in order to ensure excess P removal, the dissolved oxygen concentration in the second half of the aeration tank be kept between 2 and 5 mg/L. However, Milbury
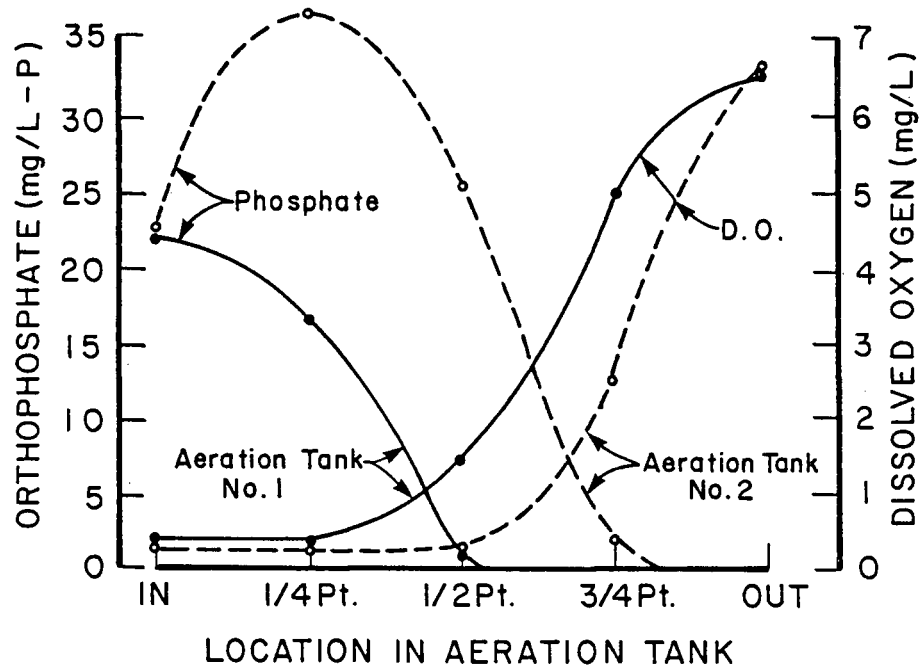
Fig. 2.5. Dissolved oxygen and P concentration
profiles for two parallel aeration
tanks exhibiting excess biological P
removal. Data reported by Scalf et
al. (1969).

et al. (1970) pointed out that excess P removal could not be achieved in a completely mixed regime and, therefore, aeration intensity alone could not guarantee excess P removal. Clearly then, the oxygen limiting conditions at the head end of the plug flow processes played some, as yet undefined, role.

The lack of an adequate understanding of the biochemical basis for excess P removal at the time is clearly illustrated in the design guidelines for excess biological P removal processes presented by Milbury et al. (1971). Their design criteria included recommendations for a plug flow regime, that the feed be introduced at the head end of the process, that sufficient aeration be supplied to the effluent end of the process, that nitrification be avoided, etc. The relevance of P release at the head end of the process and its possible connection with excess biological P removal were not understood.

Fuhs and Chen (1975) were the first research workers to specifically investigate the importance of the anaerobic-aerobic sequence and its effect on the microbiological nature of the sludge. They noted that a microscopic examination of the sludge from an anaerobic-aerobic laboratory-scale process that was not removing excess P revealed very few organisms capable of storing polyphosphate. They subjected sludge from two full-scale plants that were removing excess P to a 4 hour aerobic/20 hour anaerobic batch test sequence that had been spiked with raw sewage. Initially, all the P was removed aerobically and release values of up to 40 mg/L were noted under anaerobic conditions. They

confirmed the finding of Wells (1969) that in a batch test
subjected to a number of anaerobic-aerobic cycles, both the
uptake and release of P progressively decreased to the point that
uptake and release finally ceased. They also confirmed the
inhibitory effects of 2,4-dinitrophenol on aerobic uptake in a
parallel batch test. Using a series of microscopic
identification tests, they attributed the ability to store
excess P to one specific morphological bacterium type - to
organisms belonging to the Acinetobacter genus. Pure batch
cultures of Acinetobacter that were fed with acetate and
subjected to anaerobic-aerobic sequences had similar P release
and uptake patterns to the mixed liquor batch tests.

Fuhs and Chen reasoned that the anaerobic-aerobic sequence
allowed Acinetobacter to flourish in the process because the
anaerobic zone promoted the growth of a facultative population,
which produced compounds such as ethanol, acetate and succinate.
These intermediates then served as the carbon source for the
Acinetobacter, a relatively slow growing obligate aerobe. This
theory did not, however, explain why Acinetobacter grew
preferentially in an anaerobic-aerobic system and did not
flourish in a purely aerobic system to which the above-mentioned
intermediates were added. Furthermore, it did not explain the
release of P in the anaerobic zone or whether this was in any way
related to the excess biological P removal. Further
inconsistencies in the Acinetobacter hypothesis will be dealt
with later in this review. However, the theory of Fuhs and Chen
was significant in that it represented the first microbiological

basis for why excess biological P removal was promoted in a
process having an anaerobic-aerobic sequence.

The first researcher to propose that anaerobic P release is an
intrinsic part of the excess biological P removal mechanism was
Barnard (1976). While investigating the Bardenpho four-reactor
nitrification-denitrification process (Fig. 2.6), he noted that
excessive P release to a concentration of about 30 mg/L in the
secondary anoxic reactor occurred. This was followed by virtual
complete uptake in the reaeration reactor, resulting in excess P
removal by the process. He pointed out that the common feature
between this process and plug flow high-rate processes reported
in the literature to also exhibit excess P removal, was the
significant P release in an anaerobic zone of the process.
Furthermore, he stated that in order to reliably induce excess
biological P removal, the mixed liquor should be subjected to an
anaerobic state of such intensity that P release occurs, and that
P release was the result of a minimum level of oxidation-
reduction potential (ORP) having been attained. He pointed to
the reported difficulties of measuring ORP in mixed liquor and
suggested that P release would be a more easily measurable
parameter indicating that a sufficiently low ORP had been
attained. To this end he suggested a modification of the
Bardenpho process which included an anaerobic zone at the head
end of the process that received both the sludge recycle and the
influent stream. This process became known as the Modified
Bardenpho or Phoredox process (Fig. 2.7). Barnard also drew
attention to the adverse effect that nitrate entering the
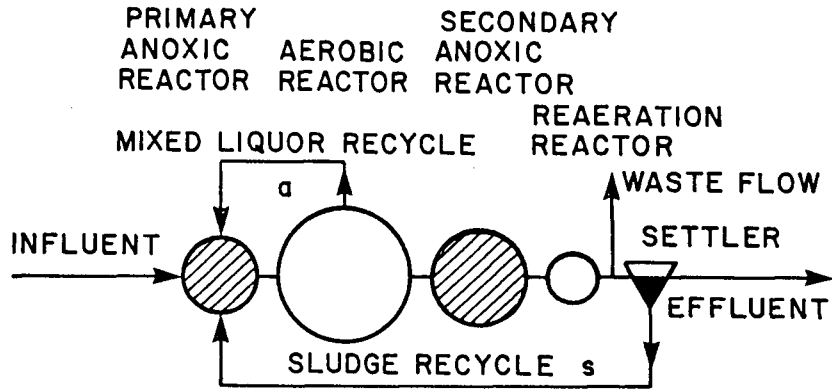
Fig. 2.6. 4-stage Bardenpho nitrification-
denitrification process configuration.
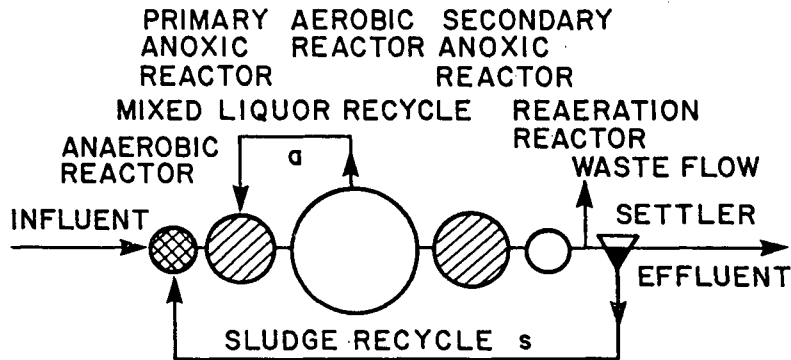
Fig. 2.7. Phoredox or Modified Bardenpho Process
configuration for biological nitrogen and
phosphorus removal.
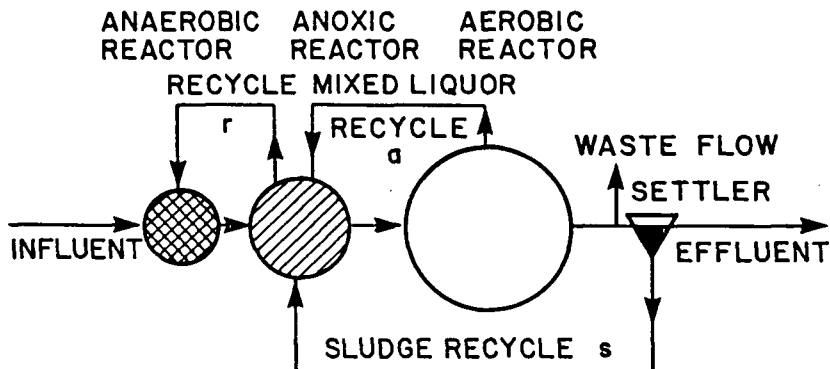
Fig. 2.8. UCT Process configuration for biological
nitrogen and phosphorus removal.

anaerobic zone would have on achieving the required minimum level of ORP or anaerobic stress. The negative effect of nitrate entering the anaerobic zone of the Bardenpho-type processes was further investigated by other researchers (McLaren and Wood, 1976, and Nicolls, 1978) and minimizing its effect proved to be a major challenge in the development of the combined nitrification-denitrification P removal process. These studies confirmed that P release was co-incident with excess P removal and that this release was most likely to occur in the anaerobic zone at the head end of the process.

Nicholls and Osborn (1979) developed a biochemical model to explain excess P removal which extended the hypothesis of Harold (1966), that poly-P accumulation was a consequence of the organisms being subjected to anaerobic stress. They maintained that the ability to store carbon in the form of poly-B-hydroxybutyrate (PHB) played an important role in the survival of aerobic organisms in the anaerobic zone. Briefly, organisms could store excess H^+ ions produced in the oxidation of sewage substrate to pyruvic acid in the form of water insoluble PHB until the onset of aerobic conditions where the ions would be released as water in the aerobic Krebs (TCA) cycle. Their model implicated two survival mechanisms: 1) The energy for ATP formation is derived from the breakup of poly-P chain in the release of P; and 2) that the formation of PHB acts as a sink for excess H^+ ions and electrons. An important recommendation of the research group was the potential benefit of the addition of short

chain volatile fatty acids (VFA's), as found in digester supernatant, to the anaerobic zone of the process, since these acids are the preferred substrates for carbon storage.

In spite of the fact that a number of full-scale Phoredox processes were being designed and constructed, with the majority of these in South Africa, problems with its basic design concept began to emerge. Simpkins and McLaren (1978) found the denitrification rates in the secondary anoxic zone to be significantly lower than in the primary anoxic zone and suggested that it, and the re-aeration zone, be omitted and the primary anoxic zone correspondingly enlarged. All studies into the Bardenpho and Phoredox processes confirmed Barnard's findings with regard to the detrimental effect of nitrate entering the anaerobic zone on P release and excess P removal. More specifically, once a particular configuration has been chosen, the mass of nitrate entering the anaerobic zone (the concentration in the sludge recycle being equal to that in the effluent) is largely a function of the influent sewage characteristics, mainly the COD and TKN concentrations. Because nitrification is usually complete in these processes, and the denitrification capacity is largely a function of the influent COD and the process configuration, any increase in the influent TKN generally results in a corresponding increase in the effluent nitrate concentration and, therefore, the mass of nitrate being recycled to the anaerobic zone.

During the period 1974-1982, an extensive research program into

the nitrification-denitrification kinetics of the activated
sludge process was carried out at the University of Cape Town.
This led to the development of a kinetic model that allowed the
determination of both the nitrification and denitrification
capacities of a particular process, and accurately predicted the
COD, TKN and nitrate concentrations at any point in the process
(Nicholls, 1982). Because the interrelationship between the
nitrification-denitrification kinetics and the excess biological
P removal mechanism were being more clearly understood, the
development of this model proved to be instrumental in the
further development of the biological P removal process.
Briefly, van Haandel et al. (1981) and Dold et al. (1980)
determined that the biodegradable fraction of the influent COD is
made up of two distinct components, each having a profoundly
different effect on the denitrification kinetics of the process.
These fractions are a readily biodegradable fraction which is
rapidly utilized in the denitrification reaction, and a
particulate component that requires enzymatic breakdown prior to
metabolism, and results in a significantly lower rate of
denitrification (that is temperature dependent). It thus became
possible, on this basis, to determine the denitrification
capacity of the anaerobic and anoxic reactors in terms of the
reactor size, process configuration and influent COD
characteristics.

Working on the assumption that redox potential measurement was
difficult in activated sludge (Barnard, 1976) and did not
quantify the anaerobic stress required for P release (Randall et

al., 1970), Rabinowitz and Marais (1980) sought an alternative parameter to quantify anaerobic stress that was based on the nitrification-denitrification kinetic model. The parameter, known as the anaerobic potential, was defined as the difference between the denitrification capacity of the anaerobic reactor and the mass of nitrate entering the reactor (both expressed as mg N per litre of influent). Furthermore, they suggested a modification to the Phoredox process that would ensure that no nitrate entered the anaerobic zone. The sludge recycle and the mixed liquor recycle were discharged into the anoxic reactor, and mixed liquor was passed from the anoxic reactor to the anaerobic reactor via an additional recycle. The configuration became known as the UCT process (Fig. 2.8). By adjusting the recycle rate from the aerobic to the anoxic reactor, the nitrate concentration in the anoxic reactor could be maintained at a low or zero concentration and the detrimental effect of nitrate entering the anaerobic zone could, therefore, be eliminated. Thus the UCT process would be more flexible in operation than the Bardenpho or Phoredox configurations, particularly in treating wastes with high TKN/COD ratios. Therefore, it was more likely to subject the sludge to the required degree of anaerobic stress for the excess biological P removal mechanism to operate. Using the concept of anaerobic potential, Rabinowitz and Marais analysed the data for laboratory scale Modified Phoredox (i.e. having no secondary anoxic zone) and UCT processes and found that in order to achieve anaerobic P release and excess biological P removal, the denitrification capacity of the anaerobic zone must

exceed the mass of nitrate entering the reactor by at least 9 mg N/L. Siebritz et al. (1980, 1983) carried out an extensive series of investigations into the applicability of the anaerobic potential hypothesis for the UCT, Modified Phoredox and Ludzak-Ettinger process configurations. They found that a UCT process, with a 7.5% anaerobic mass fraction, had an anaerobic potential of 12 mg N/L and exhibited P release and system excess biological P removal. However, a parallel anoxic-aerobic (Ludzak-Ettinger) process, with an unaerated sludge mass fraction of 70% and an anaerobic potential of 34 mg N/L, failed to release P and exhibited only minimal system P removal. On application of the UCT kinetic model for nitrification and denitrification, a fundamental difference between the two processes was revealed with regard to the substrate utilization characteristics. In the UCT process most of the denitrification capacity of the anaerobic reactor was derived from the availability of readily biodegradable COD in the influent, none of which is for the denitrification of nitrate entering the reactor. In the Modified Ludzak-Ettinger process, all of the readily biodegradable COD entering the unaerated zone is often oxidized by the nitrate entering the reactor via the sludge and internal recycles. The high denitrification capacity and the resulting high anaerobic potential of the Lutzak-Ettinger process is, therefore, derived from the inordinately large denitrification reactor and the slowly biodegradable COD component therein.

As a result of this breakdown in the anaerobic potential hypothesis, Siebritz et al. (1982, 1983) redefined the prerequisites for excess biological P removal in terms of the concentration of readily biodegradable COD available to the organisms in the anaerobic reactor. Upon the application of statistical analysis to data available for a number of process configurations, they found that anaerobic P release occurred when the readily biodegradable substrate (RBS) concentration entering the anaerobic reactor was greater than 25 mg COD/L. Furthermore, the mass of P released increased as the RBS concentration entering the anaerobic reactor increased above 25 mg/L. This new model clearly demonstrated the reason why the Bardenpho-type process configurations were not optimal for excess biological P removal, particularly for processes treating waste streams with a TKN/COD of greater than about 0.08. At the higher TKN/COD ratios the concentration of nitrate entering the anaerobic reactor can become inordinately large and because each mg of nitrate (as N) results in the oxidation of 8.6 mg of RBS (as COD) (van Haandel et al., 1981), an RBS concentration of 25 mg/L being available to the organisms in the anaerobic reactor becomes impossible. Process configurations such as the UCT process, which make it possible to guarantee a zero nitrate discharge to the anaerobic zone, are therefore at a significant advantage. A further consequence of this model is that wastewaters having a total COD concentration of less than 250 mg/L are not amenable to inducing excess biological P removal, as only about 20% of this COD is in a readily biodegradable form and it gets diluted by the sludge

recycle entering the anaerobic reactor. Siebritz et al. demonstrated the effects of increasing the readily biodegradable COD concentration of the influent to a laboratory-scale UCT process (See Fig. 2.9). The total influent COD was controlled at 800 mg/L but during the experimental period, in which the readily biodegradable component was increased from about 120 to 220 mg COD/L by the addition of acetate, a significant improvement in the system P removal was achieved.

Rensink (1981) extended the hypothesis of Osborn and Nicholls to explain how the presence of an anaerobic zone favours the proliferation of poly-P storing organisms. He hypothesized that the organisms stored carbon derived from short chain VFA's under anaerobic conditions in the form of PHB. The energy for PHB formation is generated from breakdown of poly-P to hydrolized ortho-P, i.e. by P release. Upon entering the aerobic zone, poly-P-storing organisms would have an advantage over other organisms in the form of this stored carbon even if their growth rates are lower, as in the case of Acinetobacter. An important aspect of Rensink's hypothesis is the role of stored poly-P in the storage of carbon under anaerobic conditions. This was demonstrated as follows: He converted a 10 reactor-in-series aerobic process to a 5 anaerobic/5 aerobic reactor series and added periodic dosages of acetate. Initially, there was no P release or excess uptake and very little acetate disappearance from the supernatant in the anaerobic zone (See Fig. 2.10). However, after a six week period, the added acetate disappeared by the time the plug flow reached the third anaerobic reactor and
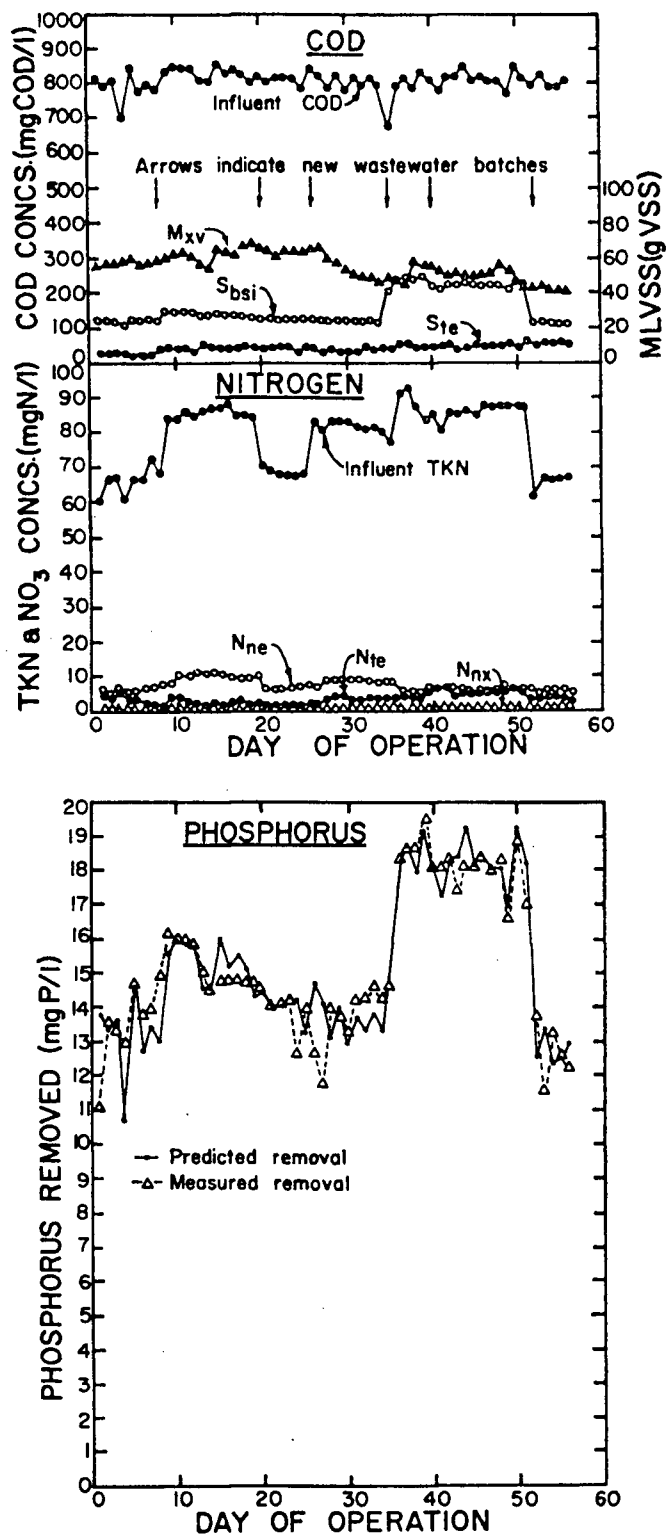
Fig. 2.9. Results from a laboratory-scale UCT process showing the effect of increasing the concentration of readily biodegradable COD in the influent (S_{bsi}). Sludge age = 20 days; Temp. = 20°C. Data reported by Siebritz et al. (1983).
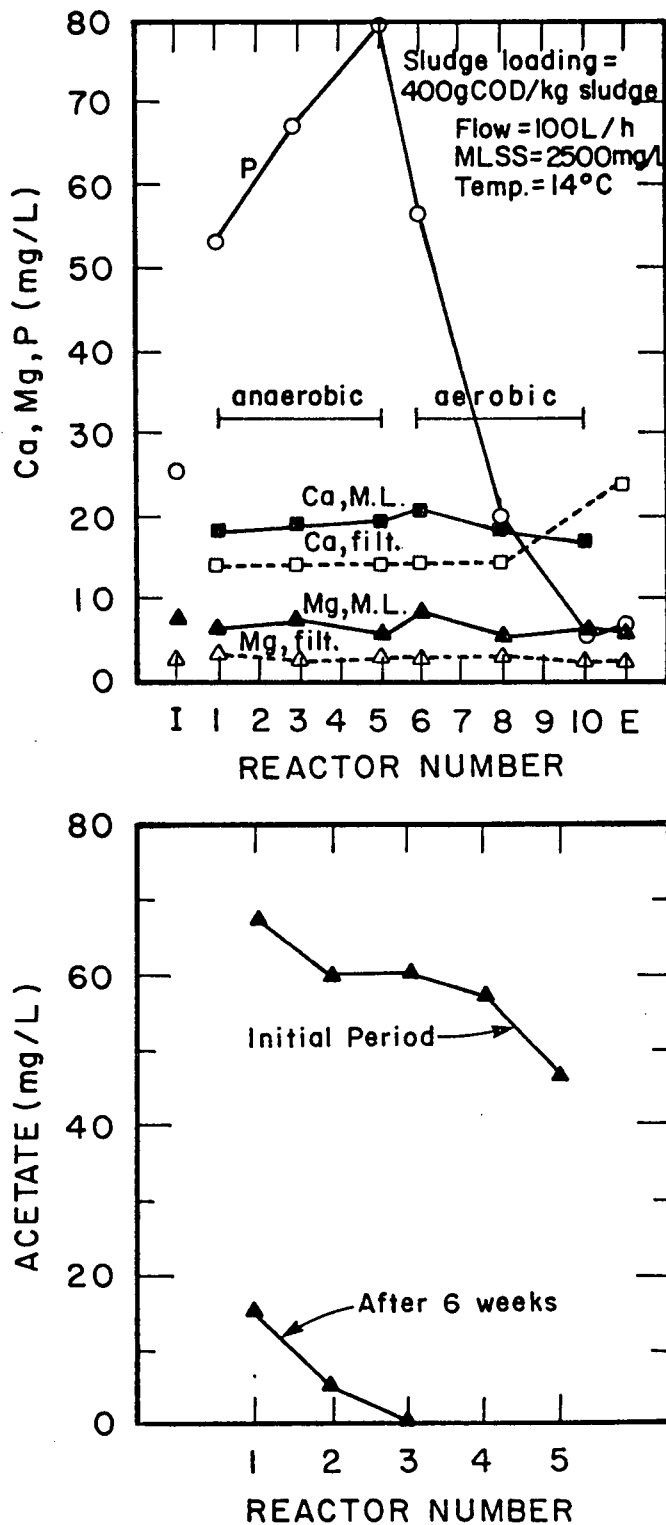
Fig. 2.10. (Top) Results of a 10 reactor-in-series
anaerobic/aerobic process showing P, Ca,
and Mg profiles.
(Bottom) Acetate profiles in the anaero-
bic zone immediately after conversion from
a purely aerobic process and after six
weeks of acetate addition. Data reported
by Rensink (1981).

P release and system excess P removal took place. A microscopic examination revealed a significant increase in the number of poly-P organisms in the sludge. In this way, Rensink demonstrated that anaerobic acetate removal and P release are associated with the presence of poly-P organisms. In contrast to the readily biodegradable COD hypothesis of Siebritz et al., in which the preferred substrates are present in the incoming waste stream, Rensink maintained that the presence of short chain VFA's in the anaerobic reactor was the result of VFA production by facultative organisms and a consequence of a sufficiently low redox potential being attained. This aspect of his hypothesis is still currently being investigated.

Perhaps the most important contribution of Siebritz et al. and Rensink towards the understanding of the excess P removal mechanism was the shift in emphasis away from the degree of anaerobic stress to the significance of the presence of preferred substrates in the anaerobic zone of the process. This contribution has been invaluable in the development of a reliable design strategy for processes in which excess biological P removal is to be guaranteed, as it clarified much about the nature of the excess biological P removal mechanism.

Once the basic hypothesis of the role of specific substrates in inducing excess biological P removal was developed, researchers then began focusing attention on developing a biochemically based model of the mechanism. Fukase et al. (1982) conducted a series of laboratory scale "fill and draw" and continuous flow

experiments using synthetic sewages made up of glucose and
acetate. They found that carbon was stored by the organisms
under anaerobic conditions in the form of glycogen when glucose
was used as the feed, and as PHB when acetate was fed. They also
found the P release was concomitant with the disappearance of
substrate from solution with molar ratios being $\Delta Glucose/\Delta PO_4 = 2$
and $\Delta Acetate/\Delta PO_4 = 1$. Dry weight percent P content of the
sludge values as high as 12.8% were measured in the aerobic zone
of the laboratory-scale unit being fed with acetate. Measurement
of poly-P showed that with sludge containing 7.23% P, 75% of it
was in the poly-P form, while with sludge containing 5.43% P,
only 35% of it was poly-P. This suggests that the additional P
taken up by the sludge was all present in the organisms in the
form of stored poly-P.

Thus far, this section has dealt with a brief overview of the
history of excess biological P removal, tracing the development
of the biochemical model and process design, up until the onset
of this research project. During the course of this study, a
number of parallel research projects were in progress at the
University of British Columbia and in other parts of the world
that are relevant to the findings of this project. For these
reasons, the results of some of these studies will be dealt with
in the Discussion Chapter of this thesis.

2.2. Volatile Fatty Acid (VFA) Production by Primary Sludge Fermentation

The concept of maximizing VFA production by fermentation or
digestion is a relatively new one in wastewater treatment. The

vast majority of research into anaerobic sludge digestion focuses on methane production, where VFA's serve as the intermediate products, i.e. as substrate for the methane formers. In the case of methane production, therefore, the basic objective is to minimize the VFA concentration in the digester final effluent, thereby maximizing methane production. However, there are a number of research projects cited in the literature, in which an attempt was made to separate out the acid and methane forming phases of anaerobic digestion, which offer vital clues to maximizing VFA production.

Jeris and McCarty (1965) found acetate to be an important intermediate fermentation product, accounting for over 70 per cent of the total methane produced during anaerobic sludge digestion. McCarty et al. (1963) pointed to propionate as an important secondary intermediate in the fermentation reaction. Andrews and Pearson (1965) conducted an important series of laboratory scale experiments on anaerobic fermentation at $37^{\circ}C$. A series of completely mixed, continuous flow reactors, with mean residence times ranging from 0.75 to 22.5 days was run using a soluble synthetic substrate made up of both organic and inorganic material. The range of residence times used included experiments in which both acid and methane predominated as the final product of the digestion process. Their results are plotted in Fig. 2.11 with the various effluent carbonaceous fractions being expressed as a function of the influent total carbon concentration. Note that the maximum VFA concentration in the system occurred at sludge ages between 2.5 and 4.5 days, with gas production
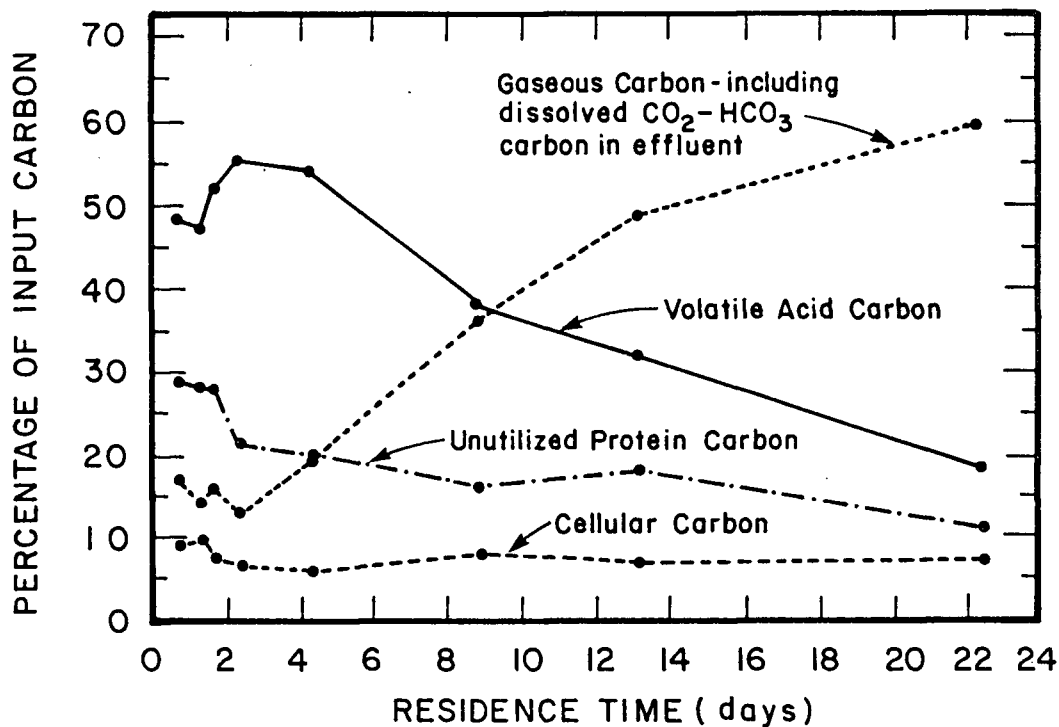
Fig. 2.11. The effect of batch reactor residence time on effluent carbon distribution. Data reported by Andrews and Pearson (1965).
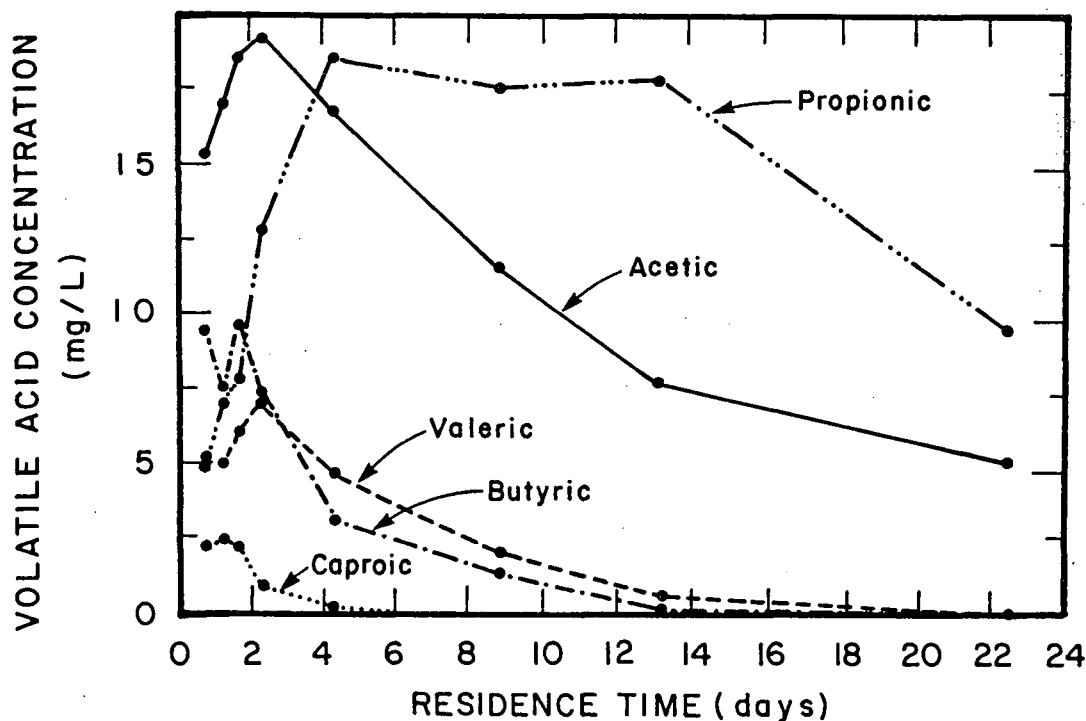
Fig. 2.12. Effluent VFA distribution as a function of sludge age. Data reported by Andrews and Pearson (1965).

(methane and carbon dioxide) predominating at the longer
residence times. They attributed the gas production at the short
residence times to not being able to completely separate the two
phases because of the low generation times of some species of
methanogenic bacteria. From the graph of the distribution of the
various VFA's presented in Fig. 2.12 it can be seen that acetic
and propionic acid represent the vast majority of the VFA
produced. Furthermore, it should also be noted that
significantly lower acetic acid concentrations at residence times
greater that 5 days clearly demonstrate acetic acid to be the
major intermediate for methane production. Although Andrews and
Pearson (1965) did not use primary sewage sludge in their
experimentation, their work demonstrated the important principle
that an anaerobic fermenter could be operated and optimized for
VFA production using mean hydraulic residence time (or sludge
age) as the main process control parameter.

Chynoweth and Mah (1970) present values reported by several
investigators for the composition of raw sewage sludge for the
three major classes of organic compounds: carbohydrates, lipids
and proteins. All three occurred in roughly equal proportions
with carbohydrates averaging 35.5%, and lipids and proteins each
comprising about 28% of the total organic compounds present.
Data is also presented regarding the decomposition of the three
classes of compounds during the fermentation process.
Carbohydrates were decomposed only by about 13%, proteins by
about 36% and lipids by about 76%. This suggests that lipids are
the most important substrate in fermentation, due to their high

degree of degradability. Chynoweth and Mah conducted a series of anaerobic batch studies at 35°C with raw sewage sludge and analysed samples for VFA concentration at 1 hour intervals. Their results are presented in Fig. 2.13 and indicate that acetate is the principle fermentation product with optimal acetate production occurring after only 4 hours. They attribute the decrease in acetate concentration after about 4 hours to the rate of acetate dissimilation due to methanogenesis exceeding its production.

Gosh et al. (1975) developed a two-phase anaerobic digestion process consisting of two separate, completely mixed reactors in series, one for acid fermentation and the other for methane fermentation. From the schematic diagram of the process presented in Fig. 2.14 it can be seen that each phase of the process has its own reactor size and sludge recycle based on growth kinetic requirements for each group of organisms. Gosh et al. found that it was possible to separate out and optimize the two phases of the digestion process by "manipulation of dilution rate and imposition of limits on the microbial generation time" i.e. by controlling the hydraulic retention time and sludge age. With regard to acid fermentation, they found that under mesophilic conditions, acidogenesis occurs at pH 5.7, with an ORP of -240 mV (E_c) and that the volatile fraction of the sludge serves as the major substrate for the acid formers. The maximum specific growth rate for acid formers was 0.16/day @ 35°C and the loading rates and retention times varied between 1.33 to 3.34 g/hr/L and 10 to 24 hours respectively.
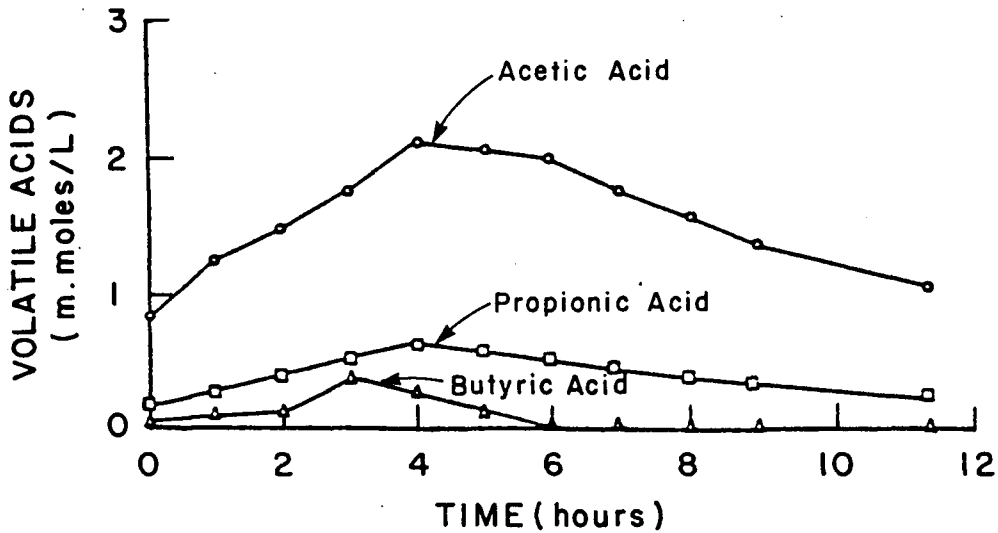
Fig. 2.13. VFA production by primary sludge
fermentation at 35°C. Data re-
ported by Chynoweth and Mah (1970).
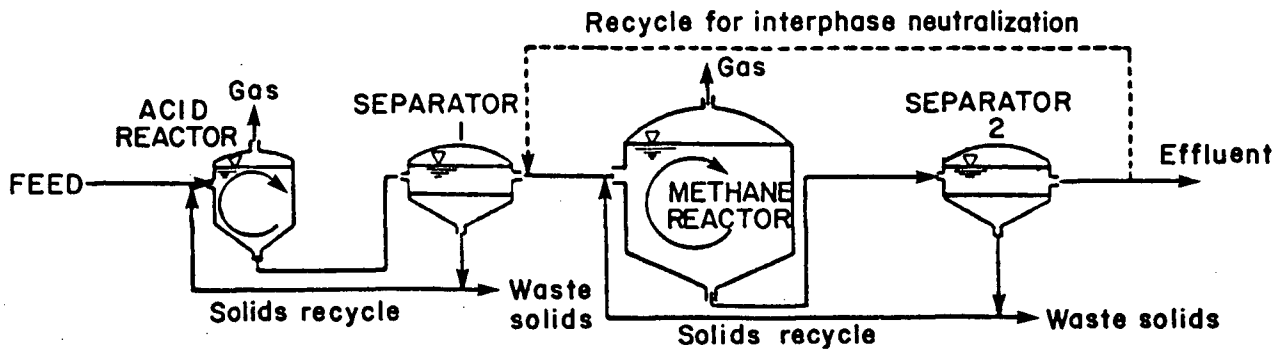
Fig. 2.14. Schematic diagram of a two-phase
anaerobic digestor with separation
of the acid and methane producing
phases by hydraulic control.
After Gosh et al. (1975).

Although the literature cited above deals with experimentation carried out in the mesophilic temperature range (i.e. at temperatures around $37^{\circ}C$), it demonstrates some important principles regarding acid fermentation in general. Firstly, that it is possible to optimize the operation of a primary sludge fermenter for acid production by hydraulic control of the sludge age. Secondly, that the growth rates of acid formers in the $14-22^{\circ}C$ temperature range is likely to be significantly lower than in the mesophillic temperature range. Thirdly, that very little is known about the effect of pH on VFA production but it might be possible to operate an acid fermenter at an uncontrolled pH in the range 5.5-6.0. It is clear, therefore, that because the concept of VFA production by primary sludge fermentation for use as a substrate in the activated sludge process is a relatively new one, knowledge of fermenter design and operation for VFA production is lacking, in particular the effects of sludge age, temperature and pH.

CHAPTER THREE

EXPERIMENTAL METHODS

The experimental methods used in this research are outlined in detail in this chapter. Section 3.1 deals with the analytical methods used on all samples analysed and measurements taken. Experimental procedures for the batch testing and pilot scale operation are outlined in Sections 3.2 and 3.3, respectively.

3.1. Analytical Methods

Chemical Oxygen Demand (COD) analysis was done in accordance with Standard Methods (15th Ed., A.P.H.A., 1980). Ortho-P analysis was done using the stannuous chloride technique also outlined in Standard Methods. In cases where a large number of samples were generated in a short space of time, (e.g. during a batch test), ortho-P analysis was done by the automated ascorbic acid reduction method on a Auto Analyser Model II (Technicon, 1973a). Total Kjeldahl Nitrogen (TKN) and total P samples were subjected to acid digestion and analysed on the Auto Analyser. The digestion method used is fully described in the block digester instruction manual (Technicon, 1974). The dry weight percent P content of the sludge was done by weighing out a given mass of oven dried sludge (104°C) and subjecting it to acid digestion and total P analysis as described above. This method was tested in an anaerobic/aerobic sequence batch test where the P release and subsequent uptake were about 40 mg/L. The changes in the percent P content of the sludge were measured at critical time intervals and found to account for all of the P released to and taken up

from the supernatant. Nitrate and nitrite analyses were done using a copper-cadmium column in which nitrate was reduced to nitrite, followed by colormetric measurement of the nitrite on the Auto Analyser (Technicon, 1973b). Total Filterable Solids (TFS) analysis was done by vacuum filtering a known volume of sample through a pre-washed 5.5 cm diameter Reeve-Angel or Whatman 934-AH glass fibre filter and oven drying it for 1 hour at $104^{o}C$. This method is an adaptation of the crucible method for determining the Total Suspended Solids (TSS) outlined in Standard Methods, and it produced comparable results.

Volatile Fatty Acid (VFA) analysis was done using a Hewlett-Packard 5880A Gas Chromatograph equipped with a flame ionization detector (F.I.D.) and using nitrogen as the carrier gas. The G.C. was fitted with a 0.91 m long, 6 mm O.D. and 3 mm I.D. glass column packed with 0.3% Carbowax/0.1% H_3PO_4 on 60/80 Carbopak C (supplied by Supelco, Inc.). The column was conditioned in accordance with instructions supplied with the packing (Supelco, 1982). A few minutes prior to injection, samples were acidified to a pH of 2 to 3 using a 1% solution of phosphoric acid. A 1 uL sample of the acidified sample was withdrawn and subjected to gas chromatographic analysis. Quantification was done by the external standard method using reagent grade standards dissolved in 0.1% aqueous phosphoric acid.

Dissolved oxygen concentration in the bio-reactors was measured using Model 54A DO Meters (Yellow Springs Instrument Co.). Probes were calibrated and membranes changed regularly. The pH

was measured using an Ionalyzer Specific Ion Meter Model No. 401 (Orion Research), and fitted with a combination electrode. Oxidation-reduction potential (ORP) was measured using a Ag–AgCl Combination Electrode No. 9176 manufactured by Broadley-James. Readout was monitored using a digital meter or a microprocessor using data logging software.

Sludge settling velocity was measured using the standard Sludge Volume Index (SVI) outlined in Metcalf and Eddy (1979). A 1 L sample of mixed liquor was drawn from the aerobic zone and allowed to settle in a 1 L measuring cylinder for 30 minutes. The SVI was calculated as follows:

$$ SVI = \frac{\text{Volume occupied by the sludge in 30 min.(\% of 1000 mL)}}{\text{Total Mixed Liquor Suspended Solids (\%)}} $$

3.2. Batch Test Procedure

For all batch tests, mixed liquor was taken from the aerobic zone of the pilot-scale process on the UBC campus or the full scale 5-stage Phoredox process in Kelowna, B.C., and placed in 1.0 or 2.8 L Erlenmeyer flasks and sealed with a rubber stopper so that no free air was trapped in the flask (See Fig. 3.1). Each stopper had a sampling tube that reached to the bottom of flask and a rubber septum through which all additions were injected. Nitrogen filled rubber balloons were fitted with syringe needles which were pierced through the septum so that the gas replaced any liquid withdrawn from the flask and a nitrogen atmosphere was maintained above the mixed liquor throughout the experiment. Each flask also had a magnetic stirrer in it to provide thoroughly mixed conditions during the course of the experiment. In
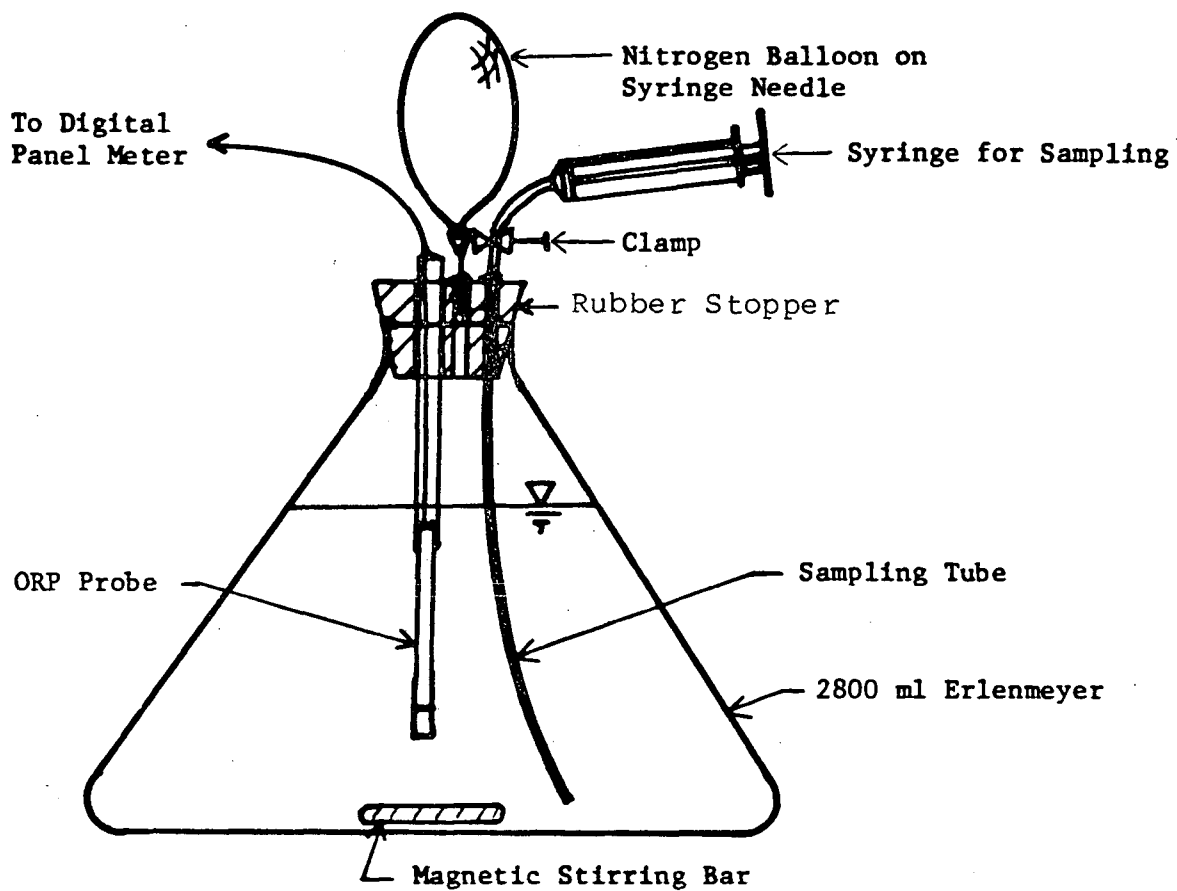
Fig. 3.1. Batch testing apparatus. After Comeau (1984).

experiments where it was important to control or eliminate the nitrate concentration at the beginning of the test, the mixed liquor was allowed to denitrify anaerobically prior to the experiment. This usually took a period of about three hours. Samples were withdrawn from the flask at periodic intervals and centrifuged immediately. The clear liquid was then vacuum filtered through a 0.45 mm millipore filter and subsampled for the various analyses. (During the later stages of the research filtering was done using Whatman #4 filters in order to save time during the experiment. It was found that this in no way affected the analytical results). Ortho-P analysis was done as soon as possible after sampling, usually concurrently with the experiment. Samples for nitrate/nitrite analysis were preserved by the addition of a mercuric acetate solution (0.1 g phenyl mecuric acetate dissolved in 20 ml acetone and 80 ml of water). Samples for VFA analysis were either analysed immediately or preserved by freezing.

3.3. Pilot Plant Operation

3.3.1. Wastewater Source

The pilot plant was situated on the University of British Columbia campus in Vancouver, B.C. A schematic diagram of the plant showing the various components is shown in Fig. 3.2. The wastewater source was a main sewer line that serviced the student residences and on-campus housing and the university sports centre. Raw sewage was pumped daily starting at 10 a.m. with a submersible macerator pump into 2 mechanically mixed plastic storage tanks, each with a capacity of 9000 L, until the
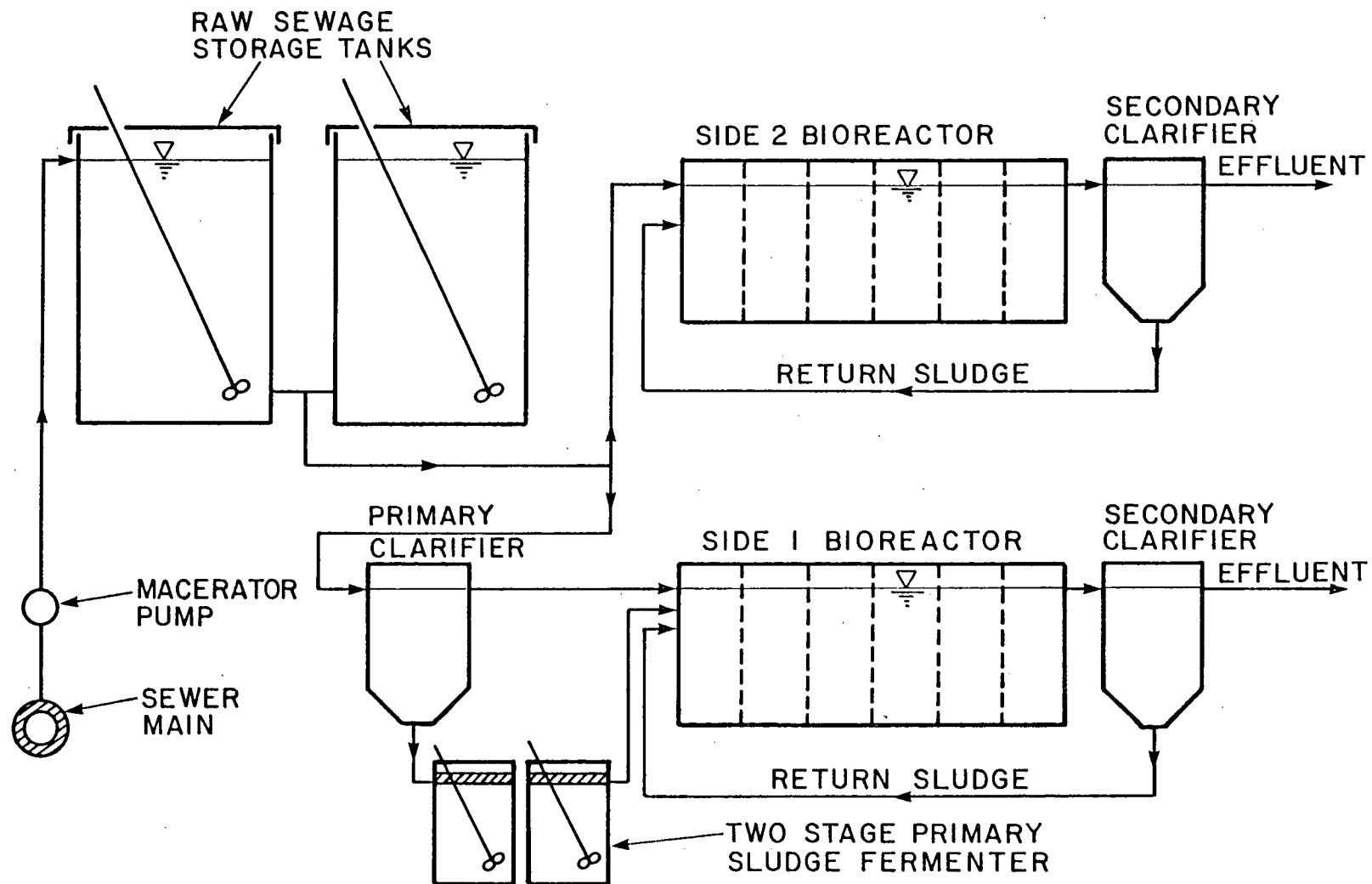
Fig. 3.2. Schematic layout of the pilot plant on the
University of British Columbia Campus

tanks were full. Since wastewater in the Vancouver area has low alkalinity (80-120 mg/L as $CaCO_3$) and is therefore poorly buffered, about 100 mg/L of alkalinity (as $CaCO_3$) in the form of sodium bicarbonate was added daily to the influent storage tanks. This kept the process pH in the 6.50-7.50 range and helped ensure complete nitrification at all times. The influent COD was generally in the range of 200-350 mg/L and the TKN in the range of 25-35 mg N/L. It was found that the sewage was generally weaker during periods of heavy rainfall, probably due to infiltration into the collection system.

3.3.2. Activated Sludge Process

The pilot plant was fitted with two mild steel rectangular aeration tanks that could each be compartmentalized into a maximum of six zones using aluminum baffles. The total process volume could be adjusted between 2500 and 3500 litres by adjusting the level of the overflow weir fitted at the end of each bioreactor. Aeration and mixing in the aerobic zone of the process was provided by coarse bubble aeration through header pipes at the bottom of the tank. The dissolved oxygen concentration in the aerobic zones was kept at between 1-2 mg/L. Mechanical mixing in the anaerobic and anoxic zones was provided by DC motors equipped with electronic speed controllers and gear drives. Mixed liquor from the aeration tanks flowed into two 540 litre secondary clarifiers each equipped with two concentric V-notch weirs. Each clarifier was also equipped with a gear driven mechanical rake set to turn at about 1 rpm.

The process sludge age was maintained by wasting sludge from the aerobic zone of the aeration tank once daily. However, it was found that when the effluent contained a relatively high concentration of suspended solids, a significant fraction of the required daily sludge wastage would occur over the clarifier weir. As a result, sludge wastage charts, such as the one shown in Fig. 3.3, were drawn up for each process configuration tested and used to calculate the required daily mixed liquor wastage, taking into account the solids lost over the clarifier weir. For example, if the effluent solids concentration was equal to 25 mg/L and the process volume and solids concentration were 2750 L and 3000 mg/L, respectively, the volume of mixed liquor to be wasted was reduced from 138 to 100 litres in order to maintain a sludge age of 20 days.

3.3.3. Pilot Plant Fermenter

The pilot plant fermenter consisted of a primary clarifier of 540 litres capacity and two fibreglass reactors (in series) with a total volume of 1000 litres. Primary sludge was pumped from the clarifier bottom into the first reactor via a positive displacement pump connected to a timer. The cycle time was 30 minutes and the mean flow rate through the fermenter was set by adjusting the length of the ON time. In this way the desired mean hydraulic retention time and the sludge age of the fermenter were maintained. Mixing was provided by air driven mixers equipped with speed controllers. In order to minimize air/liquid contact, the reactors were fitted with styrofoam floating covers. During the later stages of this research, when
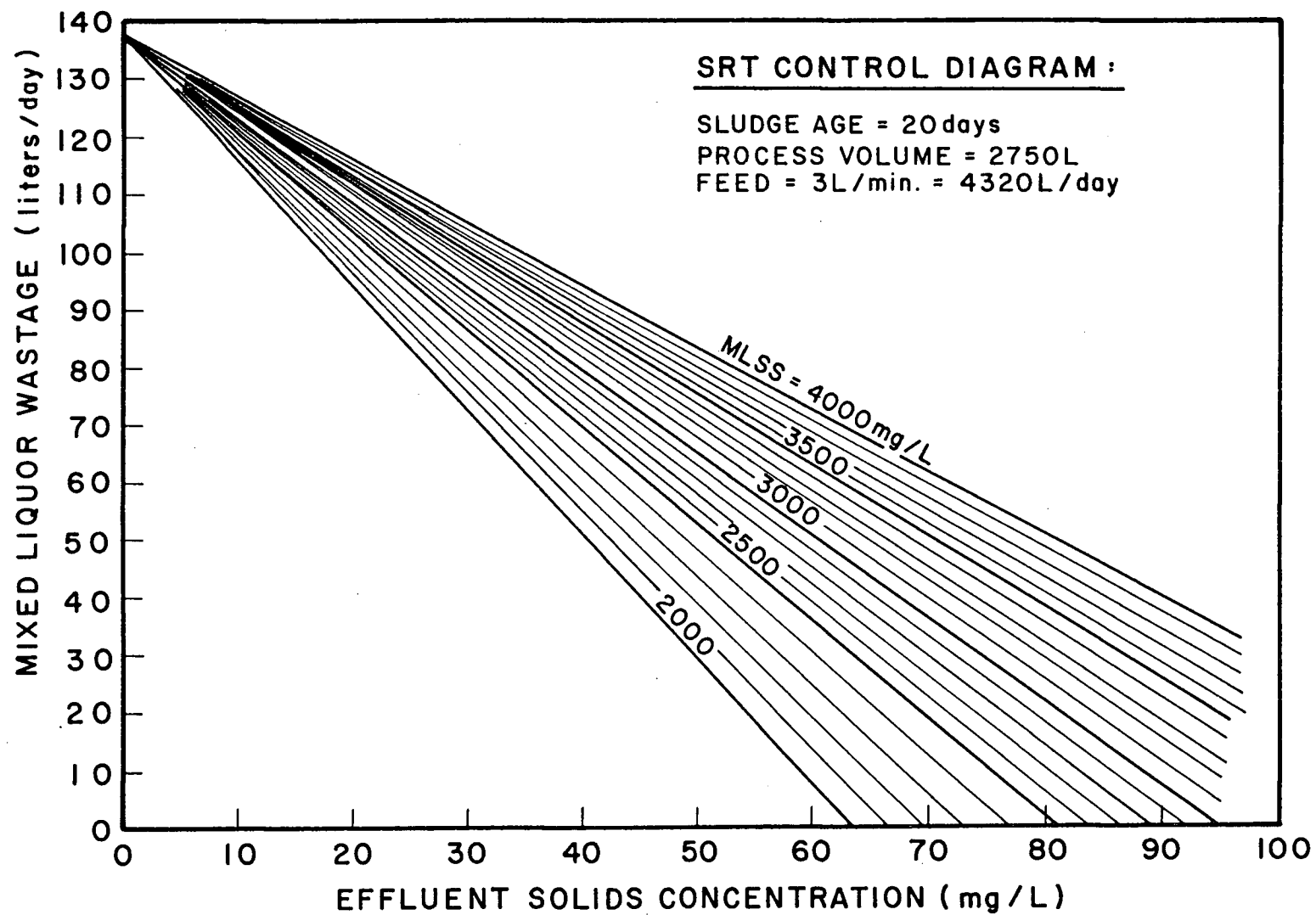
Fig. 3.3. Mixed liquor wasting chart for sludge age control
based on the effluent total solids concentration

the fermenter sludge age was increased to about 10 days, a fermenter secondary clarifier, with a volume of 130 litres, was installed. Fermented sludge was recycled from the bottom of this clarifier to the first fermentation reactor. In this case, fermenter sludge age was maintained by automated wastage from the second fermentation reactor.

3.1.4. Pilot Plant Sampling and Analysis

During the course of each pilot plant experiment, a rigorous weekly schedule of 24-hour composite and grab sampling was adhered to. Samples of the raw influent, settled sewage and effluent from each activated sludge process were taken automatically every 15 minutes, pumped directly into a refrigerator, and composited over 24 hours. All other sampling was done an a grab basis in the early morning. A summary of the sampling type and frequency and the corresponding analysis is shown in Table 3.1 and the weekly data logging schedule is shown in Table 3.2.

Table 3.1. Pilot Plant Weekly Sampling Schedule
            showing various analyses

	Raw Influent	Settled Sewage	Bio-Reactors	Fermen. Reactors	Effluent	Effluent (filt.)
COD	DC	DC	-	-	2WC	-
PO$_4$	-	-	2WG	-	-	2WC
NO$_3$,NO$_2$	-	-	2WG	-	-	2WG
Total P	DC	-	-	-	-	DC
TKN	DC	-	-	-	-	DC
VFA	-	-	-	DG	-	-
MLSS	-	-	2WG	-	DC	-
SVI	-	-	DG	-	-	-
pH	2WC	-	2WG	DG	-	-

DC - Daily Composite
DG - Daily Grab
2WC - Twice Weekly, Daily Composite
2WG - Twice Weekly Grab

Table 3.2. Pilot Plant Weekly Data Logging Schedule

	Raw Influent	Primary Sludge	Bio-Reactors	Fermen. Reactors	Recycles	Effluent
Temp.	D	-	-	D	-	D
ORP	-	-	C	C	-	-
Flow rate	3W	1W	-	-	3W	-

D - Daily
C - Continuous
1W - Once Weekly
3W - Thrice Weekly

CHAPTER FOUR

BATCH TESTING RESULTS

A series of laboratory scale batch tests was carried out in order
to gain a better understanding of the role of the anaerobic zone
of the P removal activated sludge process. In general, batch
tests were designed to simulate conditions in the anaerobic zone
and to test the effectiveness of various substrates at inducing P
release, measure the disappearance of the substrate during
release, the role of P release in the excess removal mechanism
and the negative effect of the presence of nitrate in the
anaerobic zone. In the various tests, the sludge used was either
drawn from the pilot plant at UBC or the full scale process at
Kelowna, B.C. Clearly then, some differences in the experimental
results can be attributed to variations in the biomass
characteristics. The most important of these differences was the
degree to which each flow-through process was biologically active
in removing excess phosphorus at the time of the batch test.
This was quantified by examining the dry weight percent P content
of the mixed liquor sludge used in the various experiments. An
additional factor that may explain variance in the results of
different experiments to a lesser degree is the MLSS value of the
sludge used.

4.1. The Effectiveness of Various Substrates at Inducing
    Anaerobic P Release in Activated Sludge

4.1.1. Experiment Using UBC Pilot Plant Sludge

The objective of this experiment was to compare the effect of

equivalent masses (in terms of COD) of various substrates on P

release in activated sludge under anaerobic conditions. The

mixed liquor used in this experiment was drawn from the pilot

plant which was being operated in a UCT process configuration

(See Fig. 2.8). At the time of the experiment the process and

sludge characteristics were as follows:

Sludge Age = 20 days

MLSS = 2950 mg/L

P removal = 3.50 - 1.30 = 2.20 mg P/L

COD removal = 220 - 24 = 196 mg COD/L

Sludge dry weight percent P = 2.2%

Stock solutions were prepared of the various substrates (sodium

acetate, glucose, propionic and iso-butyric acids) with

approximate COD strengths of 10,000 mg/L. The exact COD values

were then determined analytically. From the COD value of each

stock solution, the required volume to be added to each flask was

calculated so that the COD concentration of the added substrate

in the flask at the beginning of the experiment would be 100

mg/L. Using a syringe, this required volume was then injected

into sealed flasks of mixed liquor taken from the aerobic zone of

the process and samples were drawn at various time intervals.

The samples were centrifuged, filtered and the supernatant

analysed for ortho-P concentration. In this experiment two

control flasks were used; one which received no substrate and another to which 100 mg/L (as COD) of sodium acetate was added after 2 1/2 hours. The results of this experiment are presented graphically in Fig. 4.1.

From the results plotted in Fig. 4.1 it can be seen that more P was released in all flasks receiving substrate than in the control. Furthermore, although the flasks receiving substrate all had the same mixed liquor and initial COD concentration, the mass of the P released in each flask differed significantly. For example, it is clear that sodium acetate is the most effective substrate at inducing P release, followed by propionic acid, glucose etc. The effectiveness of the substrate appeared to vary as an inverse function of the carbon-chain length of the substrate. The P release rate in the Control II flask after the late addition of sodium acetate appears to be comparable to the initial release rate in the sodium acetate experimental flask.

4.1.2. Experiment using Kelowna Full-Scale Plant Sludge

In this experiment, the procedure of Section 4.1.1 was repeated on mixed liquor taken from the full scale plant at Kelowna, B.C. At the time of the experiment the plant was being operated in a 5-stage Modified Bardenpho configuration and was removing a significant mass of P biologically, as shown by the following process and sludge characteristics:
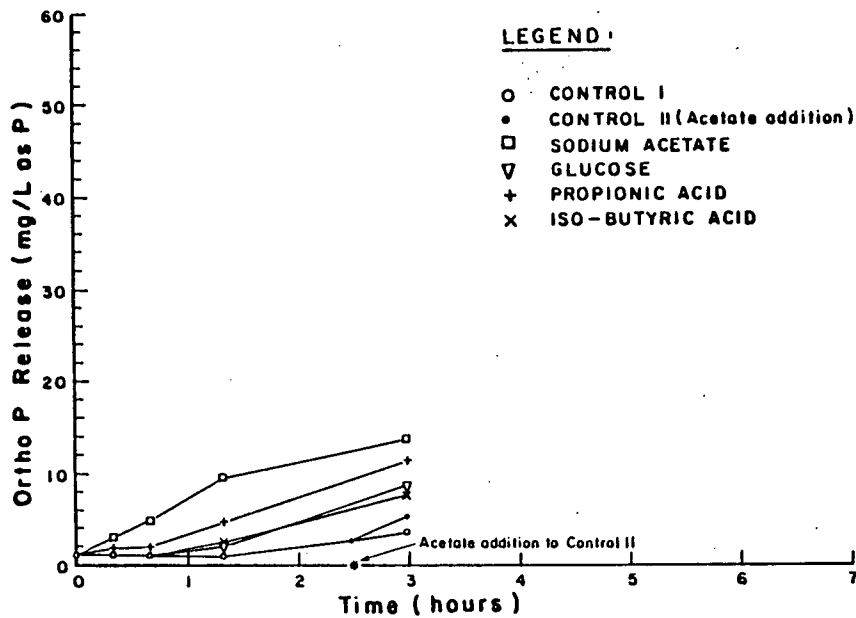
Fig. 4.1. Phosphorus release induced in UBC pilot plant sludge using various substrates
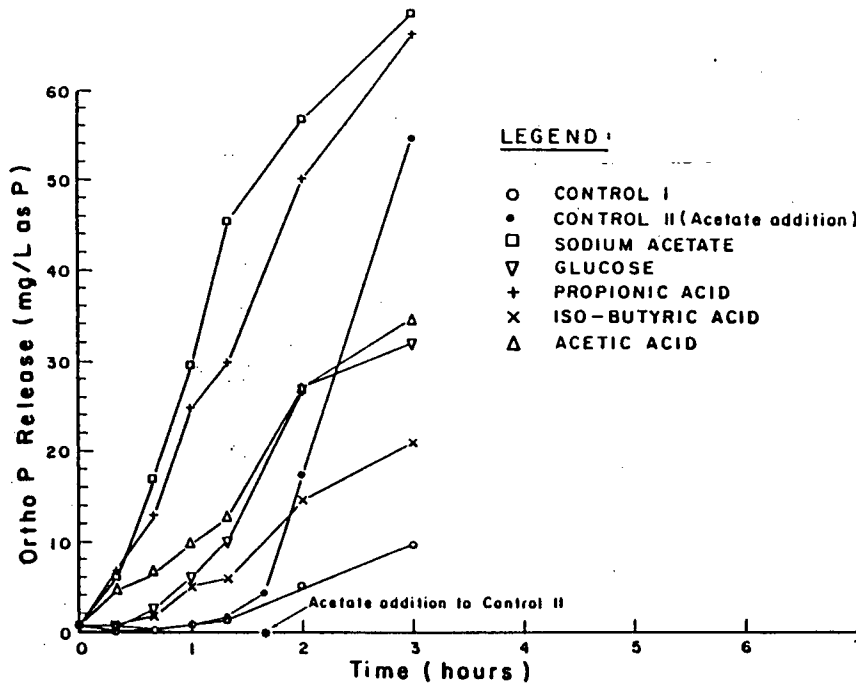
Fig. 4.2. Phosphorus release induced in Kelowna plant sludge using various substrates

Sludge Age = 35 days

MLSS = 3500 mg/L

P removal = 5.5 - 0.2 = 5.8 mg P/L

COD removal = 200 - 20 = 180 mg COD/L (approx.)

Sludge dry weight percent P = 4.5%

The results shown in Fig. 4.2 show a dramatic increase in the concentration of P released when compared with Fig. 4.1. However, the results also indicate a similar distribution of release magnitudes, i.e. the degrees of release are an inverse function of the carbon chain length. Sodium acetate (2-carbon chain) was the most effective substrate at inducing P release, followed closely by propionic acid (3-carbon chain), glucose and iso-butyric acid (4-carbon chain). An interesting observation is that acetic acid was significantly less effective than sodium acetate at inducing P release in this experiment. A possible explanation for this phenomenon is that the acid form undergoes tranformation to the ionic form (pK value = 4.80 at $25^{o}C$) on entering the flask which has a pH value close to neutrality, and this transformation might be a rate limiting step. A more likely explanation for this phenomenon is the form in which the substrate passes through the cell membrane. This explanation is dealt with in the discussion of the biochemical model for excess biological P removal of Comeau et al. (1985b) in Chapter Seven. As in the case of the previous experiment, the P release rate in the control flask that received a late acetate addition was similar to that in the experimental acetate flask.

The most striking difference between the two experiments,
however, is the vast difference in magnitude of the P released
for each substrate, e.g. 72 mg/L vs. 12 mg/L of P after 3 hours
for a substrate addition of 100 mg/L of sodium acetate (as COD).
This is probably best explained by differences in the
characteristics of the two sludges used. At the time of the
experiment the full scale plant at Kelowna was clearly exhibiting
excess biological phosphorus removal (dry weight % P = 4.5%)
while the pilot plant at UBC was only exhibiting a small degree
of excess biological removal (dry weight % P = 2.2%). If one
assumes that most, if not all, of the difference in the P content
of the two sludges was present in the form of the stored poly-P,
then it is quite clear that the Kelowna sludge had significantly
more P available for release or, at least, a higher percentage of
the biomass was capable of excess P storage. The importance of
dry weight % P in the sludge and its relevance to excess
biological P removal will be dealt with in greater detail in
Chapter Seven of this thesis.

4.2. The Effect of Varying the Concentration of a Given Substrate on Anaerobic P Release and Subsequent Aerobic P Uptake in Activated Sludge

4.2.1. Experiment Using UBC Pilot Plant Sludge

The two experiments in the previous section clearly demonstrate
that sodium acetate is an important substrate involved in the P
release mechanism. The objective of this experiment was to model
the conditions present in the anaerobic zone of the process and
subject the sludge to a range of concentrations of sodium acetate
with the view to quantifying the changes in supernatant ortho-P

and NO_3-N concentration with time. The mixed liquor used in this experiment was drawn from the aerobic zone of an anoxic/ anaerobic/aerobic pilot scale process at U.B.C. The sludge was well acclimatized to the use of sodium acetate as a substrate since the process feed had been supplemented by the continuous addition of 112 mg/L of sodium acetate (measured as mg COD per litre of influent) directly to the anoxic zone for several weeks prior to the experiment. The process was also exhibiting a small degree of excess biological P removal (this constituted virtually 100% P removal, however) as can be seen in the following process and sludge characteristics:

Sludge Age = 20 days

MLSS = 3152 mg/L

P removal = 3.40 - 0.10 = 3.30 mg P/L

COD removal = 198 + 112 - 26 = 284 mg COD/L

Sludge dry weight percent P = 2.3%

At the start of the experiment, a range of concentrations of sodium acetate (20, 40, 60, 80 and 100 mg/L as COD) were injected into the anaerobic flasks. A control flask that received no acetate addition was included for the sake of comparison. Because of the low nitrate concentration in the original mixed liquor (2.2 mg/L NO_3-N) it was decided to spike each reactor with a sodium nitrate solution to bring the concentration up by 6 mg N/L. It was felt that this would more closely approximate the characteristics of the anaerobic zone of some of the Bardenpho type-process and give some preliminary information regarding the effect of nitrate entering the zone.

The ortho-P, nitrate and ORP profiles are presented in Fig. 4.3.
Examination of the ortho-P profiles shows that each profile
appears to have two distinct release phases; a very rapid
initial release rate which appears to be similar for all the
experimental flasks and in which the degree of P release
appears to be a function of the initial substrate concentration,
followed by a significantly slower rate at which release
continues for the duration of the anaerobic period. Furthermore,
in the flasks that received lower acetate dosages and exhibited
this initial rapid release (40 and 60 mg COD/L experimental
flasks), a certain degree of P uptake occurred in the time period
after the initial release. Examination of the nitrate profiles
(Fig. 4.3b) shows that over this period of P uptake, nitrate was
still present in the reactors, i.e. complete denitrification had
not yet occurred. It is assumed that in the flasks that received
the lower initial substrate concentration, substrate limiting
conditions were present in the flasks at this point. This
situation is analogous to the anoxic zone of the Bardenpho-type
processes where P uptake is known to occur in cases where
substrate is limiting and nitrate is entering the zone via the
internal recycle from the aerobic zone of the process. Many
activated sludge organisms are known to follow almost similar
metabolic pathways when using either oxygen or nitrate as the
final electron acceptor.

The ORP profiles presented in Fig. 4.3c (only 4 flasks were
monitored) do not demonstrate that a minimum absolute level
of redox potential defines the necessary prerequisite for P
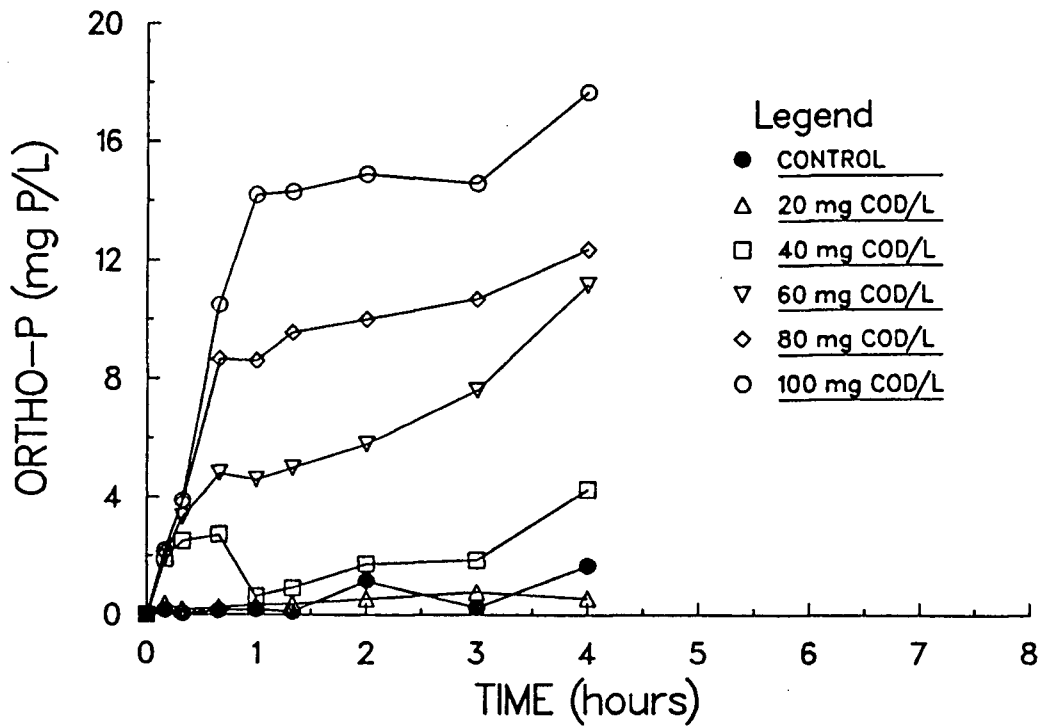
Fig. 4.3a. Ortho-P concentration profiles: The effect of substrate concentration on anaerobic P release - nitrified sludge (Section 4.2.1)
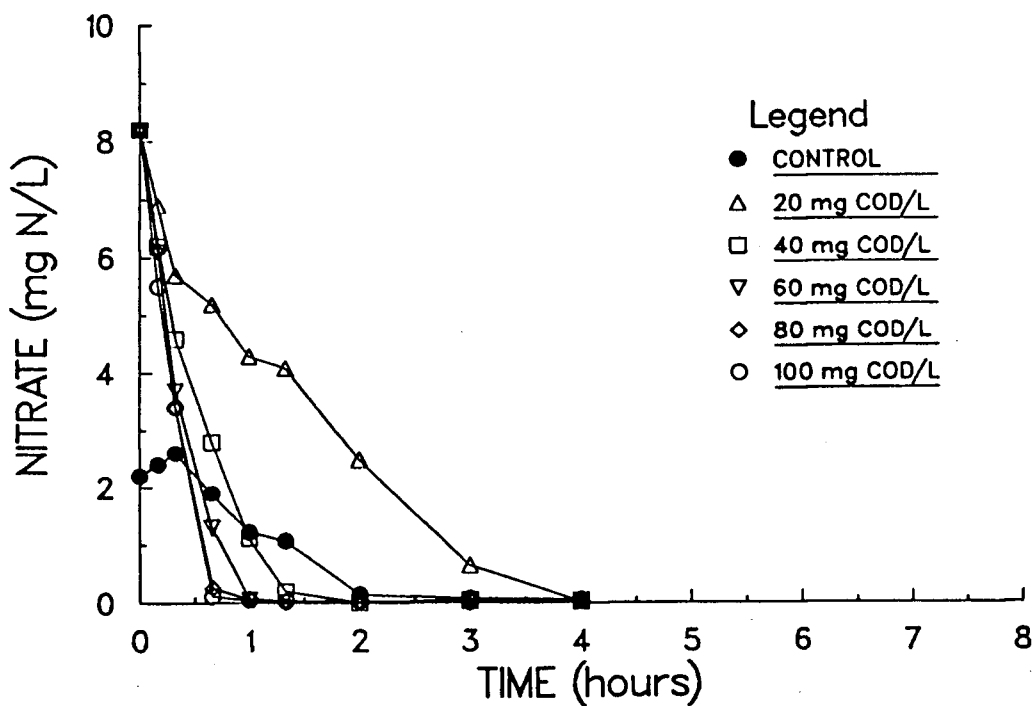
Fig. 4.3b. Nitrate concentration profiles: The effect of substrate concentration on anaerobic P release - nitrified sludge (Section 4.2.1)
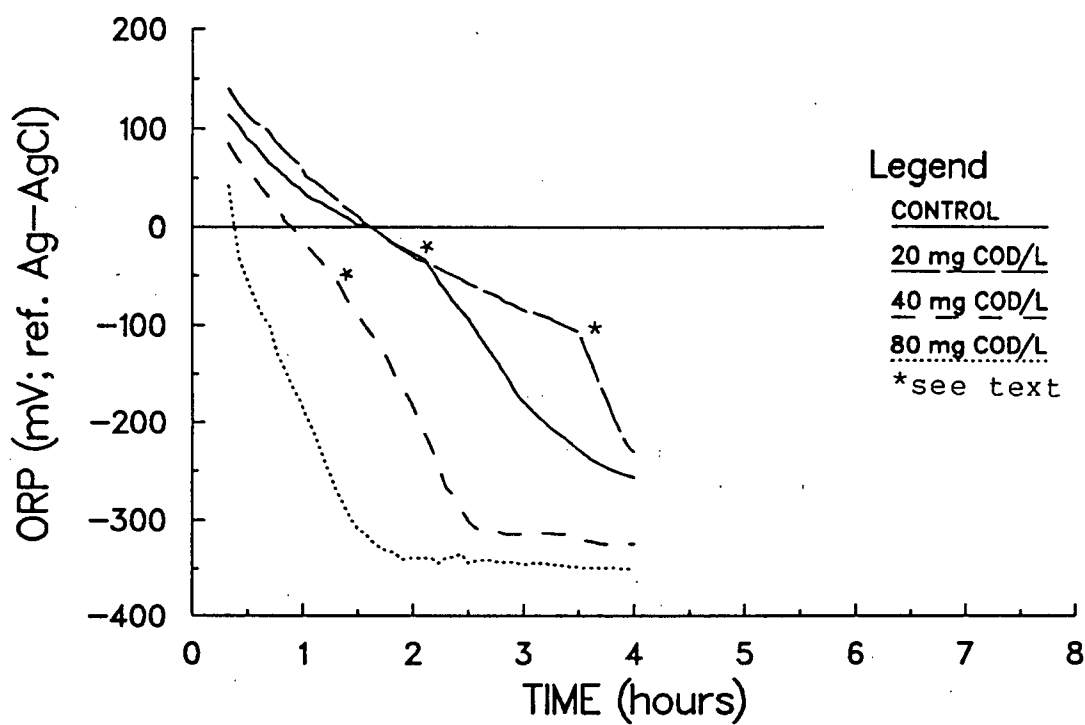
Fig. 4.3c. ORP profiles: The effect of substrate
concentration on anaerobic P release –
nitrified sludge (Section 4.2.1)

release. Quite clearly, higher levels of substrate caused the ORP level of the flask to drop at a higher rate and to greater negative values than in the control flask, but P release commenced virtually immediately upon substrate addition and prior to the high negative values being attained. This may be attributable to the experimental procedure used. It is known, for example, that ORP electrodes may require several hours or even days of conditioning time in a given solution before giving accurate absolute measurement. It is, therefore, possible that this time lag·between highly negative ORP values being attained in the flasks and the onset of P release is primarily due to the long electrode response time. The ORP profiles do, however, show an interesting phenomenon with regard to the denitrification kinetics. For the three profiles measured, there exists a definite increase in the rate of the drop in ORP as marked by an asterisk * in Fig. 4.3c (this is clearly apparent in the 20 and 40 mg COD/L flasks, but not easily visible in the 80 mg COD/L flask, where the initial rate of change in ORP was the greatest. The nitrate profiles in Fig. 4.3c clearly show that these "bends" in the ORP profiles are coincident with the total disappearance of nitrate from solution. This can be explained by the fact that nitrate is a strong oxidizing agent as demonstrated by its high standard electrode potential (E^o = +0.42v @ 25°C).

This experiment clearly demonstrates that the mass of P released is a function of the mass of substrate available to the organisms under anaerobic conditions. However, in order to obtain information regarding the fate of the substrate and its

possible connection to P release, substrate profiles are also required. In addition, because some of the substrate is also utilized in the denitrification reaction (8.6 mg COD per mg N by stoichiometry), it is clear that the presence of nitrate in the flask at the start of the experiment introduced a confounding variable, primarily due to the substrate requirement for denitrification. These two factors were considered in the design of the experiment reported in Section 4.2.2.

4.2.2. Experiment Using Denitrified UBC Pilot Plant Sludge

This experiment had a similar procedure to that discussed in Section 4.2.1 except that the mixed liquor was stored anaerobically for about three hours prior to the start of the experiment in order to allow complete denitrification to take place. All samples drawn were acidified and analysed for VFA concentration so that the fate of the substrate during anaerobic P release could be determined. A third modification to this experiment was that an additional 4-hour aerobic phase was included immediately after the 3-hour anaerobic phase, in order to measure the P uptake and to determine whether it was in any way related to the degree of anaerobic release. The mixed liquor used was drawn from a 3-phase anoxic/anaerobic/aerobic process that was receiving a continuous acetate addition of 24 mg/L (measured as mg COD per litre of influent) at the time of the experiment. The process and sludge characteristics were as follows:

Sludge Age = 20 days

MLSS = 3550 mg/L

P Removal = 3.80 - 0.10 = 3.70 mg P/L

COD removal = 249 + 24 - 25 = 248 mg COD/L

Sludge dry weight percent P = 3.2%

In order to simplify the experiment, the number of experimental flasks was reduced to four. At the start of the experiment, the four experimental flasks received acetate dosages of 25, 50, 75, and 100 mg/L as COD, respectively. The control flask received no substrate addition. Just prior to turning on the air supply (i.e. 15 minutes before the end of the anaerobic phase) the flasks were each spiked with a sodium phosphate solution (approximately 45 mg P/L) in order to ensure that P limiting conditions did not occur during the aerobic phase.

The ortho-P, acetate and ORP profiles are presented in Fig. 4.4. The most apparent difference between the results of this experiment and those of Section 4.2.1 is the significant increase in the mass of P released under anaerobic conditions for the same substrate addition, i.e. 60 mg P/L vs 12 mg P/L after three hours in the flask that received an initial substrate concentration of 100 mg COD/L. This can probably be attributed to two factors: firstly, by completely denitrifying the sludge prior to the start of the experiment the substrate requirement for denitrification was removed; and secondly, the dry weight % P content of the sludge used in this experiment was significantly higher than in the previous experiment (3.2% vs. 2.2%) and, therefore, more
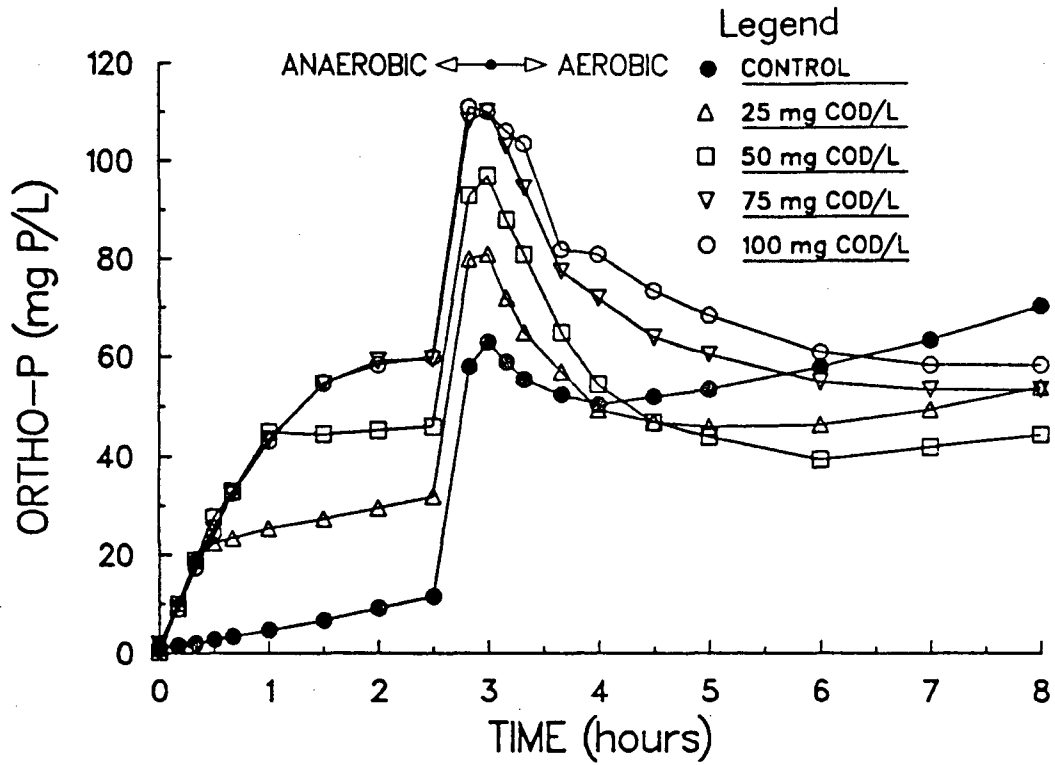
Fig. 4.4a. Ortho-P concentration profiles: The effect of
substrate concentration on P release and
uptake - denitrified sludge (Section 4.2.2)
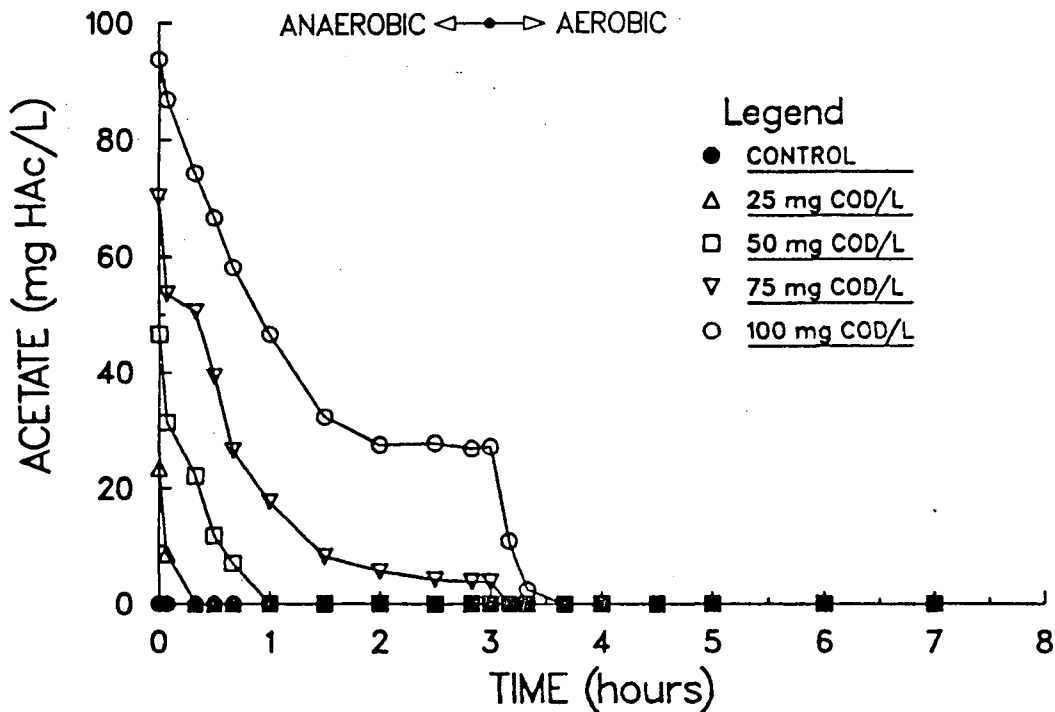
Fig. 4.4b. Acetate concentration profiles: The effect of
substrate concentration on P release and
uptake - denitrified sludge (Section 4.2.2)
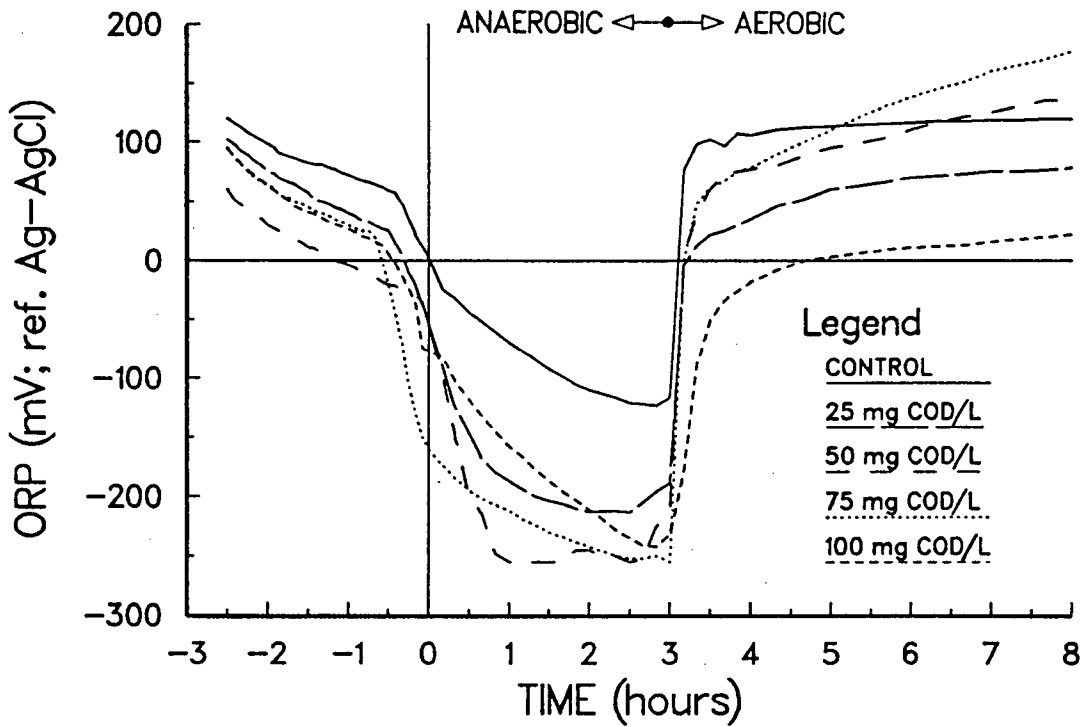
Fig. 4.4c. ORP profiles: The effect of substrate
concentration on P release and
uptake - denitrified sludge (Section 4.2.2)
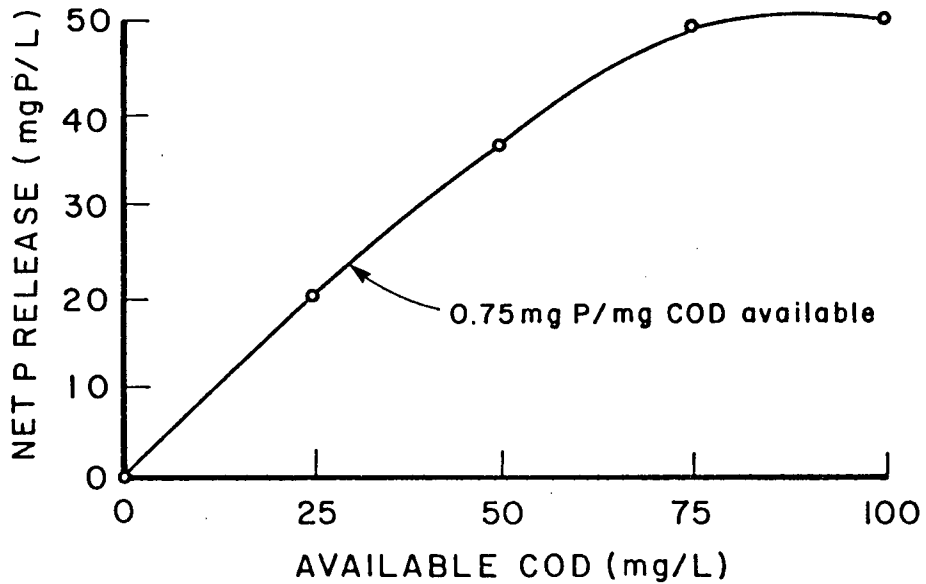
Fig. 4.5. The effect of increasing the initial substrate
concentration on the net P release after 2
hours in Section 4.2.2

stored P was available for release. From Fig. 4.4 it can be seen that once again the same 2-stage phosphorus release profiles were observed with the extent of the initial rapid release being a function of the initial substrate concentration. Furthermore, it can be seen that the ortho-P profiles for the 75 and 100 mg COD/L experimental flasks are almost identical, indicating that there exists a definite upper limit to the mass of P that can be released by a given sludge. Increasing the concentration of substrate available to the organisms beyond this limit does not result in additional P release. This upper limit is, in all likelihood, a function of the characteristics of the sludge used in the experiment. A total P release of 60 mg/L from a sludge with a MLSS concentration of 3550 mg/L represents a percent dry weight loss of 1.7%. Therefore, the cells returned to a dry weight % P content of 3.2 - 1.7 = 1.5%, i.e. the accepted P content of cells in which there is no excess biological P accumulation (Hoffmann and Marais, 1977). Thus, given sufficient substrate and a long enough period of anaerobiosis, cells that have accumulated excess P will release all of this stored P to the supernatant. A graph of net P release (i.e. experimental flask P release minus control flask P release) after two hours for each of the experimental flasks plotted against the available COD concentration at the start of the experiment is shown in Fig. 4.5. The graph shows a linear relationship between P release and available substrate concentration with a slope of about 0.75 mg P/mg COD for lower substrate concentrations. At higher available substrate concentrations the plot becomes horizontal at

a higher net release of 50 mg P/L. The acetate concentration profiles in Fig. 4.4 present some important clues regarding the interrelationship between substrate utilization and P release. The HAc profiles for all the experimental flasks show that substrate utilization by the cells (as represented by the disappearance of sodium acetate from the supernatant)˙occurs during the rapid P release phase at a rate that is independent of the initial substrate concentration. Furthermore, the cessation of substrate utilization is coincident with the end of the rapid release phase. In the case of the 25 and 50 mg COD/L experimental flasks this occurs due to the onset of substrate limiting conditions in the flasks, but in the case of the 75 and 100 mg COD/L flasks the cells appear to have lost the ability to take up the remaining substrate. It seems likely, therefore, that P release plays an integral role in the transport of the substrate from the supernatant into the cell and that the interrelationship of these two phenomena warrants closer examination. A plot of the P release vs. the corresponding substrate utilization during the rapid release phase for time intervals of 10, 20, 30, 40, 60, 90 and 120 minutes is shown in Fig. 4.6. There is an excellent correlation (r = 0.989) between the two phenomena indicating that there exists some direct or indirect exchange of the two molecules across the cell wall under anaerobic conditions. The least squares line drawn through the points has a slope of 0.91 indicating that 0.91 mg of P are released for every mg of HAc taken up by the cells. Converting this ratio to the molar form:

$$= 0.91 \ (60/30.94)$$

$$= 1.76 \ \text{moles P/mole HAc}$$

i.e. 1.76 moles of P are released for every mole of HAc taken up by the cells. This observation, together with its relevance to the concept of specific substrate induced excess biological P removal in the activated sludge process, is dealt with in greater detail in Chapter Seven. From Fig. 4.6 it can be seen that the line drawn through the points does not pass through the origin but intersects the HAc axis at about 4 mg/L. Two possible explanations for this phenomenon are that a small mass of substrate is adsorbed onto the sludge or experimental apparatus, or that there exists some small metabolic requirement that is independent of the P release mechanism.

From the ORP profiles presented in Fig. 4.4, it can be seen that ORP measurement was used during the pre-experimental sludge denitrification phase in order to determine when the zero nitrate condition had been reached. Slope changes in the profiles, similar to those observed in the previous experiment, occurred during a period of approximately 10 minutes prior to the start of the experiment, probably indicating that a zero nitrate condition had been reached. During the course of the experiment, however, there appears to be no specific trend in the ORP measurements. Although the experimental flasks had consistently more negative ORP readings than the control flask during the anaerobic phase, this may have been a function of the electrode used in the control flask as the control readings were also less negative during the pre-experiment denitrification phase.
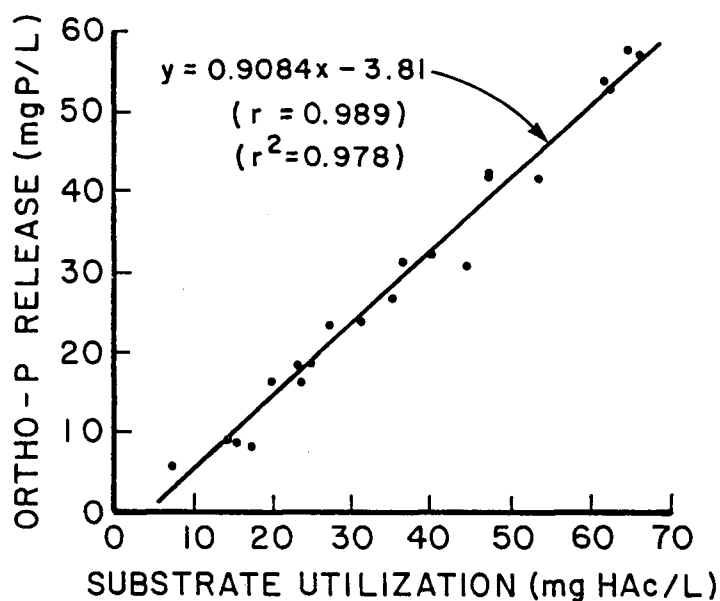
Fig. 4.6. Phosphorus release versus substrate utilization in Section 4.2.2
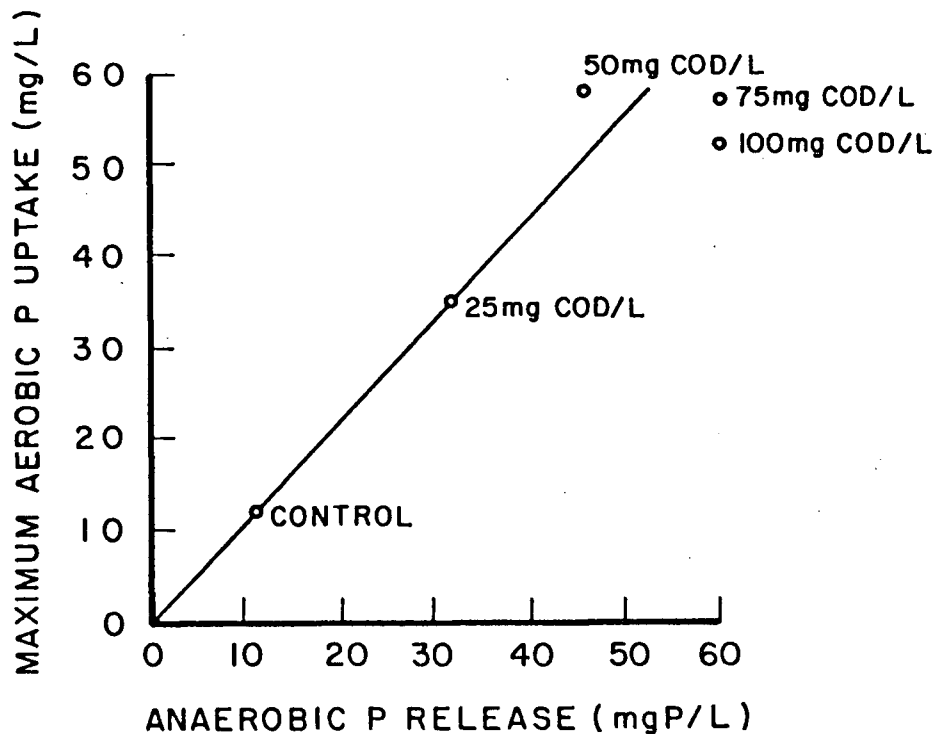
Fig. 4.7. Aerobic P uptake versus anaerobic P release in Section 4.2.2

Results for the aerobic phase were, in general, less conclusive than those of the anaerobic phase. The ortho-P profiles in Fig. 4.4 show that although the flasks exhibited a significant degree of P uptake under aerobic conditions, the control and the 25 and 50 mg COD/L experimental flasks (in that order) began to release P back into the supernatant during the course of the aerobic phase. A possible explanation may lie in the experimental procedure itself - the long anaerobic time period between the drawing of the mixed liquor from the pilot plant and the onset of aerobic conditions, 6 hours, may have caused a significant degree of cell lysis under the aerobic conditions (where substrate limiting conditions prevailed), which would result in the re-release of the stored P back into the supernatant. In spite of this observation, there does appear to be a loose correlation between the mass of P released under anaerobic conditions and the mass of P taken up under subsequent aerobic conditions (See Fig. 4.7). However, the aerobic phase of the ortho-P profiles shown in Fig. 4.4 show that there appears to be an upper limit to the P uptake capacity of the cells of a given biomass under batch testing conditions.

4.3. The Effect of Varying the Level of Nitrate for a Given
     Initial Substrate Concentration on Anaerobic P Release and
     Subsequent P Uptake in Activated Sludge.

4.3.1. Experiment Using UBC Pilot Plant Sludge and Excess
       Substrate.

In Section 4.2 it was shown that the presence of nitrate at the start of an anaerobic batch test appears to have a negative effect on the P release mechanism due to the substrate

requirement for denitrification. In addition, the nitrate
entering the anaerobic zone of full-scale activated sludge
processes is known to have a confounding effect on the P release
mechanism and on the P removal characteristics of the processes.
The objective of this experiment was to quantify the effect of
the presence of nitrate in the flasks at the start of a batch
test on the anaerobic P release and aerobic P uptake mechanisms.
Sludge used in this experiment was drawn from a 3-stage anoxic/
anaerobic/aerobic process that was receiving a continuous sodium
acetate addition of 37 mg COD/L and exhibiting excess biological
P removal. The process and sludge characteristics were as
follows:

Sludge age = 20 days

MLSS = 3900 mg/L

P removal = 3.84 - 0.57 = 3.27 mg P/L

COD removal = 266 + 37 - 30 = 273 mg COD/L

Sludge dry weight percent P = 3.4%

Prior to the start of the batch experiment, the sludge was
allowed to denitrify completely so that the initial nitrate
concentration could be controlled. In order that P limiting
conditions did not occur during the aerobic phase, the flasks
were each spiked with 24 mg/L of P. At the start of the
experiment, the control flask and each of the experimental flasks
received an initial sodium acetate substrate concentration of 100
mg/L (as COD). In addition, the four experimental flasks
received 3, 6, 9 and 12 mg/L of nitrate (as N), respectively, by
injecting the appropriate volumes of a stock sodium nitrate

solution. The ortho-P, acetate and nitrate concentration
profiles for the five flasks are presented in Fig. 4.8. From the
ortho-P concentration profile it can be seen that the P release
mechanism was virtually unaffected by the initial nitrate
concentrations of 3, 6 and 9 mg N/L. In the flask with the
initial nitrate concentration of 12 mg N/L, P release was slightly
inhibited with the final concentration of P released being
approximately 20% lower than in the other flasks. The nitrate
concencentration profiles show that denitrification occurred at a
steady rate that was virtually identical for all the flasks. The
VFA profiles in Fig. 4.8 for the control flask and the 3 mg N/L
experimental flask show that the substrate was not limiting in
these flasks for the duration of the anaerobic phase. In the
case of the 6 and 9 mg N/L experimental flasks, substrate became
limiting after complete denitrification had taken place. Only in
the case of the 12 mg N/L flask did the substrate concentration
reach zero prior to the completion of denitrification.
Furthermore, there appears to be a significant decrease in the
substrate utilization rate that is coincidental with the nitrate
concentration reaching zero. It appears, therefore, that the
presence of up to 9 mg/L of nitrate had virtually no effect on
the P release mechanism and that it may be possible to explain
this phenomenon by the fact that an excess concentration of
substrate was present in the flasks at the start of the
experiment.
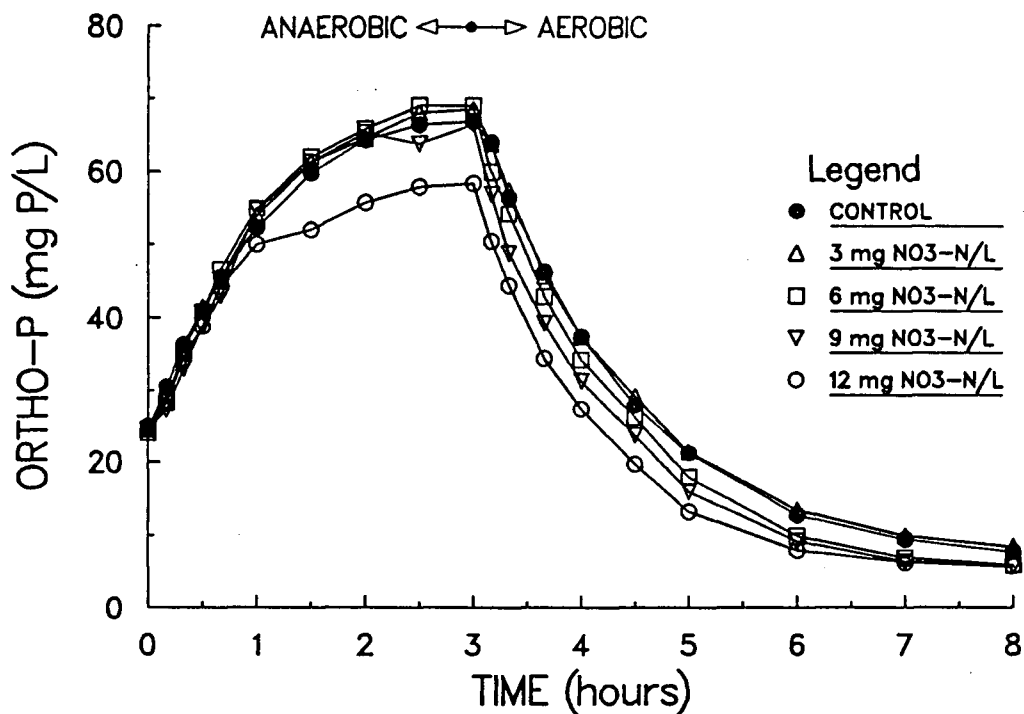
Fig. 4.8a. Ortho-P concentration profiles: The effect
of nitrate on P release and uptake - excess
substrate conditions (Section 4.3.1)
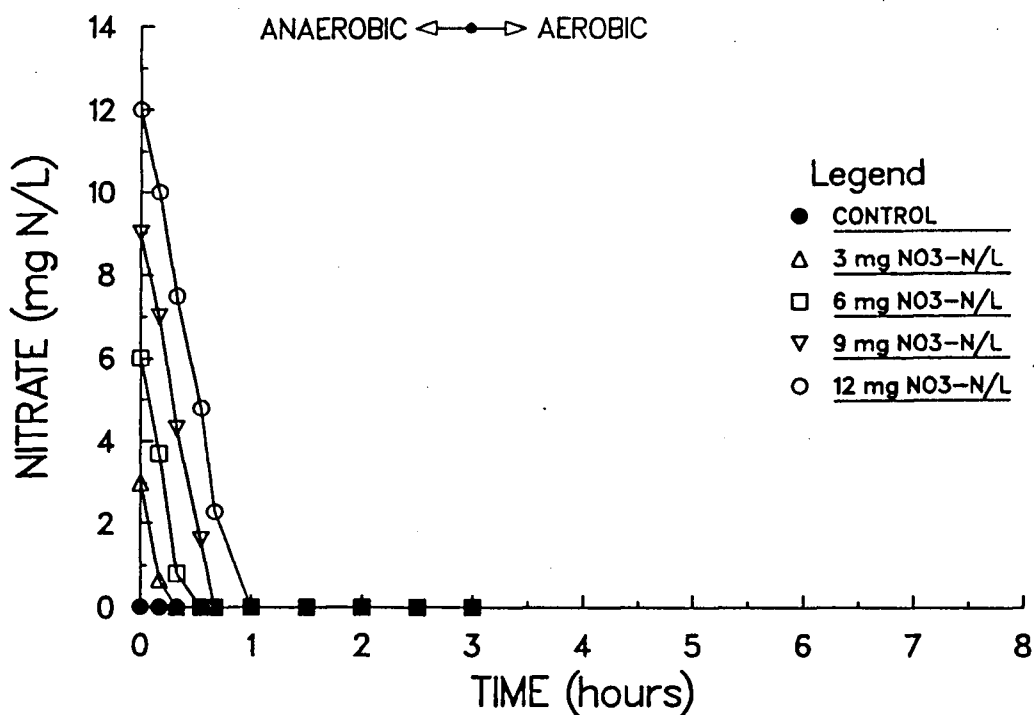
Fig. 4.8b. Nitrate concentration profiles: The effect of
nitrate on P release and uptake - excess
substrate conditions (Section 4.3.1)
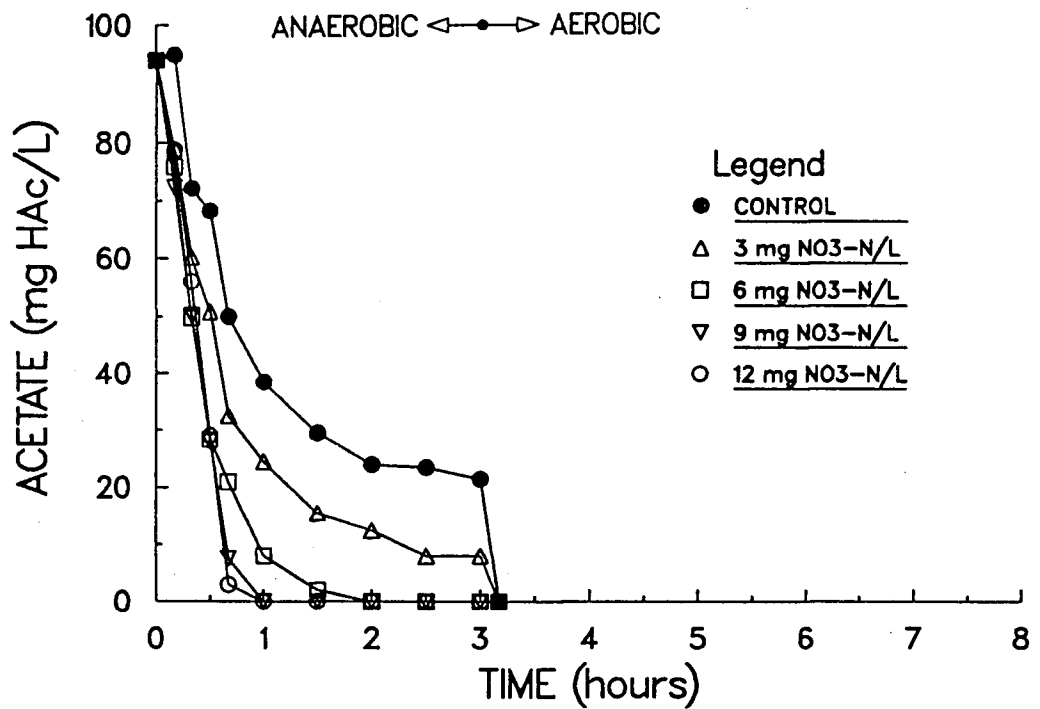
Fig. 4.8c. Acetate concentration profiles: The effect of
nitrate on P release and uptake - excess
substrate conditions (Section 4.3.1)

The results of this experiment suggest that denitrification and the P release mechanism are substrate utilization mechanisms that are not mutually exclusive. For example, the denitrification kinetic theory of van Haandel et al. (1981) is based on a stoichiometric relationship in the oxidation of substrate using nitrate as the final electron acceptor. The model assumes that 8.6 mg of substrate (as COD) are required in the reduction of 1 mg of nitrate (as N). According to the theory of Siebritz et al. (1983), substrate that is utilized for denitrification in the anaerobic reactor is rendered unavailable for the excess biological P removal mechanism in processes designed for enhanced biological P removal. However, in this experiment it seems unlikely that there was preferential usage of the available acetate in the denitrification reaction, i.e. that substrate utilized for denitrification was rendered unavailable for the P release mechanism. For example, in the case of the 9 mg N/L experimental flask, the hypothetical substrate requirement for denitrification was 9.0 X 8.6 = 77 mg COD/L, according to the theory of van Haandel et al. This implies that only 23 mg COD/L was available for the P release mechanism. This seems unlikely when one considers that P release was virtually unaffected by the initial presence of up to 9 mg/L of nitrate in the experimental flasks. However, it is possible that the substrate that is first taken up by the cells in the P release mechanism, is stored intracellularly and later utilized in the denitrification reaction; this would allow the same substrate to perform two functions in a given experimental flask. This situation is

unlikely to occur in a single zone of a continuous flow-through process. This aspect will be dealt with in greater detail in the presentation of an integrated model for anaerobic P release in Chapter Seven of this thesis. An additional question that is raised by the results of this experiment is whether or not the presence of the same initial concentrations of nitrate at the start of the experiment would have had a more detrimental effect on the anaerobic P release mechanism in a case where the initial substrate concentration in the flasks had been significantly lower, i.e. had substrate limiting conditions prevailed in the flasks for a greater period of time. The question of the effect of nitrate on the P release mechanism under limited substrate availability is addressed in the following experiment.

4.3.2. Experiment Using UBC Pilot Plant Sludge and Limited Substrate

The results of the previous experiment show that when excess simple substrate is available, anaerobic P release is not severely inhibited by the presence of nitrate in concentrations likely to be found in a nitrifying nutrient removal process. The objective of this experiment was to quantify the effect of the same range of initial nitrate concentrations on the P release mechanism under substrate limiting conditions. At the time of the experiment, the pilot plant was operating under conditions similar to those of the previous experiment, except that the continuous sodium acetate addition was reduced to 28 mg/L (as COD). The process and sludge characteristics were as follows:

Sludge Age = 20 days

MLSS = 4365 mg/L

P Removal = 5.3 - 0.8 = 4.5 mg P/L

COD Removal= 309 + 28 - 24 = 313 mg COD/L

Sludge dry weight percent P = 3.0%

The initial nitrate concentrations in the experimental flasks
were kept at 3, 6, 9 and 12 mg N/L, respectively, and the initial
sodium acetate dosage was reduced from 100 to 50 mg/L (as COD).
The ortho-P, nitrate and acetate profiles are presented in Fig.
4.9. From the ortho-P profiles in Fig. 4.9, it can clearly be
seen that the presence of nitrate in the flasks at the beginning
of the experiment had a significantly greater negative effect on
the P release mechanism than in the previous experiment. The
ortho-P profiles for the control and the 3 and 6 mg N/L
experimental flasks show similar release trends to those observed
in Section 4.2.2, where no nitrate was present at the start of
the experiment, i.e. a rapid initial release rate followed by
a slower release rate which continued for the remainder of the
anaerobic time period. The magnitude of the initial P release
was adversely affected by the initial presence of 3 mg/L of NO_3-N
and, to a greater extent, by the presence of 6 mg/L of NO_3-N.
This effect was similar in fashion to the way in which the
magnitude of the initial release phase was determined by the
initial substrate concentration in Section 4.2.2. The ortho-P
profiles for the 6 and 9 mg/L of nitrate experimental flasks
demonstrate an interesting phenomenon previously observed
in Section 4.2.1 - after the rapid initial P release, P uptake
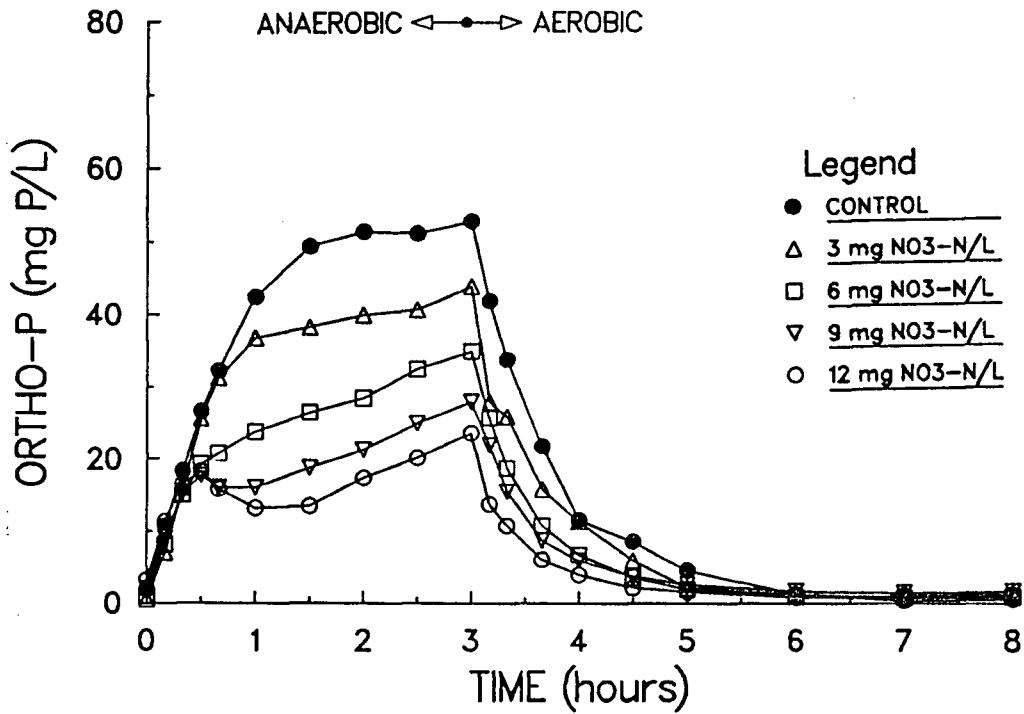
Fig. 4.9a. Ortho-P concentration profiles: The effect of nitrate on P release and uptake - substrate limiting conditions (Section 4.3.2)
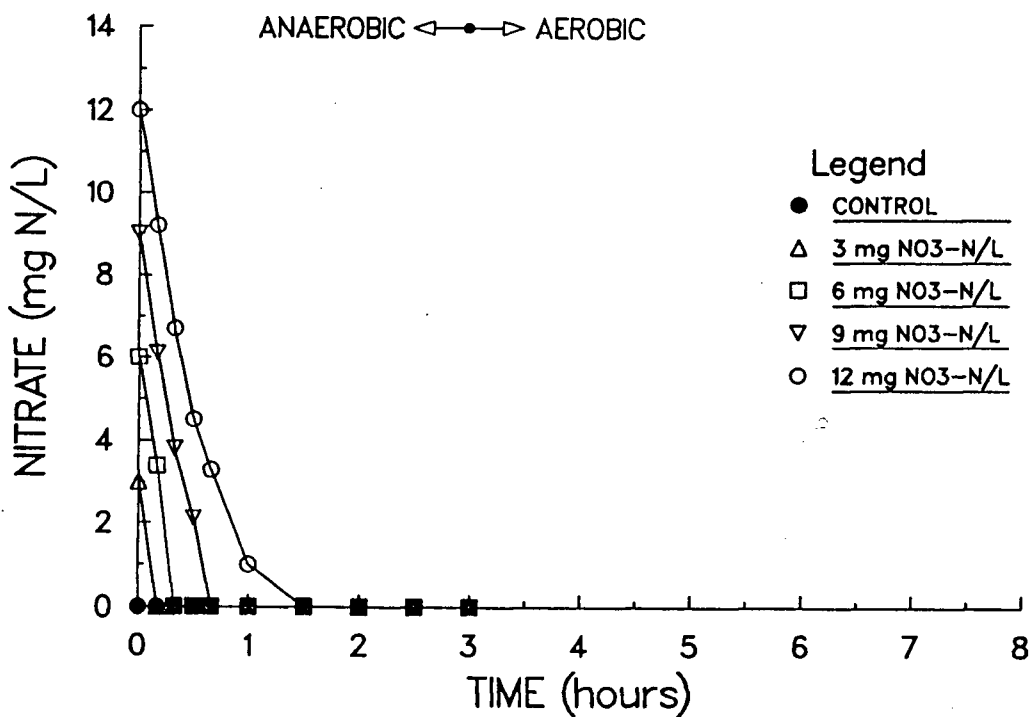
Fig. 4.9b. Nitrate concentration profiles: The effect of nitrate on P release and uptake - substrate limiting conditions (Section 4.3.2)
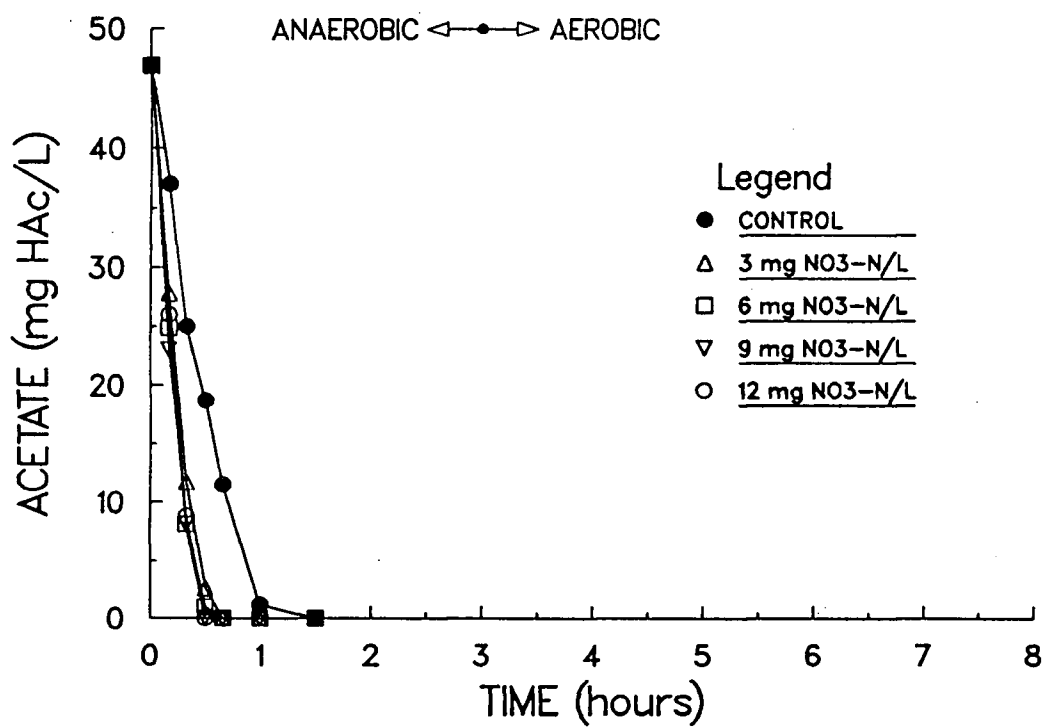
Fig. 4.9c. Acetate concentration profiles: The effect of
nitrate on P release and uptake - substrate
limiting conditions (Section 4.3.2)

occurs before the slower release rate commences. Examination of nitrate and acetate profiles in Fig 4.9 indicate that for these two reactors, the end of the rapid P release phase is coincident with the substrate concentration reaching zero but that nitrate is still present in solution, i.e. complete denitrification has not yet occurred. P release only resumes at the slower rate once all the nitrate has disappeared from solution. This seems to suggest that some of the substrate stored by the organisms during the rapid P release phase is utilized by the organisms for denitrification. Such denitrification in the absence of substrate being available from the solution is characterized by a net uptake of P.

In order to quantify the negative effect of nitrate on P release, P release after three hours is plotted against the initial nitrate concentration in Fig. 4.10. It can be seen from the graph that the relationship appears to be linear for nitrate concentrations up to about 6 mg N/L with a loss of 3.0 mg/L of P release for every mg N/L of nitrate initially present at the start of the experiment. For initial concentrations of nitrate greater than about 6 mg N/L, the negative effect of the presence of nitrate tends to decrease slightly.

The relationship between the substrate utilized and the corresponding P release in the control reactor (no nitrate addition) is shown in Fig. 4.11. The graph shows a linear relationship with a slope of 0.89 which is within 2% of the slope obtained for the same two variables in Section 4.2.2 (See Fig.
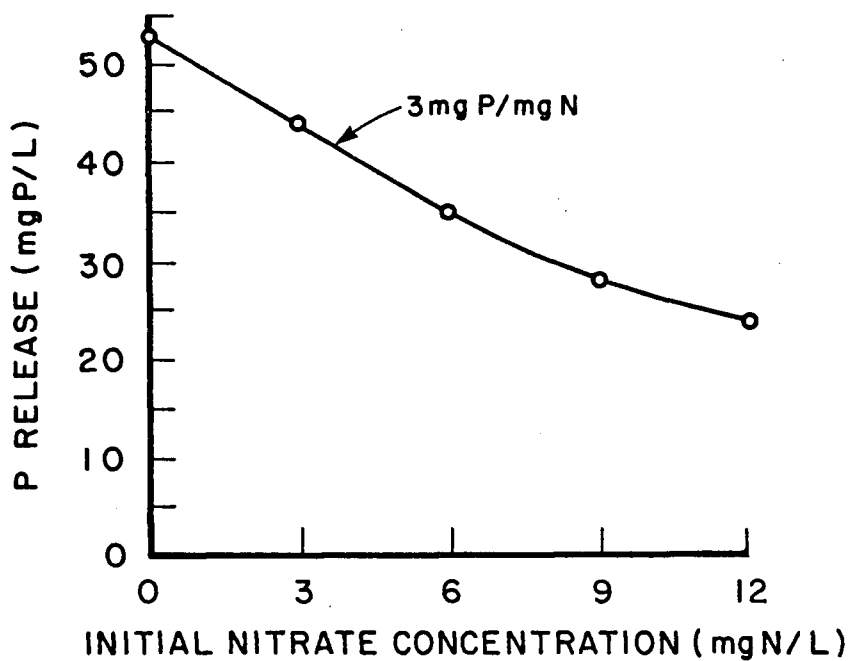
Fig. 4.10. Effect of increasing the initial nitrate
concentration on P release in Section 4.3.2
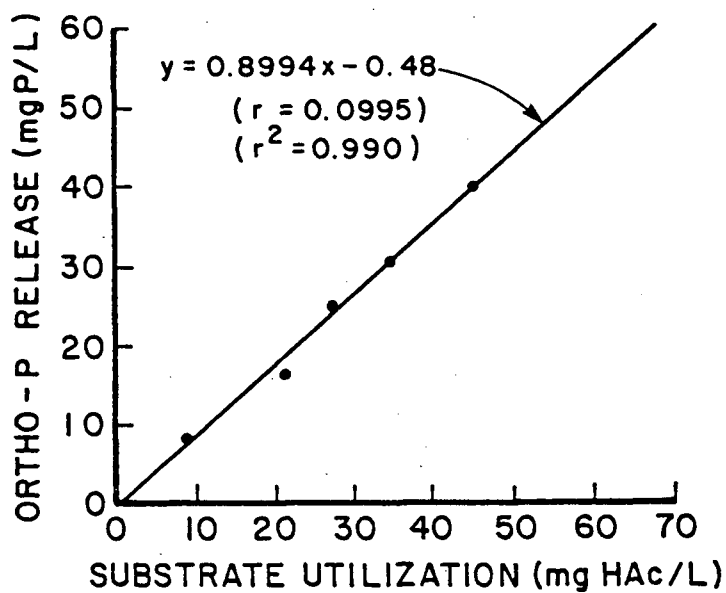
Fig. 4.11. Substrate utilization versus P release for the
control reactor in Section 4.3.2

4.6). This confirms that for every mg acetate (as HAc) lost from solution in the absence of nitrate, 0.89 mg of P are released. This represents a molar ratio of:

$$0.89 \ (60/30.94) = 1.75 \ \text{moles P/mole HAc}$$

On the other hand, denitrification theory of van Haandel et al. (1981) states that 8.08 mg of HAc (1 mg COD = 0.94 mg HAc) are required to denitrify 1 mg of NO_3-N. This, in turn, represents a molar ratio of:

$$8.08 \ (14/60) = 1.89 \ \text{moles HAc/mole N}$$

By combining the two ratios, the theoretical detrimental effect of the presence of nitrate on P release (assuming preferential use of the substrate in the denitrification reaction) can be made as follows:

Change in P Release/N = 1.75 X 1.89 = 3.31 moles P/mole N

Expressing this in the mass concentration form:

$$3.31 \ (30.94/14) = 7.31 \ \text{mg P/mg N}$$

which is more than double that shown for this experiment in Fig. 4.10 (a maximum of 3.0 mg P/mg N).

The results of this experiment show that when the substrate available to the organisms is limiting, the presence of increased concentrations of nitrate has an increasingly negative effect on P release. However, this negative effect is clearly not as great as predicted by denitrification theory, assuming that the substrate utilized in the denitrification reaction is rendered unavailable for P release. This confirms the findings of Section

4.3.1, that the two phenomena are clearly not mutually exclusive, since the same substrate may be utilized in both reactions simultaneously. As in the case of the previous experiment, it appears that substrate that is stored intracellularly under anaerobic conditions can later be utilized in the denitrification reaction, should nitrate still be present in the flask after all the available substrate in the supernatant is utilized.

With regard to the aerobic P uptake in this experiment, it can be seen from Fig. 4.9 that there were no clear differences observed between the various flasks and that P became limiting after about three hours of aeration. The relationship between P release and uptake in batch tests of this nature, and its relevance to P removal in a continuous process requires further investigation.

CHAPTER FIVE

PILOT-SCALE SUBSTRATE ADDITION

The objective of the five experiments detailed in this chapter
was to determine to what extent the P removal characteristics of
the pilot scale nutrient removal activated sludge process could
be enhanced by the addition of a sodium acetate solution directly
into the process. Phosphorus removal efficiency was measured by
the $\Delta P/\Delta COD$ ratio and the dry weight percent P content of the
sludge. It was assumed that the dry weight percent P content of
organisms not exhibiting any excess biological P removal is 1.5%
(Hoffmann and Marais, 1977).

5.1. Various Levels of Sodium Acetate Addition to a Simplified Nutrient Removal Activated Sludge Process Configuration

In the experiments reported in this section the process used had
a simplified unaerated/aerated configuration. The unaerated
fraction made up 30% of the process volume and was further
divided into two zones of approximately equal size. The first
unaerated zone received the raw sewage, the return sludge
(recycle rate 2:1 with respect to the influent flow rate) and the
sodium acetate feed solution. The process sludge age was
hydraulically controlled at 20 days by wasting the appropriate
volume of mixed liquor daily from the aerated zone. Section
5.1.1 served as the control period, during which no sodium
acetate was added to the bioreactor. In Sections 5.1.2, 5.1.3
and 5.1.4 the process received sodium acetate additions of 56, 86
and 112 mg/L (expressed as mg COD/L of influent flowing into the

process), respectively, which was fed continuously into the process. Influent and effluent total P and TKN, and influent and added COD analyses were done on daily 24-hour composite samples, and effluent and COD analysis was done on twice-weekly 24-hour composite samples for the entire duration of each experiment. Ortho-P and nitrate analyses were done twice weekly on the mixed liquor using filtered grab samples drawn from each reactor of the process. Raw data from this series of experiments is listed in Appendix A1 and the mean values of the results of each section are presented in Table 5.1.

The results presented in Table 5.1 for Section 5.1.1 indicate that when no acetate was added to the simplified process, the P removal efficiency was 0.0069 mg P/mg COD and the dry weight percent P content of the sludge was 1.46%. This value is close to the accepted value for organisms not exhibiting excess biological P removal. A slight improvement in removal efficiency, from 0.0066 to 0.0071 mg P/mg COD, was achieved by the addition of 56 mg/L of COD to the process in Section 5.1.2. However, in Sections 5.1.3 and 5.1.4 in which dosages of 86 and 112 mg COD/L were added to the process, removal efficiency improved to approximately 0.01 mg P/mg COD, an improvement of about 50%. The mean dry weight percent P content of the sludge increased from around 1.5% to over 2.1% for these two sections. The results of these two sections confirm the findings of Siebritz et al. (1983), that a certain basic minimum concentration of "readily biodegradable" substrate is required to be available to the organisms in the anaerobic zone of the

Table 5.1. Results of Pilot-Scale Substrate Addition Experiments

Parameter	Section No.				
	5.1.1 (Control)	5.1.2	5.1.3	5.1.4	5.2
Influent Total P (mg/L)	4.02	3.87	3.32	3.06	4.01
Ortho-P:					
1st Unaerated (mg/L)	2.50	2.72	5.37	6.41	0.94
2nd Unaerated (mg/L)	2.76	3.42	7.26	7.51	10.65
Aerobic (mg/L)	2.42	2.13	0.69	0.42	0.27
Effluent (mg/L)	2.42	2.07	0.49	0.30	0.25
Effluent Total P (mg/L)	2.62	2.23	0.73	0.32	0.28
ΔP (mg/L)	1.40	1.64	2.59	2.74	3.73
% P Removal	34.8%	42.4%	78.0%	89.5%	93.0%
Influent COD (mg/L)	253	247	226	203	268
Added COD (mg/L)	0	56	86	112	39
Effluent COD (mg/L)	50	71	59	39	44
ΔCOD (mg/L)	203	232	253	276	263
$\Delta P/\Delta$COD	0.0069	0.0071	0.0102	0.0099	0.0142
Influent TKN (mg/L)	21.02	20.86	21.42	18.64	26.00
NO_3-N (mg/L):					
1st Unaerated (mg/L)	1.27	0.15	0.12	0.03	0.25
2nd Unaerated (mg/L)	0.14	0.09	0.09	0.08	0.12
Aerobic (mg/L)	7.43	4.99	3.87	3.88	5.93
Effluent (mg/L)	7.56	3.72	3.94	3.48	5.89
Effluent TKN (mg/L)	-	2.23	1.49	1.12	1.41
% N Removal	57.0%	71.5%	74.6%	75.3%	71.9%
Process Temp. (^{o}C)	18.5-21.0	20.0-21.5	12.5-14.5	17.5-21.5	10.0-16.0
Aerobic MLSS (mg/L)	2783	2791	2732	3033	3105
Sludge Dry Wt. % P	1.46%	-	2.34%	2.06%	2.54%

process to ensure excess biological P removal. According to their theory, 25 mg/L of readily biodegradable COD (measured as mg of COD/L of influent flow) must enter the anaerobic reactor, taking into account the dilution effect of any recycles entering the reactor. In Sections 5.1.1-5.1.4 the sludge return recycle was 2:1 with respect to the influent flow rate, meaning that a readily biodegradable COD concentration of 3X25 = 75 mg/L in the raw sewage is required. Furthermore, the theory of Siebritz et al. also states that approximately 24% of the biodegradable COD (or about 20% of the total COD) present in South African wastewaters is in the readily biodegradable form. If these fractions are applicable to North American wastewaters, there should have been approximately 40 mg/L of readily biodegradable COD present in the influent sewage in these experiments and the added sodium acetate dosages should have been supplementary to the preferred substrate concentration already present in the wastewater. However, the minimum readily biodegradable substrate concentration required for excess biological P removal to occur, as determined by Siebritz et al., is based on the assumption that the nitrate concentration entering the anaerobic zone is zero. Should any nitrate enter the zone, according to Siebritz et al. (1983), the readily biodegradable substrate is utilized preferentially in the denitrification reaction at a rate of 8.6 mg COD/mg N and is thus rendered unavailable for the excess biological P removal mechanism. This preferential use of the readily biodegradable COD in the denitrification reaction was the main motivation for the UCT process configuration, as described

in Chapter Two, in which a zero nitrate discharge to the anaerobic reactor can be ensured by using the operational flexibility of the process. The batch test results presented in the previous chapter (Section 4.3) confirm that the presence of nitrate in concentrations likely to be found in a nitrifying activated sludge process is inhibitory to the anaerobic P release mechanism, particularly in cases where the availibility of substrate is limited. In Sections 5.1.2-5.1.4 the nitrate concentration in the effluent (and therefore in the return sludge recycle) was in the 3.5-5.5 mg N/L and the sludge recycle ratio was maintained at 2:1 with respect to the influent flow. Assuming that the substrate was consumed in the denitrification reaction at a rate of 8.6 mg COD/mg N, between 60 and 95 mg/L of substrate (as COD) was required to denitrify the mass of nitrate entering the anaerobic zone via the return sludge recycle. Therefore, it is clearly important to discharge any substrate that is added to the process into a zone that receives a zero nitrate discharge.

With regard to biological N removal, the results of Section 5.1 indicate increasing N removal efficiency with increasing sodium acetate dosages even though the process configuration used in this series of experiments was not specifically designed to optimize N removal. The most probable explanation is that by incrementally increasing the acetate dosage, the influent TKN/COD ratio was decreased (viz. 0.0831, 0.0688, 0.0687 and 0.0592, respectively), thereby improving the influent characteristics for biological N removal. Furthermore, in addition to being a

preferred substrate for excess biological P removal, acetate is
also a preferred substrate for biological N removal, with
denitrification rates using acetate as a substrate being
much higher than those achieved with the more complex organic
substrates.

The supernatant ortho-P concentrations in the various zones of
the process show that the degree of P release in the unaerated
zone is a function of the acetate available to the organisms
present in the zone, with increasing release being observed with
increasing acetate dosages. This phenomenon confirms the
findings of Section 4.2 of the previous chapter, that the degree
of anaerobic P release is a function of substrate availability
and that there appears to be an exchange phenomenon that occurs
between the acetate and the phosphate molecules under anaerobic
conditions. The results of Section 5.1, therefore, confirm the
findings of many researchers (Barnard (1974, 1975, 1976),
Rabinowitz and Marais (1980) and Siebritz et al. (1983)) that the
P removal characteristics of the activated sludge process are
improved with increasing degrees of anaerobic P release.

5.2. The Effect of Feeding Sodium Acetate to a Zone that Receives a Zero Nitrate Discharge

The objective of this section was to determine to what extent the
P removal characteristics of a simplified nutrient removal
activated sludge process receiving sodium acetate can be improved
by discharging the added substrate to a zone that received no
influent nitrate concentration. To this end the sodium acetate
solution was fed into the second unaerated zone, rather than the

first, of a process having its unaerated zone at the head end of
the process further divided into two zones of equal size. In
this way, it was reasoned, complete denitrification would occur
in the first unaerated zone making use of the readily
biodegradable COD and the more complex organics present in the
incoming wastewater, and the nitrate concentration entering the
second unaerated zone would, therefore, be zero. This would
facilitate a more efficient use of the added substrate, as all of
it would be available for the excess biological P removal
mechanism. In addition, the following two changes were made to
the process configuration - the unaerated volume fraction was
increased to 40% but was still equally divided into two zones of
approximately equal size, and the sludge recycle ratio was
decreased from 2:1 to 1:1 in order to minimize the mass of
nitrate being recycled to the unaerated zone. The results of
this experiment are also included in Table 5.1 for the purposes
of comparison.

The results of this experiment clearly show that a significantly
more efficient use was made of the added acetate solution by
feeding it into a zone that received little or no influent
nitrate. A continuous substrate addition of 39 mg/L (as
mg COD/L of influent flowing into the process) into the second
unaerated zone resulted in an overall P removal efficiency of
0.0142 mg P/mg COD. This level of substrate addition is about
40% less than the dosage used in Section 5.1.2 (56 mg COD/L)
where no significant improvement in P removal efficiency was
achieved by substrate addition. The removal efficiency achieved

in this experiment is more than 100% greater than that achieved
in the control, (Section 5.1.1) and 40% greater than that
achieved in the two higher substrate addition experiments,
reported in Sections 5.1.3 and 5.1.4.

With regard to biological N removal, the N removal efficiency of
Section 5.2 is similar to that achieved in Section 5.1.2 in which
a comparable acetate dosage was fed into the process. However,
in this experiment the effluent nitrate concentration was
approximately 60% higher than that in the other three experiments
in which the process received acetate addition. There are two
possible explanations for the increased effluent nitrate
concentration. Firstly, the mean influent TKN concentration in
this experiment was approximately 30% higher than in the other
three experiments and secondly, a lower mass of nitrate was being
recycled to the unaerated zone via the reduced sludge recycle,
meaning that the denitrification capacity of the process was not
as efficiently utilized.

The ortho-P concentration profiles of the various zones presented
in Table 5.1 confirm the more efficient use of the added acetate
in the process for the excess biological P removal mechanism.
The mass of P released by the end of the unaerated zone was
greatest in this experiment in spite of the fact that the added
acetate was lowest in this experiment and all of this release
took place in the second unaerated zone only. All of the factors
outlined above clearly demonstrate the advantage of feeding any
additional substrate, whether it be in the synthetic form or

generated on-site by primary sludge fermentation, directly into
an unaerated zone that receives no influent nitrate. However,
additional research is still required to determine what
percentage of micro-organisms invovled in the excess biological P
removal mechanism are also capable of denitrification. This
would facilitate quantification of the relative mass of substrate
utilized for excess biological P removal only, for
denitrification only, and in both mechanisms simultaneously.

CHAPTER SIX

PRIMARY SLUDGE FERMENTATION RESULTS

In this chapter the results of a pilot scale study into short-chain volatile fatty acid (VFA) production by primary sludge fermentation are detailed. The principle objective of the study was to determine the amount of VFA's that can reasonably be produced from primary sludge, and the distribution of the acids that make up the production. The three main operational parameters controlling anaerobic fermentation, sludge age or mean solids retention time, temperature and pH, were investigated. Section 6.1 describes a detailed study into the effect of sludge age in the 2.5-10 day range at ambient temperatures between 16 and 20°C. Sections 6.2 and 6.3 describe a somewhat less detailed study into the effects of temperature and pH.

The fermentation unit used in this series of experiments consisted of a 540 L primary clarifier and a two-stage anaerobic sludge fermenter with a total volume of 1000 L. Primary sludge was pumped on a half-hourly cycle from the bottom of the primary clarifier into the first fermentation reactor. The underflow pumping rate was regulated between 6 and 10% of the influent flow rate by adjusting the length of the "ON" cycle of the primary sludge pump. The primary sludge COD loading entering the fermenter was calculated on the basis of a COD mass balance on the primary clarifier, because of the variation in the organic strength of the primary sludge over the course of the day and the difficulties associated with measuring the COD value of the

primarily particulate material. The calculation was based on the difference between the mass of COD entering the primary clarifier in the raw sewage and that leaving the clarifier via the settled sewage, i.e. it was assumed that there was no COD destruction in the primary clarifier and that all the COD entering the clarifier left either via the settled sewage or the primary sludge. Raw influent and settled sewage COD analysis was done on daily 24-hour composite samples and VFA analysis was done on the filtered supernatant of grab samples drawn daily from the fermentation reactors. VFA production was compared on the basis of a yield factor which was defined as the ratio of the daily mean total VFA produced (as HAc) per unit mass of primary sludge (as COD) entering the fermenter for each study period.

6.1. The Effect of Sludge Age on VFA Production

The objective of this series of experiments was to investigate the effects of sludge age on VFA production. An attempt was made to maintain the fermenter temperature close to $20^{o}C$ by the use of space heating during the winter months. The pH was not controlled in any way, except that about 100 mg/L of alkalinity (as $CaCO_3$) was added to the raw influent daily in order to improve its buffering capacity and thus maintain the pH of the activated sludge processes above 6.5. During the first three experiments the sludge age was regulated at 2.5, 3.5 and 5.0 days, respectively, by adjusting the pumping rate from the primary clarifier to the fermenter, with both fermentation reactors operating under completely mixed conditions (i.e. sludge age equal to the hydraulic retention time). In the fourth

experiment a secondary clarifier, from which settled, fermented sludge was returned to the first fermentation reactor at a recycle rate of 2:1, was installed. In this experiment the sludge age was hydraulically controlled by wasting the appropriate volume of mixed liquor from the second fermentation reactor. The operational parameters and results of the four experiments are presented in Table 6.1 and the raw data are listed in Appendix A2. All of the values reported in Table 6.1 are the mean values for each experiment, with the exception of pH which is reported as the median values for each experiment.

The VFA results presented in Table 6.1 show the largest percentage of acid produced for all sludge ages to be in the acetic acid form, which comprised 57%, 56%, 53% and 56%, respectively, of the production. The second major VFA form produced was propionic acid which made up 41%, 43%, 43% and 42% of the acid production. In each case the balance of the acid produced was in the butyric acid form. No other short chain VFA's were present in detectable concentrations (>3 mg/L as HAc, using gas chromatography) in the fermenter supernatant. The fermenter sludge age did not appear to significantly affect either the yield of VFA produced from the primary sludge or the distribution of the various component acids in the 2.5-10.0 day range. Optimum yields were attained in the 3.5 and 5.0 sludge age experiments, with yields being slightly greater than 0.09 mg HAc/mg COD of primary sludge. However, even the lower acid production in the 2.5 day sludge age experiment could possibly be explained by the significantly lower mean fermenter operating

Table 6.1. The Effect of Sludge Age on VFA Production from
Primary Sludge (Section 6.1)

Parameter	Section No.			
	6.1.1	6.1.2	6.1.3	6.1.4
Sludge Age (days)	2.5	3.5	5.0	10.0
Influent (mg COD/L)	277	250	245	351
Sett. Sewage (")	179	140	151	187
Prim. Sludge (")	1142	1794	1718	1823
Underflow rate (% of raw sewage flow rate)	10.0%	6.67%	6.0%	10.0%
Temperature ($^\circ$C):				
Ferm. 1	15.7	18.1	20.8	19.7
Ferm. 2	15.7	19.6	20.8	20.9
MLSS (mg/L):				
Ferm. 1	1344	1961	1358	2310
Ferm. 2	2460	2139	2433	2432
Median pH:				
Ferm. 1	6.05	5.90	5.60	6.10
Ferm. 2	5.85	5.60	5.15	6.00
VFA Production (mg/L)				
Fermenter 1:				
Acetic acid (as HAc)	27.6	61.8	60.7	69.0
Propionic acid (")	14.7	43.9	44.6	47.3
Butyric acid (")	0.5	0.7	2.8	3.4
Total acid (")	42.8	106.4	108.1	119.7
Fermenter 2:				
Acetic acid (as HAc)	51.0	90.9	85.0	79.0
Propionic acid (")	37.4	70.1	69.2	59.1
Butyric acid (")	1.8	1.5	5.5	4.2
Total acid (")	90.2	162.5	159.7	141.7
Fermenter Clarifier:				
Total acid (as HAc)	-	-	-	151.0
Yield (mg HAc/mg COD)	0.0790	0.0906	0.0930	0.0828

temperature (15.7^{o} vs about $20^{o}C$) during this period. In general, the median pH values for all the experiments were in the 5.1-6.1 range, with the lower values being attained where VFA production was most efficient. The pH dropped by between 0.25 and 0.55 pH units between the two fermentation reactors in the first three experiments, in which there was no internal recycle. In the fourth experiment, where the fermented sludge was recycled from the clarifier to the first fermentation reactor, the median pH dropped by only 0.10 pH units between the two reactors. It should be noted that the MLSS concentration difference between the two reactors was also smallest in the 10 day sludge age experiment, probably also as a result of the sludge recycling.

The VFA yields achieved in these experiments appear to be below both the sodium acetate addition requirements found in the previous chapter, and the "readily biodegradable COD" requirement in the activated sludge process influent stream according to Siebritz et al. (1982) (i.e. 25 mg COD/L entering the anaerobic reactor). One possible way of improving fermenter efficiency is to increase the operating temperature of the fermenter. Microbiologists point to a doubling of microbial activity for every $5-10^{o}C$ increase in temperature for psychrophiles and, therefore, far greater VFA yields may be expected at the higher temperatures. In addition, the viability of fermenter pH control with the view to maximizing VFA production also requires examination. There are indications in the literature that the uncontrolled pH range at which this experiment was conducted (5.1-6.1) may prove not to be the optimal range for the

acid-forming bacteria. For example, Borchardt (1970) found that optimum acid production occurs at a pH of 6.95, with production dropping off significantly on either side of this optimum value.

6.2. The Effect of Temperature on VFA Production

The objective of this experiment was to determine to what extent the operating temperature of the fermenter affects the VFA production efficiency. As a result of the inflexibility of the pilot plant, it was only possible to raise the fermenter temperature to about $22^{o}C$ using space heating. For this reason, it was decided to do a cold temperature study over the winter months. The study period lasted approximately two months during which the fermenter was operated at a sludge age of 3.5 days with a mean fermenter temperature of $12.7^{o}C$. The results of the experiment are, therefore, compared with the results of Section 6.1.2 in which the sludge age was also kept at 3.5 days, but where the process temperature was raised to around $19^{o}C$ using space heating. The raw data from this experiment are shown in Appendix A2 and the mean experimental results are presented together with those of Section 6.1.2, which served as the control for this experiment, in Table 6.2.

The results presented in Table 6.2 indicate that when the mean operating temperature of the fermenter at a sludge age of 3.5 days was lowered by $6^{o}C$, the resultant drop in VFA production efficiency was about 20%, the yield dropping from 0.0906 to 0.0732 mg HAc/mg COD. This drop in efficiency is significantly less than that predicted by microbiologists for general anaerobic

Table 6.2. The Effect of Temperature on VFA Production from Primary Sludge (Section 6.2)

Parameter	Control	Experiment
Sludge Age (days)	3.5	3.5
Influent (mg COD/L)	250	265
Sett. Sewage (")	140	147
Prim. Sludge (")	1794	1910
Underflow rate (% of raw sewage flow rate)	6.67%	6.67%
Temperature (^{o}C):		
Ferm. 1	18.1	12.7
Ferm. 2	19.6	12.7
MLSS (mg/L):		
Ferm. 1	1961	3166
Ferm. 2	2139	3647
Median pH:		
Ferm. 1	5.90	5.80
Ferm. 2	5.60	5.65
VFA Production (mg/L)		
Fermenter 1:		
Acetic acid (as HAc)	61.8	52.5
Propionic acid (")	43.9	34.1
Butyric acid (")	0.7	0.7
Total acid (")	106.4	87.3
Fermenter 2:		
Acetic acid (as HAc)	90.9	77.8
Propionic acid (")	70.1	60.7
Butyric acid (")	1.5	1.2
Total acid (")	162.5	139.7
Yield (mg HAc/mg COD)	0.0906	0.0732

activity. However, a detailed study of the effects of operating temperatures greater than $20^{\circ}C$ is still required. In addition, the economics of operating a primary sludge fermenter in the mesophilic temperature range (around $37^{\circ}C$), a temperature range commonly used in the operation of anaerobic sludge digesters, also warrants closer examination as the costs involved may prove to be prohibitive. The lower fermenter operating temperature had no significant effect on the distribution of the component acids making up the VFA production, with acetic and propionic acids making up about 56% and 43%, respectively, at both temperatures.

6.3. The Effect of pH on VFA Production

In this experiment the operating pH of the fermenter reactors was raised to around neutrality, in order to determine if this would enhance the VFA production. It was reasoned that raising the pH might possibly improve the VFA yield for the following two reasons: firstly, a pH of 7.0 may, in fact, be a better pH for the acid-forming bacteria, as indicated in the literature (Borchardt, 1970), and the uncontrolled pH approach of Section 6.1 may have resulted in sub-optimal conditions due to product inhibition; secondly, the pK values for acetic and butyric acid are 4.76 and 4.83 at $25^{\circ}C$, respectively, meaning that at the higher pH values, a significantly smaller fraction of the acids produced are in the molecular form from which volatilization is possible, i.e. the produced VFA is largely in the more stable salt form in which it can be fed directly into the subsequent activated sludge process. In this experiment the fermenter was once again operated at a 3.5 day sludge age and a mean

temperature of around 20°C so that, as in the case of the previous experiment, the results of the experiment outlined in Section 6.1.2 served as the control for this experiment. Because automated pH control apparatus was not available for use in this experiment, the fermenter pH was adjusted manually by the addition of sodium hydroxide pellets to each fermentation reactor. The pH value of each reactor was recorded immediately before the sodium hydroxide addition and again about 15 minutes later. The raw data for this experiment are listed in Appendix A2 and the mean results are presented in Table 6.3. The pH results are presented in the form of a "low" and a "high" reading for each day, indicating the pH value immediately before and after the sodium hydroxide addition, respectively.

The results of this experiment, presented in Table 6.3, indicate that raising the pH of a primary sludge fermenter operating at a 3.5 day sludge age resulted in a mean drop in VFA production yield of about 30%, from 0.0906 to 0.0633 mg HAc/mg COD of primary sludge. This result is in spite of the fact that the operational parameters of the experimental and control periods were similar except that the mean temperature was about 1°C higher in the experimental period. Examination of the VFA production raw data in Appendix A2 points to an interesting phenomenon in this experiment. The concentration of VFA's produced in the first three weeks of the experiment was around 200 mg/L of acid (as HAc), indicating that there was some initial period of improved production. However, in the final four weeks of the experiment, the VFA production dropped to around 80 mg/L,

Table 6.3. The Effect of pH on VFA Production from Primary
Sludge (Section 6.3)

Parameter	Control	Experiment
Sludge Age (days)	3.5	3.5
Influent (mg COD/L)	250	282
Sett. Sewage (")	140	165
Prim. Sludge (")	1794	1920
Underflow rate (% of raw sewage flow rate)	6.67%	6.67%
Temperature (^{o}C):		
Ferm. 1	18.1	18.9
Ferm. 2	19.6	20.8
MLSS (mg/L):		
Ferm. 1	1961	1357
Ferm. 2	2139	1513
Median pH:		Low/High
Ferm. 1	5.90	6.75/7.05
Ferm. 2	5.60	6.85/7.30
VFA Production (mg/L)		
Fermenter 1:		
Acetic acid (as HAc)	61.8	46.2
Propionic acid (")	43.9	29.4
Butyric acid (")	0.7	1.0
Total acid (")	106.4	76.6
Fermenter 2:		
Acetic acid (as HAc)	90.9	66.7
Propionic acid (")	70.1	50.8
Butyric acid (")	0.7	4.0
Total acid (")	162.5	121.5
Yield (mg HAc/mg COD)	0.0906	0.0633

resulting in a mean VFA production for the experiment of 121.5 mg/L. This large variation in production during the course of the experiment may have been the result of the unstable operating conditions in the fermenter itself, particularly with regard to the pH control. Because the pH was only adjusted once daily, as opposed to being constantly maintained at a given value, the pH of the fermentation reactors was in a constant state of flux, i.e. it was raised by about 0.5 pH units once daily and then allowed to drop by that amount during the following 24 hours. It is possible that the constantly changing pH environment itself was responsible for the diminished VFA production and outweighed any possible benefit of the mean increase in the fermenter pH. For this reason, an additional pilot-scale study with automated pH control in the fermentation reactors is still required in order to accurately determine the potential for improved VFA production using pH control. With regard to the distribution of the component acids, there appears to be very little difference in the acid production except that the butyric acid component increased from 1% to around 3%. However, it is not anticipated that this will have a major effect on the usefulness of the VFA as a substrate in the activated sludge process.

CHAPTER SEVEN

THE USE OF PRIMARY SLUDGE FERMENTATION

IN THE ACTIVATED SLUDGE PROCESS

The principle objective of the experiments described in this

chapter was to determine to what extent the P removal

characteristics of the nutrient removal activated sludge process

can be enhanced by the use of primary sludge fermentation. This

series of experiments was conducted at the pilot-scale with the

fermenter incorporated into the activated sludge process. The

raw sewage flowed into the primary clarifier, from which the

settled sewage passed into the head end of the process. The

primary sludge was pumped into the two-stage fermenter, which was

operated at a sludge age of 10 days with the use of the fermenter

secondary clarifier (See Chapter Six, Section 6.1.4). The

effluent from the fermenter clarifier then flowed into the

anaerobic zone (one that received little or no incoming nitrate)

of the process so that efficient use of the VFA's present in the

fermenter liquor could be made in the excess biological P removal

mechanism (See Chapter Five, Section 5.2) rather than the

substrate being utilized in the denitrification reaction. The

process schematics used are shown in Figs. 7.1 and 7.2.

Fermenter and activated sludge process operation and sampling was

done using similar methodologies to those described in Chapters

Six and Five, respectively. In each section the results of the

performance of the process with the incorporation of primary

sludge fermentation into the design are compared with those for a

similar process configuration that received raw sewage only and, therefore, served as the control. The mean P removal efficiency was measured by the $\Delta P/\Delta COD$ ratio (i.e. the mass of P removed per unit mass of COD removed from the incoming wastewater) of the combined fermenter and activated sludge process in the experimental periods. The degree to which excess biological P removal was occurring in the activated sludge process was determined by the extent to which the dry weight percentage P content of sludge drawn from the aerobic zone of the process exceeded 1.5%, the accepted P content of organisms not storing excess polyphosphate.

7.1. The Use of the Fermenter in the Simplified Nutrient Removal Activated Sludge Process

The objective of this section was to determine to what extent the P removal characteristics of a simplified unaerated/aerated process can be enhanced by the use of primary sludge fermentation. Schematic diagrams of the process configurations used in the control and experimental periods are shown in Fig. 7.1.

7.1.1. Control

The process configuration used and the results of this experiment are those of Section 5.1 (Chapter Five) for the simplified unaerated/aerated nutrient removal process that received no sodium acetate addition. The process had a 30% unaerated volume fraction which was further divided into two zones of approximately equal size. The first unaerated zone received the raw influent and the return sludge recycle which had a recycle

CONTROL
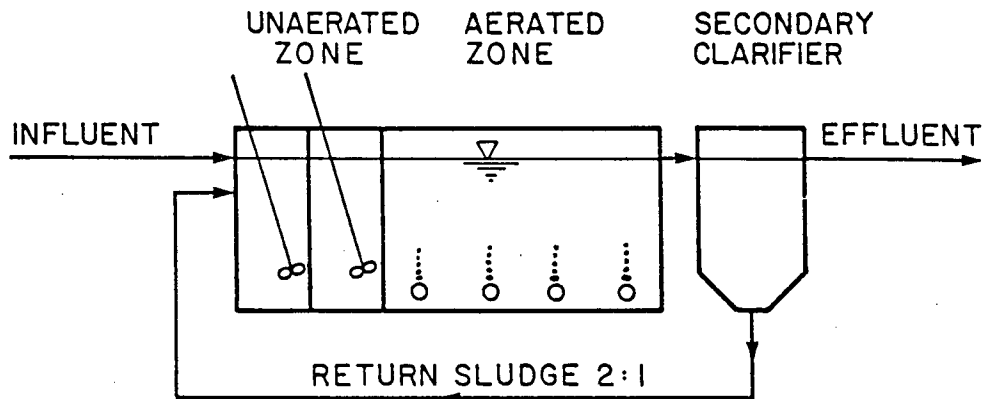
EXPERIMENT
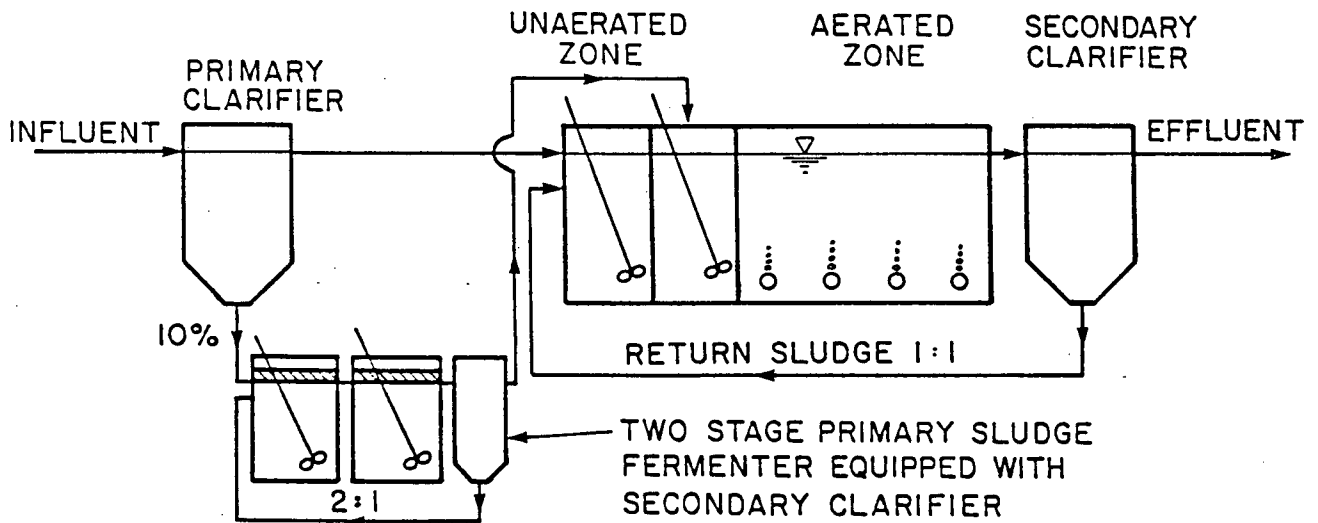
Figure 7.1. The use of primary sludge fermentation in the
simplified nutrient removal process - Control and
Experimental process configurations

ratio of 2:1 with respect to the influent flow rate. The raw
data for this experiment are shown in Appendix A1 (Section 5.1.1)
and the mean results are presented in Table 7.1.

The results of Experiment 7.1.1 indicate that no excess
biological P removal took place in the simplified nutrient
removal process. The $\Delta P/\Delta COD$ ratio was 0.0069 while the mean dry
weight percent P content of the sludge drawn from the aerobic
zone of the process was 1.46%. Furthermore, no P release took
place in the anaerobic zone of the process. This result is not
unexpected in terms of the present understanding of the
prerequisites for excess biological P removal, as it can be
assumed that little or none of the simple substrate required for
the excess biological P removal mechanism was available to the
organisms in the second unaerated zone, the only zone not
receiving a high influent nitrate concentration.

7.1.2. Experiment
In the experimental period, the primary sludge fermenter was
incorporated into the design of the simplified nutrient removal
process (See Figure 7.1). The first unaerated zone received the
settled sewage and the return sludge recycle, whose recycle ratio
was reduced to 1:1 with respect to the influent flow rate, and
the second unaerated zone received the fermenter effluent. It
was reasoned that all of the nitrate entering the first unaerated
zone would be denitrified using the more complex organics present
in the settled sewage as substrate and as such, the second
unaerated zone would receive little or no nitrate loading. In

Table 7.1. The Use of Primary Sludge Fermentation in the
Simplified Nutrient Removal Process (Section 7.1)

Parameter	Section No.	
	7.1.1 (Control)	7.1.2 (Experiment)
Raw Influent (mg COD/L)	253	332
Influent Total P (mg/L)	4.02	5.64
Influent TKN (mg/L)	21.02	33.06
Sett. Sewage (mg COD/L)	-	183
Fermenter		
Primary Sludge (mg COD/L)	-	1682
Primary Clarifier Underflow	-	10%
Sludge Age (days)	-	10
VFA Production (mg HAc/L)	-	184
Activated Sludge Process		
Ortho-P (mg/L):		
1st Unaerated (mg/L)	2.50	2.58
2nd Unaerated (mg/L)	2.76	12.49
Aerobic (mg/L)	2.42	1.70
Effluent Total P (mg/L)	2.62	1.72
ΔP (mg/L)	1.40	3.92
% P Removal	34.8%	69.5%
Effluent COD (mg/L)	50	53
ΔCOD (mg/L)	203	279
ΔP/ΔCOD	0.0069	0.0141
NO_3-N (mg/L):		
1st Unaerated (mg/L)	1.27	0.26
2nd Unaerated (mg/L)	0.14	0.21
Aerobic (mg/L)	7.43	13.24
Effluent (mg/L)	7.56	12.70
Effluent TKN (mg/L)	-	1.79
Process Temp. (^{o}C)	18.5-21.0	19.0-22.5
Sludge Age (days)	20	20
Aerobic MLSS (mg/L)	2783	2619
SVI	90.8	69.6
Sludge Dry Wt. % P	1.46%	3.20%

this way all of the VFA present in the acid-rich fermenter liquor would be available for the excess biological P removal mechanism. In Chapter Five it was shown that far more efficient use of the added sodium acetate solution was made by adding it to the second, rather than the first, unaerated zone. An additional modification made to the activated sludge process was that the unaerated volume fraction was increased from 30% to 40%, but was still divided into two zones of equal size. The raw data for this experiment are shown in Appendix A3 and the mean results are presented in Table 7.1.

The results presented in Table 7.1 clearly show a significant improvement in the P removal characteristics when primary sludge fermentation was incorporated into the design of the process. The percentage of influent P removed by the process increased from 34.8% to 69.5%. The $\Delta P/\Delta COD$ ratio doubled from 0.0069 to 0.0141 while the dry weight percent P content of the sludge drawn from the aerobic zone increased from 1.46% to 3.20%. It is quite clear, therefore, that a significant degree of excess biological P removal was occurring in the process throughout the duration of this experiment. In addition, a considerable degree of P release, up to a concentration of 12.49 mg/L, took place in the second unaerated zone. It appears as if the additional substrate present in the acid-rich fermenter liquor was responsible for this large concentration of P released into the supernatant of the second unaerated zone and for the significant degree of excess biological P removal exhibited by the process.

With regard to the N removal efficiency of the process, the incorporation of primary sludge fermentation into the process design does not appear to have had a significant effect on the process performance. Although the effluent nitrate concentration increased from 7.56 to 12.70 mg N/L, this can largely be attributed to the significantly higher influent TKN concentration (33.06 vs 21.02 mg/L) during the experimental period and the reduced sludge recycle rate. The sludge settling characteristics of the process operated with primary sludge fermentation showed some improvement, with the SVI decreasing from 91 to 70.

7.2. The Use of the Fermenter in the UCT Process

The objective of this section was to determine the extent to which the P removal characteristics of the UCT process, a process configuration designed to satisfy the prerequisites for excess biological P removal, could be further enhanced by the use of primary sludge fermentation. Briefly, the UCT process, as described in Chapter Two, consists of two unaerated zones followed by an aerobic zone. The first unaerated zone, known as the anaerobic reactor, receives the influent sewage and the r-recycle from the second unaerated zone, or anoxic reactor (See Chapter Two, Fig. 2.8). The anoxic reactor receives the return sludge recycle and the internal recycle from the aerobic zone and it is in this reactor that all of the process denitrification takes place. The basic objective of the process operation is to limit the mass of nitrate entering the anoxic reactor via these two recycles so that it approximates the denitrification capacity of the reactor. In this way, the nitrate concentration in the

anoxic reactor is maintained at a near-zero value and only a minimal mass of nitrate is discharged to the anaerobic reactor via the r-recycle. As such, it is in the anaerobic reactor that the prerequisite conditions for excess biological P removal are most likely to be met, as little or no denitrification takes place in the reactor and all of the simple substrate entering the reactor in the incoming sewage is therefore available for the excess biological P removal mechanism. However, in the case of UCT processes treating typically low organic strength North American wastewaters, it is thought that there are insufficient quantities of the required substrates present in the wastewater, and that the concentration of these substrates can be increased by using primary sludge fermentation. During the control period, the UCT process received raw sewage only, and during the experimental period the influent was pretreated using primary sludge fermentation in order to increase the VFA concentration entering the process. Both the settled sewage and the fermenter liquor were discharged into the anaerobic reactor of the process. Schematic diagrams of the two process configurations used are presented in Figure 7.2.

7.2.1. Control

The UCT process configuration used in this experiment had both the anaerobic and the anoxic reactors each comprising 20% of the process volume with the remaining 60% of the process volume being maintained under aerobic conditions. The sludge return and the r-recycle were maintained at a ratio of 1:1 with respect to the influent flow rate and the internal recycle was generally kept at

CONTROL

ANAEROBIC ANOXIC AEROBIC
ZONE ZONE ZONE

1:1 3:1

SECONDARY
CLARIFIER

INFLUENT EFFLUENT

RETURN SLUDGE 1:1

EXPERIMENT

ANAEROBIC ANOXIC
ZONE ZONE

1:1

PRIMARY
CLARIFIER

AEROBIC
ZONE

SECONDARY
CLARIFIER

INFLUENT EFFLUENT

10%

RETURN SLUDGE 1.5:1

TWO STAGE PRIMARY SLUDGE
FERMENTER EQUIPPED WITH
SECONDARY CLARIFIER

2:1
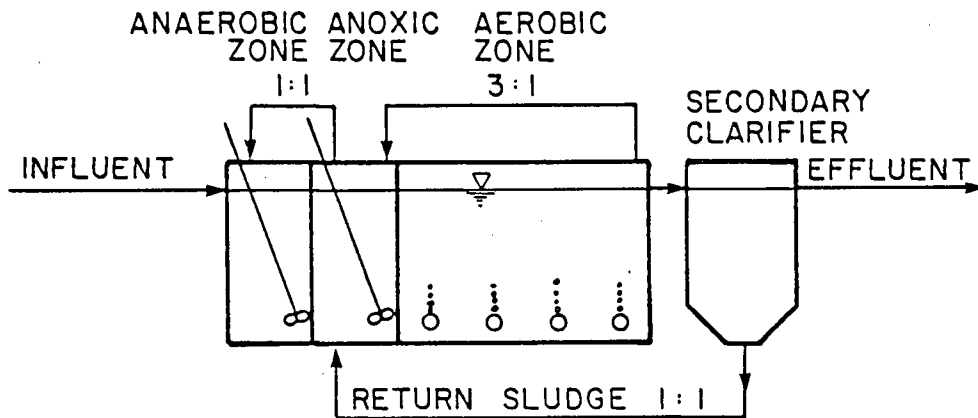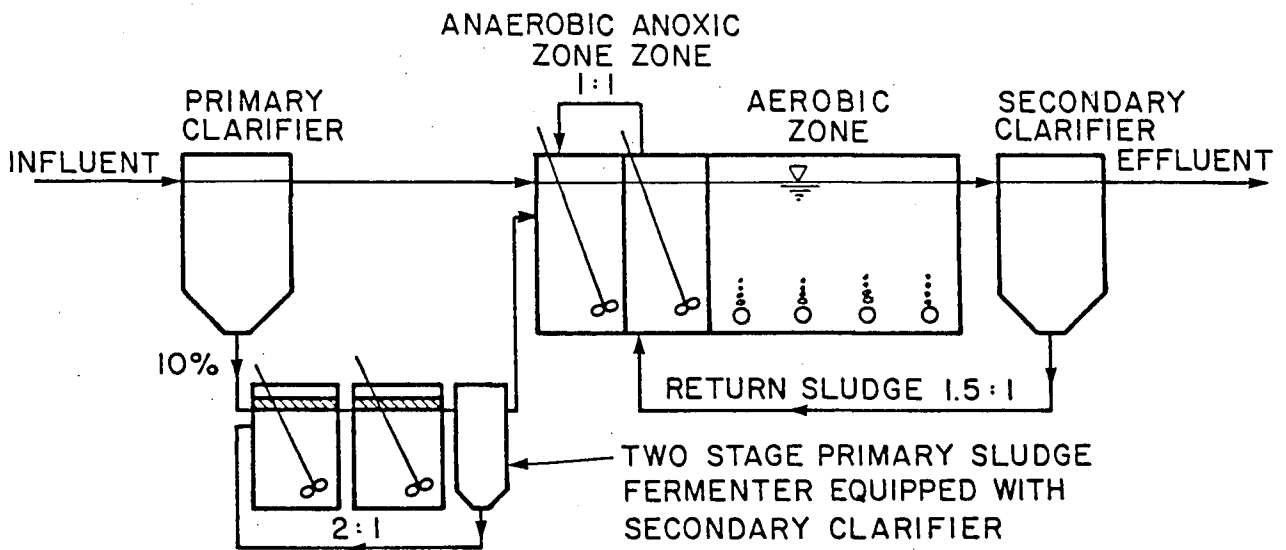
Figure 7.2. The use of primary sludge fermentation in the
UCT process - Control and Experimental process
configurations

a 3:1 recycle ratio, although this was periodically reduced in order to minimize the nitrate concentration in the anoxic reactor. The raw data from this experiment are shown in Appendix A3 and the mean values are presented in Table 7.2.

The results of this experiment indicate that there was some degree of excess biological P removal occurring in the UCT process, even without the use of primary sludge fermentation. The P removal efficiency, as measured by the $\Delta P/\Delta COD$ ratio, was 0.0086. This figure is about 25% greater than that for the simplified unaerated/aerated nutrient removal process used in the control period of Section 7.1. The mean dry weight percent P content of the sludge was 2.11%, an increase of about 44% over that achieved in Section 7.1.1, indicating that some degree of excess poly-P storage took place during the experiment. Furthermore, the reactor ortho-P concentrations presented in Table 7.2 show that a small degree of P release occurred in the anaerobic reactor.

7.2.2. Experiment

In this experiment, the settled sewage from the primary clarifier was discharged into the anaerobic zone of the process, as in the case of the experimental period of Section 7.1. The primary sludge was pumped into the 2-stage primary sludge fermenter which was operated at a sludge age of 10 days, and the fermenter liquor from the clarifier was discharged together with the settled sewage into the anaerobic reactor at the head end of the process. Two additional changes were made to the operation of the UCT

Table 7.2. The Use of Primary Sludge Fermentation in the UCT
Process (Section 7.2)

Parameter	Section No.	
	7.2.1 (Control)	7.2.2 (Experiment)
Raw Influent (mg COD/L)	236	330
Influent Total P (mg/L)	3.54	5.43
Influent TKN (mg/L)	20.20	32.40
Sett. Sewage (mg COD/L)	-	186
Fermenter		
Primary Sludge	-	1682
Primary Clarifier Underflow	-	10%
Sludge Age (days)	-	10
VFA Production (mg HAc/L)	-	148
Activated Sludge Process		
Ortho-P (mg/L):		
Anaerobic Zone (mg/L)	3.88	10.78
Anoxic Zone (mg/L)	2.21	5.27
Aerobic Zone (mg/L)	1.82	1.59
Effluent Total P (mg/L)	1.72	1.47
ΔP (mg/L)	1.72	3.96
% P Removal (mg/L)	50.0%	72.9%
Effluent COD (mg/L)	24	39
ΔCOD (mg/L)	212	291
$\Delta P/\Delta COD$	0.0086	0.0136
NO_3-N (mg/L)		
Anaerobic Zone (mg/L)	0.08	0.35
Anoxic Zone (mg/L)	0.94	2.74
Aerobic Zone (mg/L)	3.95	12.19
Effluent (mg/L)	3.07	11.88
Effluent TKN (mg/L)	-	1.50
Process Temp. (^{o}C)		
Sludge Age (days)	20	20
Aerobic MLSS (mg/L)	3777	3791
SVI	207.3	109.1
Sludge Dry Wt. % P	2.11%	3.10%

process. The internal mixed liquor recycle was eliminated and the return sludge recycle ratio was increased to 1.5:1 with respect to the influent flow rate. In this experiment, therefore, the process was not optimized for nitrogen removal. The raw data for this experiment are shown in Appendix A3 and the mean values are presented in Table 7.2.

As in the case of the previous section, the P removal characteristics of the UCT process were significantly enhanced, although not to the same extent, by the incorporation of primary sludge fermentation into the process design. The P removal efficiency of the process increased from 0.0086 to 0.0136 mg P/mg COD between the experimental and control periods, an increase of 58%. The mean dry weight percent P content of the sludge increased from 2.11% to 3.10%, indicating that a significant degree of excess biological P removal occurred during this experiment. A relatively large concentration of P was released into the supernatant of the anaerobic reactor, probably as a result of the increased mass of VFA's entering the reactor via the acid-rich fermenter liquor. It is interesting to note, however, that when comparing the performance of the UCT and the simplified nutrient removal process, with both processes having primary sludge fermentation incorporated into the design (i.e. Sections 7.1.2 and 7.2.2), the P removal characteristics of the simplified process were slightly better. Both the P removal efficiency and the dry weight percent P content of the sludge were a little higher, viz. 0.0141 vs 0.0136 mg P/mg COD and 3.20% vs 3.10%, respectively. This can, in all likelihood, be

attributed to the fact that the VFA production of the fermenter was significantly lower during the experiment described in Section 7.2.2, than it was in the earlier experiment viz. 148 vs 184 mg HAc/L. The higher effluent nitrate concentration observed in the experimental period over that in the control period can partially be attributed to the fact that the process configuration was no longer optimized for nitrogen removal. In addition, the influent TKN concentration was significantly higher in this experiment than in the control period (32.4 vs 20.2 mg/L) which resulted in a much higher mass of nitrate being nitrified and consequently, a higher effluent nitrate concentration. Incorporation of primary sludge fermentation into the design of the UCT process configuration also appears to have improved the settling characteristics of the sludge, with the mean SVI value decreasing from 207 to 110 between the control and experimental periods.

CHAPTER EIGHT

DISCUSSION OF RESULTS

8.1. Introduction

In recent years excess biological P removal in the activated
sludge process has received an increasing amount of attention
from many research workers worldwide, primarily due to its
recognition as a viable method of removing large masses of P from
wastewater point sources. Since the onset of the research
reported in this thesis, a number of international conferences
and seminars dealing with biological P removal have been held.
In addition, an international study group on "P Removal in
Biological Sewage Treatment Processes", with a membership of over
100 research workers, has been formed under the auspices of the
International Association of Water Pollution Research and Control
(IAWPRC). As a result, considerable progress has been made in
many aspects of biological P removal while the research reported
herein was in progress. For this reason, it was decided to
discuss the findings of this research by dealing with each aspect
and its contribution to the further development of excess
biological P removal technology in the activated sludge process,
within the context of some of the recent parallel research that
has been carried out by others during the course of this study.

8.2. The Excess Biological P Removal Mechanism

The definition of the prerequisites of the excess biological P
removal mechanism in terms of the concentration of the "readily
biodegradable COD" surrounding the organisms in the anaerobic

zone of the process, as set out by Siebritz et al. (1982,1983) was a milestone in the understanding of the nature of the P removal mechanism. This is primarily because it represented a major shift in emphasis away from defining the prerequisites of the removal mechanism in terms of a certain minimum degree of anaerobic stress in the anaerobic zone, in favour of recognizing the importance of the availability of carbonaceous substrate to the organisms in the zone. However, the "readily biodegradable" hypothesis suffers from a number of theoretical and practical limitations. Firstly, the determination of the readily biodegradable COD concentration present in a wastewater is not based on any specific chemical analysis of the wastewater. Instead, it requires the operation of a short sludge age, completely aerobic, cyclically fed activated sludge process treating the specific waste, and measuring the drop in oxygen consumption rate at the termination of the feed cycle. Recent experience in the operation of these units at the N.I.W.R. laboratories in Pretoria, South Africa and at the University of British Columbia (Manoharan, 1985) has revealed a number of practical problems affecting the usefulness of this technique. The two most important of these problems are filamentous sludge bulking, and difficulties associated with the suppression of nitrification, which introduces a confounding oxygen demand on the system. Furthermore, the lack of a precise chemical definition of "readily biodegradable COD" does not recognize the possibility of some readily biodegradable wastewater components being more effective than others in inducing the excess

biological P removal mechanism. Substrate that is readily utilizable by aerobic heterotrophs in general is not necessarily readily utilizable in the excess biological P removal mechanism. Finally, the hypothesis merely states that a certain minimum concentration of readily biodegradable COD be available to the organisms in the anaerobic zone but does not clarify the role of the substrate in the P removal mechanism.

The research presented herein clearly demonstrates that a number of substrates which could be classified as being "readily biodegradable COD" have widely differing effects on the P release mechanism under anaerobic conditions. For example, sodium acetate and propionic acid proved to be the most effective in inducing anaerobic P release while glucose and butyric acid were only half as effective on the basis of their COD value. This seems to suggest that the effectiveness of a specific substrate is some inverse function of the complexity of the substrate molecule. Furthermore, the fact that acetic acid was significantly less effective than sodium acetate in inducing anaerobic P release on an equivalent COD basis suggests that the ionic form of the substrate is also important to its role in the P removal mechanism.

Anaerobic batch testing, using various concentrations of sodium acetate at the start of the test and measuring the ortho-P and acetate supernatant profiles, revealed much about the fate of the substrate during anaerobic P release. For all initial substrate concentrations, P release and substrate utilization appear to be

integral parts of an exchange phenomenon, with the cessation of P release and substrate utilization being coincident. The end of the P release phase occurred either due to the onset of substrate limiting conditions or due to the exhaustion of intracellular reserves of stored P. In the latter case, no more substrate was removed from the supernatant, indicating that P release plays an integral role in the transport of carbonaceous substrate into the cell. During the P release phase, 0.91 mg of P were released for every mg of sodium acetate (measured as HAc) utilized. This represents a molar ratio of 1.76 moles P/mole HAc. In a series of batch experiments similar in concept to those reported here, Fukase et al. (1982) obtained a phosphate:acetate molar ratio of 0.9:1 while Arvin (1985) and Comeau et al. (1985) reported molar exchange ratios of 1.4:1. Siebritz et al. (1983) reported a phosphate:acetate molar ratio of 2:1 but this value was based on substrate availability rather than utilization.

In batch tests where an aerobic phase followed the anaerobic P release period, P uptake was observed to occur at a rapid initial rate which slowed down somewhat after a period of about one hour. There appeared to be some loose correlation between the mass of P released anaerobically and the mass of P taken up under subsequent aerobic conditions. However, these results were not as conclusive as those of Wentzell et al. (1984) who were able to develop the following relationship between P release uptake:

$$P \text{ uptake (mg/L)} = 5.5 + 1.13 X P \text{ release (mg/L)}$$

The above equation suggests that the greater the degree of anaerobic P release, the greater is the system P removal of the

process. This supports an early observation of Barnard (1976), who, in addition to being the first to explicitly state that there was a connection between P release and excess biological P removal, also observed that the degree of P release determined the degree of system P removal.

In general, the findings of this research support the proposed biochemical models for the excess biological P removal mechanism of a number of research workers who endeavoured to explain why the anaerobic/aerobic sequence is essential for the proliferation of organisms capable of storing excess quantities of phosphorus in the activated sludge process. For example, significant amounts of carbon storage in the form of poly-B-hydroxybutyrate (PHB) have been reported to occur in the anaerobic zone of processes exhibiting excess biological P removal (Nicholls and Osborn (1979), Fukase et al. (1982) and Comeau (1984)). It seems likely that the ability to store carbon as PHB in a stressed environment, where aerobic metabolism is impossible (in the absence of both dissolved oxygen and nitrate), is the key to the proliferation of these organisms in the process. Upon entering the aerobic zone of the process, where substrate limiting conditions often occur, organisms that have intracellularly stored carbon have a decided advantage over organisms that must rely on membrane transport in the highly competitive activated sludge environment. It is in the aerobic environment that these organisms store vast quantities of phosphorus not needed for basic metabolic purposes, in long chains of inorganic polyphosphate known as volutin granules. Fukase et al. (1982)

reported a dry weight percent P content of around 12% in sludge drawn from the aerobic zone of a laboratory-scale anaerobic/ aerobic activated sludge process treating a synthetic feed largely made up of sodium acetate. It is these polyphosphate reserves that are broken down and hydrolyzed when the organisms re-enter the anaerobic zone of the process, presumably in order to facilitate the transport and intracellular storage of the available carbonaceous substrate in the zone. Comeau et al. (1985b) refers to Bio-P bacteria (bacteria responsible for enhanced biological P removal) as capable of storing both polyphosphate under aerobic conditions and carbon under anaerobic conditions. In their comprehensive biochemical model of the excess biological P removal mechanism for both anaerobic and aerobic conditions (see Fig. 8.1), they suggest that anaerobic P release plays a passive role in the P removal mechanism. Because acetate must be transported into the cell in an electrochemically neutral form (i.e. as HAc), the primary function served by the degradation of polyphosphate is to re-establish the pH gradient across the cell membrane, thereby facilitating more acetate uptake. They suggest that P release takes place as a result of the intracellular accumulation of this unusable metabolite. Furthermore, they theorize that poly-P also serves as the energy source in the synthesis of acetyl CoA from substrates such as acetate prior to their storage as PHB. They also point out that K^+, Mg^{++} and Ca^{++} are co-transported across the cell wall together with the phosphate in ratios that remain the same for both uptake and release conditions. It appears, therefore, that
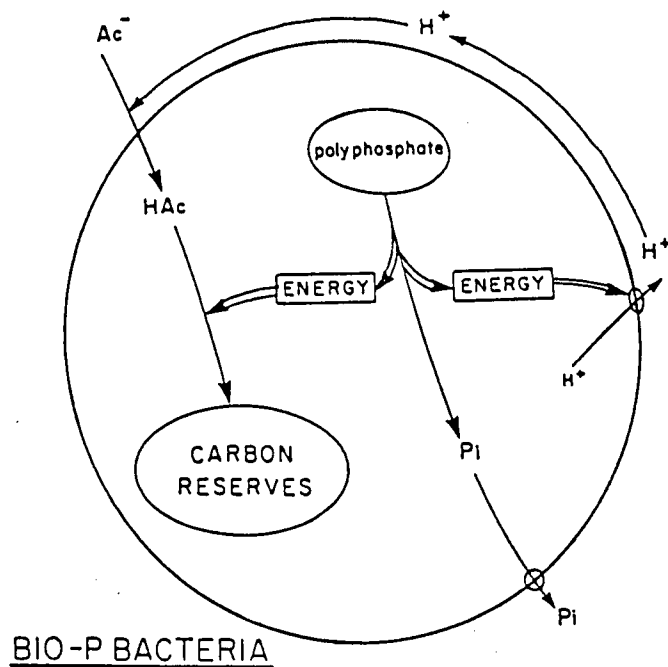
Fig. 8.1a. Simplified model for anaerobic metabolism of Bio-P bacteria. Transport and storage of simple carbon substrates such as acetate require energy obtained from the breakdown of polyphosphate reserves. Phosphate, an unusable molecule by the cell under such conditions, is released into solution. Adapted from Comeau et al. (1985b).
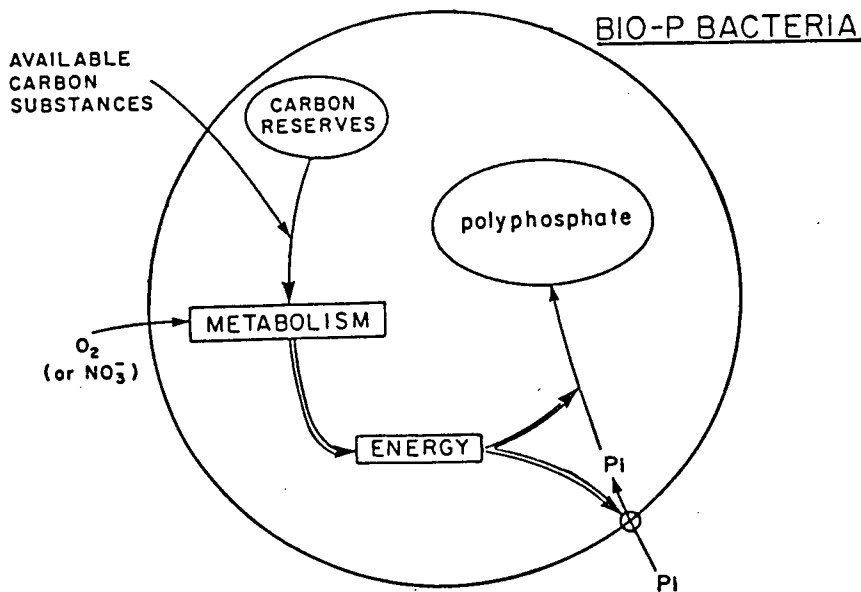
Fig. 8.1b. Simplified model for aerobic metabolism of Bio-P bacteria. Carbon from internal reserves or from solution is used with oxygen (or nitrate) to produce energy for the growth of Bio-P bacteria. Energy is used for phosphate transport and its storage as polyphosphate. Adapted from Comeau et al. (1985b).

polyphosphate storage, and therefore excess biological P removal in general, plays a role that is secondary to carbon storage in the proliferation of Bio-P bacteria in the activated sludge process. This, of course, leads to the question of why cells store polyphosphate at all when subjected to stressful conditions. The role of polyphosphate as a reserve material for growth is well recognized but its role as an energy source is still under investigation. Harold (1966) points out that polyphosphate appears to be synthesized by the transfer of phosphate molecules from adenosine triphosphate (ATP) to growing chains of polyphosphate and suggests that, from an evolutionary viewpoint, polyphosphate may predate ATP as the chief energy carrier in bacterial cells. Therefore, ability of certain bacteria to store polyphosphate may be the result of a "genetic throwback" from the evolution of early bacterial cells.

8.3. Effect of Nitrate on Excess Biological Phosphorus Removal

Nitrate is the most abundant form of combined oxygen present in nutrient removal activated sludge processes, largely because of the incorporation of both nitrification and denitrification into the design of these processes. The detrimental effect of nitrate entering the anaerobic zone of processes designed for excess biological P removal was first pointed out by Barnard (1976). He stated that nitrate would increase the redox potential of the zone, thereby reducing the degree of anaerobic stress to a level that may not induce anaerobic P release, which at the time he considered to be the prerequisite for the excess biological P removal mechanism to operate. Barnard's observation regarding

the detrimental effect of nitrate entering the zone was confirmed
by McLaren and Wood (1976), Nicholls (1977), Simpkins and McLaren
(1978) and others, and was the primary motivation for the
development of the UCT process by Rabinowitz and Marais (1980)
and Siebritz et al. (1980). The UCT process configuration is one
in which a zero nitrate discharge to the anaerobic reactor can be
guaranteed under most operating conditions. However, it was in
the quantification of the negative effect of nitrate entering the
anaerobic zone of the process that there existed much
uncertainty. In the development of the "readily biodegradable
COD" hypothesis to define the prerequisites for excess biological
P removal, Siebritz et al. (1982, 1983) assumed that any nitrate
entering the anaerobic zone would result in the oxidation of
readily biodegradable COD in the zone in a ratio determined by
the kinetic theory of denitrification of van Haandel et al.
(1981). The denitrification theory states that in the oxidation
of substrate using nitrate as the terminal electron acceptor,
8.6 mg of substrate (as COD) are oxidized for every mg of nitrate
(as N) denitrified. Siebritz et al. (1982) hypothesized that in
the case of the enhanced biological P removal process, the
readily biodegradable COD required for the excess biological P
removal mechanism is utilized preferentially in the
denitrification reaction at a rate of 8.6 mg COD/mg NO_3-N
entering the anaerobic zone of the process.

The anaerobic batch tests presented in this thesis (Section 4.3,
Chapter 4) clearly show that, although the presence of nitrate is
inhibitory to the anaerobic P release mechanism, the inhibitory

effect is not as great as that suggested by Siebritz et al. (1982). Furthermore, the inhibitory effect, while increasing with increasing nitrate concentrations, is an inverse function of the mass of substrate available to the organisms. For example, in the flasks that received an initial substrate concentration of 100 mg COD/L of acetate, initial nitrate concentrations of up to 9 mg N/L had no effect on the P release mechanism and in the flask that had an initial nitrate concentration of 12 mg N/L, the P release mechanism was only slightly inhibited. However, in the flasks that received an initial substrate concentration of 50 mg COD/L, the presence of nitrate in concentrations as low as 3 mg N/L proved to be inhibitory to the P release mechanism. In the former case, the initial presence of 12 mg N/L of nitrate (all of which was denitrified in the first hour of the experiment) required 12 X 8.6 = 103.2 mg/L of COD for denitrification, i.e. all of the initial substrate concentration, according to the denitrification theory of van Haandel et al. (1981). In the latter case, the inhibitory effect of the nitrate on the P release mechanism was only a maximum of 3.0 mg P release per mg of nitrate (as N) present in the reactor at the start of the experiment. Combining this value with the $\Delta P/\Delta HAc$ value of 0.89 mg P/mg HAc (equivalent to 0.83 mg P/mg COD) found in the control flask that received no initial nitrate concentration, the inhibitory effect of the nitrate is equal to:

$$3.0/0.83 = 3.60 \text{ mg COD/mg } NO_3\text{-N}$$

which is less than one half of that predicted by the denitrification theory, assuming preferential usage of the

substrate in the denitrifition reaction. The results of these experiments clearly show that the inhibitory effect of nitrate on the P release mechanism is not as great as that suggested by current theory. There are two possible explanations for this: either the substrate requirements for denitrification (8.6 mg COD/mg N) is overestimated in the denitrification theory of van Haandel et al. (1981), or the substrate is not utilized preferentially in the denitrification reaction (and thus rendered unavailable for the P release mechanism), as hypothesized by Siebritz et al. (1982), i.e. that there exists some phenomenon whereby the same substrate can be utilized in both the P release mechanism and the denitrification reaction. These two possible explanations require further examination.

The substrate requirements for denitrification due to the oxidation of readily biodegradable COD, according to the theory of van Haandel et al. (1981) (i.e. 8.6 mg COD/mg NO_3-N) was derived in the following way:

   According to the biological growth kinetic theory of Marais and Ekama (1976), for every mg of COD utilized under aerobic conditions, $(1-PY_h)$ mg of oxygen are required, where P is the COD/VSS ratio of the sludge = 1.48 mg COD/mg VSS and Y_h is the yield coefficient for heterotrophic organisms = 0.45 at $20\,^{o}$C. Both of these constants were determined experimentally. Therefore, numerically $(1-1.48 \times 0.45) = 0.33$ mg of oxygen are required for every mg of COD utilized by the organisms of the process. Equating the electron equivalence of nitrate and oxygen, 2.8 g nitrate (as N) = 8.0 g oxygen (as O). Thus the

nitrate consumption per unit mass of COD oxidized

$$= 0.33 \times 8.0/2.8 = 0.116 \text{ mg N/mg COD.}$$

Conversely, 8.6 mg of COD are metabolized for every mg of NO_3-N reduced.

From the above derivation it is clear that the substrate requirement for denitrification, as estimated by van Haandel et al. (1981), is not based on the stoichiometry of denitrification but rather on the biomass yield factor for the aerobic oxidation of any carbonaceous substrate and the stoichiometric equivalence of nitrate and oxygen. However, an alternative approach is to estimate the required mass of a particular substrate, such as acetate, in the denitrification of a given mass of nitrate using a stoichiometrically balanced equation.

The following equation for the denitrification of nitrate (in this case nitrate serves as the terminal electron acceptor) using acetate as the electron donor is derived from the work of McCarty et al. (1969) using a consumptive ratio (defined as the ratio of the total mass of substrate consumed during denitrification to the stoichiometric requirement for denitrification and deoxygenation alone), Cr=1.30:

$$0.771 \text{ } CH_3COO^- + NO_3^- => 0.475 \text{ } N_2 + 1.288 \text{ } CO_2 + 1.670 \text{ } OH^-$$
$$+ 0.144 \text{ } H_2O + 0.051 \text{ } C_5H_7O_2N$$
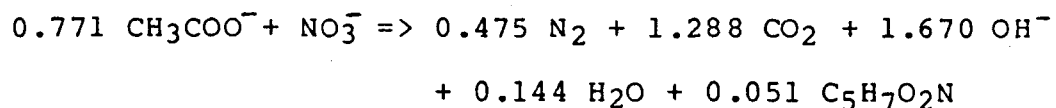
From the above equation, 0.771 moles of acetate are oxidized for every mole of nitrate denitrified. Expressing the acetate in the COD equivalence form, 3.53 mg of acetate (as COD) are utilized for every mg of nitrate (as N) denitrified. This ratio is

extremely close (within 2%) of the ratio determined in this research by quantifying the detrimental effect of the presence of nitrate on the anaerobic P release mechanism, and the acetate/phosphorus molar exchange ratio in the control reactor of the same experiment. It follows, therefore, that the model of Siebritz et al. (1982) overestimates the detrimental effect of nitrate by assuming that the readily biodegradable COD required for the excess biological P removal mechanism is utilized preferentially in the denitrification reaction at a rate of 8.6 mg COD per mg of nitrate (as N) entering the anaerobic zone of the process.

With regard to the simultaneous use of the available substrate in both the excess biological P removal mechanism and the denitrification reaction, the batch testing reported in this thesis shows an interesting phenomenon. When nitrate is present in an unaerated flask of activated sludge, the P release mechanism is virtually unaffected, provided that there exists an excess concentration of a carbonaceous substrate such as acetate. However, as soon as the substrate becomes limiting, the P release mechanism is reversed and P uptake immediately commences. This observation was confirmed in the work of Comeau (1984) who injected a sodium nitrate solution into a series of anaerobic batch flasks in which anaerobic P release had been induced three hours previously by the addition of acetate. At the time of the nitrate addition, the rapid P release phase was complete and substrate limiting conditions prevailed in the flasks.

Immediately upon injection of the nitrate solution, P uptake
commenced and continued for the period while nitrate was present
in the flasks. As soon as denitrification was complete, P
release once again resumed at a somewhat lower rate. From these
observations, Comeau (1984) concluded that at least a fraction of
the organisms involved in excess biological P removal were
capable of denitrification and therefore, had the ability to
accumulate polyphosphate in the presence of nitrate and would be
capable of such accumulation in the anoxic zone of nutrient
removal processes. Furthermore, Comeau (1984) observed the
disappearance of PHB from the sludge during the denitrification
in the absence of excess substrate and concluded that the
intracellularly stored PHB serves as the carbon source for
denitrification. In this way, the carbon plays a dual role in
the nutrient removal process. Firstly, it is stored under
anaerobic conditions, thereby initiating the excess biological P
removal mechanism and then, under subsequent anoxic conditions,
it is utilized as the carbon source for denitrification. The
incorporation of this dual function of short chain VFA's and
other sources of "readily biodegradable COD" has proved to be
highly successful in the design of processes for the simultaneous
removal of both P and N, such as the Bardenpho and UCT type
processes, in which significant amounts of P uptake occur in the
anoxic zone of the process.

The research using the UCT process reported in this thesis
(Section 7.2, Chapter 7) confirms the findings of Florentz and
Granger (1982), Siebritz et al. (1983), Comeau et al. (1985b)

and others that P uptake is an anoxic as well as an aerobic phenomenon, i.e. that significant masses of P are taken up in the presence of nitrate (but in the absence of dissolved oxygen) in processes that are designed to include both nitrification and denitrification. The mean P concentration and the net change in P concentration, ΔP (expressed as mg P/L of influent flow) for each of the reactors in the experiments using the UCT process in Chapter Seven are presented in Table 8.1. The ΔP values for each reactor were calculated on the basis of a mass balance for the reactor, with negative values indicating P release and positive values indicating P uptake.

Table 8.1. Mean P Concentrations and Net changes in P Concentration in the experiments using the UCT process (Section 7.2)

| Reactor | Control | | Experiment | |
	P (mg/L)	ΔP (mg/L)	P (mg/L)	ΔP (mg/L)
Influent	3.54	-	5.43	-
Anaerobic	3.88	-2.01	10.78	-10.86
Anoxic	2.21	+1.68	5.27	+5.32
Aerobic	1.82	+1.95	1.59	+9.20
Clarifier	1.72	+0.20	1.47	+0.30
System P Removal		1.82		3.96

From the above table, it is clear that in both experiments conducted on the UCT process, a significant degree of P uptake occurred in the anoxic reactor of the process. In the control period, where a small degree of excess biological P removal took place, but where the combined nitrate-rich mixed liquor flow was equivalent to four times the influent flow rate, approximately 44% of the P uptake occurred in the anoxic zone of the process. In the experimental period, primary sludge fermentation was

incorporated into the design of the process and the system P removal, as well as the degree of anaerobic P release, increased significantly. The total recycle from the end of the process to the anoxic reactor was reduced to 1.5 times the influent flow rate and the percentage of P uptake occurring in that reactor was about 36%. It is important to note that in both of these experiments the anoxic reactor only comprised about 20% of the total process volume and that anoxic P uptake was, therefore, occurring at a rapid rate. It is known that for many aerobic organisms, in the absence of dissolved oxygen, nitrate readily replaces oxygen as the terminal electron acceptor, largely because the metabolic pathway for nitrate is similar to that for oxygen. Chang and Morris (1972) found that the replacement of oxygen with nitrate as the terminal electron acceptor is facilitated by the formation of an enzyme, nitratase, whose production is inhibited by the presence of dissolved oxygen. It seems likely, therefore, that a significant number of organisms involved in the excess biological P removal mechanism are also capable of denitrification and that there are significant advantages in incorporating an anoxic reactor into the design of the activated sludge process where both denitrification and P uptake can occur simultaneously.

8.4. Primary Sludge Fermentation

Research using the pilot-scale primary sludge fermenter clearly showed that the two most efficient substrates for inducing anaerobic P release in the activated sludge process, acetate and propionate, can be produced on-site by primary sludge

fermentation. It was found that the VFA yield (measured as mg HAc per mg of primary sludge COD entering the fermenter) was optimal in the 3.5-5.0 day sludge age range at about 0.09 mg HAc/mg COD, with production dropping off slightly at shorter and longer sludge ages. It should be noted, however, that the drop in production observed at a sludge age of 2.5 days may be attributed to the lower fermenter operating temperature during that period. With regard to the effect of temperature, increasing the operating temperature of a fermenter operating at a 3.5 day sludge age from about $12^{o}C$ to about $20^{o}C$ only increased the VFA yield by about 20%, which is considerably less than that predicted by the general theory of microbial activity. Attempts to raise the operating pH of the fermenter to a value of about 7.0 using sodium hydroxide did not result in any improvement in the VFA yield, with the sludge age and operating temperature remaining constant. It should be noted that in this fermentation study the solids concentration and temperature control, as well as the pH adjustment, were poor and, therefore, additional pilot-scale research is still required in order to accurately determine the effect of these variables on VFA production.

The results of the fermentation study were confirmed by the findings of Gupta et al. (1985), who carried out a comprehensive laboratory scale primary sludge fermentation study using "fill and draw" sealed reactors to measure the effects of temperature, pH and detention time on VFA production. These researchers found that, although there was much interaction between the three

variables examined, certain clear trends emerged. For example, they found that VFA production improved consistently with increasing temperature in the 10-30°C temperature range. In general, optimum acid production was achieved at a 6 day detention time with a slight increase in production at a 10 day detention time at 10 and 20°C and a decrease in production at 30°C. However, these trends are based on net acid production values and it is possible that at 30°C and a 10 day sludge age, more acid was produced but subsequently converted to CH_4 and CO_2 by methogenic organisms. They also found that controlling the pH to a level of 7.0 did not result in an increase in VFA production but that, as in the case of the research reported in this thesis, no rigorous study into the effect of pH was carried out using a range of pH values. A comparison was made only between an uncontrolled pH condition and controlling the pH at one particular level.

It is clear that a comprehensive pilot-scale study of VFA production by primary sludge fermentation at sludge ages in the range of 1-6 days, together with the interaction of the effects of temperature, is still required in order to accurately evaluate the economics of the best combination of sludge age and temperature. In addition, ways of simplifying the design and operation of the fermenter so that the fermenter volume requirements can be minimized and the need for a fermenter secondary clarifier eliminated must still be examined (See Section 8.6).

8.5. The Use of Primary Sludge Fermentation in the Nutrient
     Removal Activated Sludge Process

The results of Chapter Seven clearly show that, in addition to

being able to produce the specific substrates required in the

activated sludge process for the excess biological P removal

mechanism to operate, these substrates can be produced in

sufficient quantities to significantly enhance the P removal

characteristics of the process. The P removal efficiency (as

measured by the $\Delta P/\Delta COD$ removal ratio) and the dry weight percent

P content of the sludge were both doubled by the incorporation of

primary sludge fermentation into the design of a simple

unaerated/aerated process configuration that previously did not

exhibit excess biological P removal. Similarly, the P removal

characteristics of a UCT process configuration that was

exhibiting some degree of excess biological P removal were

improved by about 50% by the use of primary sludge fermentation.

These results point to the increasing realization of the

importance of the nature of the COD entering the process rather

than simply considering the total influent COD concentration.

For example, the incorporation of primary clarification and

primary sludge fermentation lowers the mass of COD entering the

process due to the inefficient conversion of the primary sludge

COD into simpler organics, fermenter sludge wastage, etc.

However, in spite of the lower mass of COD entering the process,

the waste stream COD is preprocessed into a form that is far more

suitable for excess biological P removal. The complex organic

material is removed in the primary clarifier and from it simpler

short chain volatile fatty acids, the principle substrates

required for the excess biological P removal mechanism in the subsequent activated process, are produced. This form of pretreatment partially compensates for the problems associated with the operation of plants designed for excess biological P removal that treat low organic strength wastewaters, as typically found in North America. It should be noted that in areas having warm temperate climates and sewer lines with long hydraulic detention times, a considerable degree of this pre-treatment may occur in the sewage collection system, prior to the wastewater entering the treatment plant. Extensive investigation of the nature of the biodegradable COD present in a waste stream, with particular reference to the specific substrates required by the excess biological P removal mechanism should, therefore, be carried out in the planning stages of all plants in which this mechanism is to be encouraged. This shift in emphasis towards the importance of characterizing the biodegradable components of the influent COD has, to a certain extent, made influent characterization in terms of the P/COD and TKN/COD ratios largely invalid, as these ratios are based on the total COD present in the wastewater and disregard the chemical nature of the biodegradable COD components. A rational design procedure that includes the recent understanding of the prerequisites of excess biological P removal with regard to the specific substrate required for the excess biological P removal mechanism to operate is, therefore, strongly recommended.

Primary sludge fermentation has been used to enhance the P removal characteristics of a number of full-scale nutrient

removal processes. Oldham (1984) presented data on the benefits of discharging liquor from the primary sludge thickener into the anaerobic zone of the 5-stage Bardenpho plant at Kelowna B.C., Canada. The plant consists of two parallel modules and when all of the VFA-rich fermenter liquor (the thickener sludge blanket height is controlled in an attempt to maximize the VFA production) was alternately discharged into one of the two modules, the module receiving the liquor discharge immediately exhibited a greater degree of anaerobic P release and excellent system P removal within a few days. At the same time, the anaerobic P release and system P removal rapidly declined in the module from which the thickener liquor discharge was withdrawn. In the final stages of the experiment, when the thickener liquor was evenly distributed between the two modules, effluent ortho-P concentrations of less than 1 mg/L were achieved on both sides of the plant within about 4 days of operation. The plant has been operated successfully using this form of primary sludge fermentation, with mean effluent total P concentrations of less than 1 mg/L being achieved for extended periods of time. This is in spite of the fact that application of the UCT biological P removal model predicts that no excess biological P removal will occur in the plant treating raw sewage (Barnard, 1985). Nicholls et al. (1984) found that it was possible to generate 73 mg/L of VFA (expressed as mg HAc per litre of influent flow) by fermenting primary sludge in an acid digester with a 3 day retention time at the Northern Works 5-stage Bardenpho plant in Johannesburg, South Africa. By discharging the supernatant from

the acid digester into the anaerobic zone of the process, they were able to achieve an effluent ortho-P concentration of 2.2 mg/L (influent total P = 17.5 mg/L) in the process. By measuring the "readily biodegradable COD" concentration in the anaerobic zone of the process using the denitrification method outlined in their paper, Nicholls et al. were able to demonstrate the disappearance of the substrate under anaerobic conditions and the perceived role of the substrate in the excess biological P removal mechanism.

8.6. Future Design and Operational Considerations

In this research it was clearly demonstrated that the P removal characteristics of the nutrient removal process can be significantly enhanced by the incorporation of primary sludge fermentation into the process design. The use of primary sludge fermentation, therefore, shows great potential as a viable method of generating the specific substrates required to ensure that excess biological P removal takes place in future activated sludge processes and in existing plants that undergo retrofitting for such removal. Primary sedimentation and primary sludge digestion are commonly incorporated into the design of conventional activated sludge processes as a means of reducing the organic loading and, therefore, the oxygen requirements of the process. In such cases, the addition of primary sludge fermentation to produce VFA's on-site for excess biological P removal would be a relatively simple modification. In the planning of new plants, the primary sludge treatment equipment can easily be designed to provide for acid fermentation, with

the acid-rich fermenter liquor being discharged into the
anaerobic zone of the process. Existing plants, particularly
those already having primary clarification, can easily be
retrofitted to include these design features by adapting the
existing primary sludge treatment facilities in order to optimize
VFA production. The aeration tank of the main process would have
to be modified to include an anaerobic zone, i.e. a zone that
receives no dissolved oxygen and minimal nitrate loading in the
sludge recycle, if nitrification is anticipated in the aerobic
zone of the process. This zone would receive the fermenter
liquor which would then be available solely for the excess
biological P removal mechanism. To this end, it is important
that the process configuration be designed either on the
principle of the UCT process, in which a zero nitrate discharge
to the anaerobic zone can be guaranteed, or that it have a
2-stage anaerobic zone in which complete denitrification occurs
in the first stage and the fermenter liquor is discharged into
the second stage of the unaerated zone of the process. In this
way, organisms having the ability to store excess P under aerobic
conditions, and to store carbonaceous substrate in the form of
PHB under anaerobic conditions, would have a decided advantage in
the highly competitive activated sludge environment. Additional
research is still required to investigate the feasibility of
using this technique in high rate activated sludge processes,
i.e. those designed specifically not to nitrify. This would
eliminate the problems associated with the utilization of the
substrates required for the excess biological P removal mechanism

in the denitrification reaction. Ultimately, the decision to include primary sludge fermentation into the design of new works, or the retrofitting of existing plants, is one that will be made on economic considerations. The anticipated additional capital and operating costs must be carefully weighed against the costs of alternative methods of enhancing the P removal characteristics of the activated sludge process, e.g. chemical precipitation by the in-plant addition of metal salts, addition of a freely available highly carbonaceous industrial wastewater rich in the preferred substrates for excess biological P removal, sand filtration of the final effluent to reduce the total P concentration, etc.

It is clear that additional research is required into the kinetics of the fermentation of primary sludge, particularly with regard to the interrelationship of the sludge age and temperature control parameters. It seems likely that a sludge age of between 2 and 6 days would suffice and that strict control of the sludge age would be required to optimize the VFA production for all operating conditions. For example, longer sludge ages may be required to compensate for lower fermenter operating temperatures in the winter months. However, in order to operate a fermenter that receives between 5% and 10% of the influent flow at sludge ages in this range, a sludge age control strategy that is independent of the hydraulic detention time of the fermenter is going to be required in order to avoid inordinately large fermenter volumes. This sludge age control strategy requires that the fermenter have its own secondary clarifier and sludge

return recycle and that fermenter liquor be wasted daily in some
controlled fashion, as was carried out at the pilot-scale in the
research reported in this thesis. An alternative method of
fermenter sludge age control, which eliminates the requirement
for a fermenter secondary clarifier, is schematically presented
in Fig. 8.2. In this proposed configuration, primary sludge is
pumped from the bottom of the primary clarifier into a completely
mixed anaerobic primary sludge fermenter, whose liquid surface
level is higher than that of the primary clarifier. The
fermenter mixed liquor is returned by gravity, together with raw
influent, back to the primary clarifier, which would have to be
designed to handle the increased solids loading. The fermenter
sludge age would have to be hydraulically controlled by wasting
the appropriate volume of mixed liquor from the reactor daily,
settling out and wasting the solids, and discharging the
fermenter supernatant into the fermenter effluent stream. A
major perceived advantage of this approach over the "sidestream"
method used in this thesis is that all of the influent flow is
brought into contact with the fermented solids, thereby
maximizing the opportunity for VFA production in the primary
clarifier and in the fermenter itself. In addition, this method
allows for the removal of the primary sludge from the wastewater,
thereby decreasing the dissolved oxygen requirements of the
process, while at the same time, the VFA content of the settled
sewage is maximized, thereby enhancing the P removal
characteristics of the process. This design strategy is similar
in concept to the "activated primary tanks" recommended by

PRIMARY
SLUDGE
FERMENTER

FERMENTER
EFFLUENT
COMBINED
WITH RAW
INFLUENT

FERMENTER MIXED
LIQUOR WASTAGE—
DETERMINES FERMENTER SRT

PRIMARY
CLARIFIER

ACTIVATED SLUDGE PROCESS CONFIGURATION
DESIGNED FOR ENHANCED BIOLOGICAL P REMOVAL

INFLUENT

VFA-RICH SETTLED
SEWAGE TO
BIOREACTOR

EFFLUENT

PRIMARY SLUDGE—
PUMPING RATE
DETERMINES
FERMENTER HRT
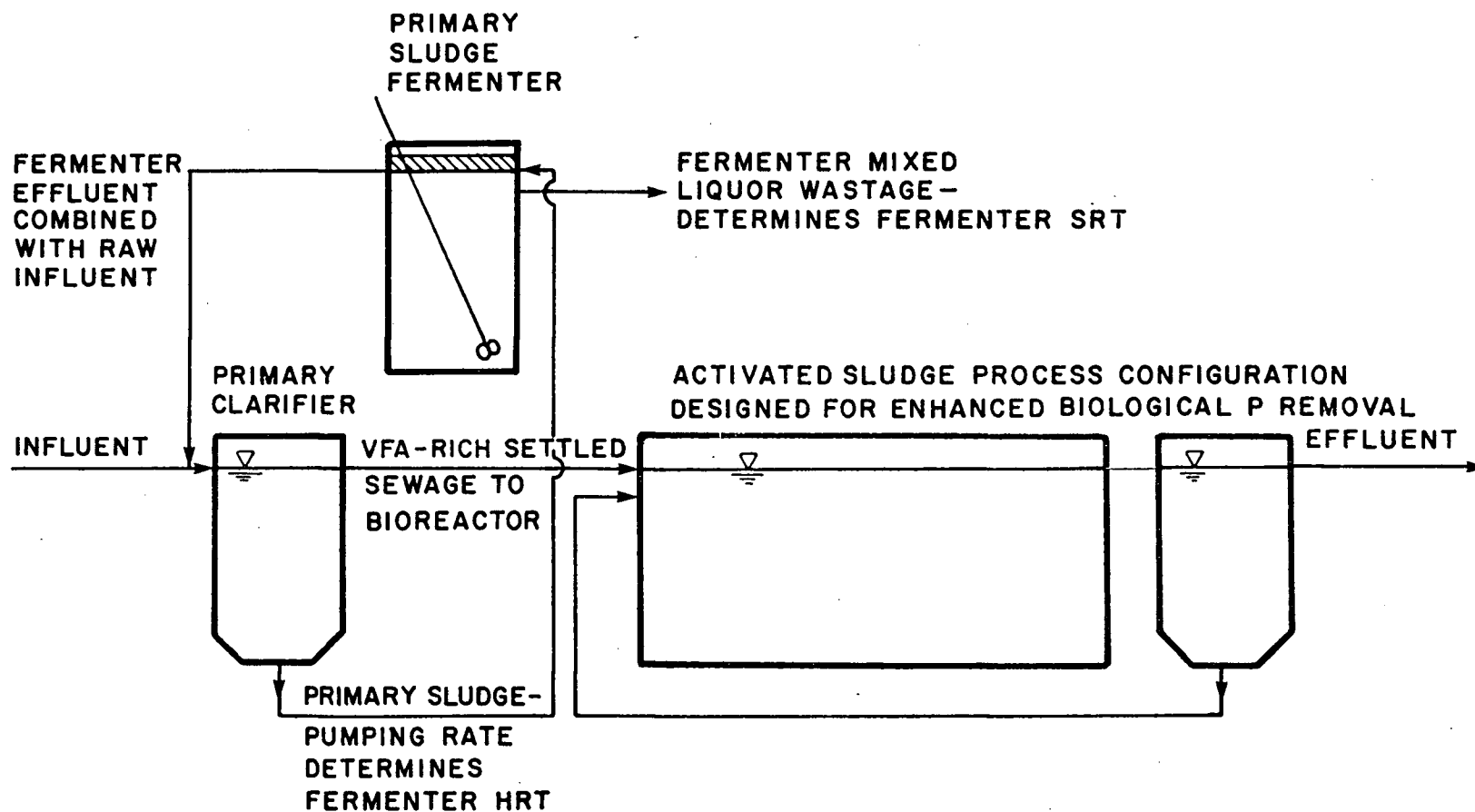
Fig. 8.2. Schematic layout of proposed method of operating
primary sludge fermenter having independent SRT and
HRT control and no fermenter secondary clarifier

Barnard (1984), except that it allows for significantly more stringent control over the operating sludge age of the fermenter, thereby facilitating the optimization of VFA production for different operating conditions. With regard to the most suitable activated sludge process configuration for this proposed strategy, it is recommended that the UCT process be chosen as it guarantees a zero nitrate discharge to the anaerobic zone at the head end of the process and, therefore, all of the VFA generated by primary sludge fermentation will be available for the excess biological P removal mechanism. If, however, relatively low effluent nitrate concentrations of around 3 mg N/L are anticipated, the Bardenpho-type process configurations will also be suitable, as the mass of substrate required for denitrification should not significantly affect the P removal characteristics of the process.

The results of this research also demonstrate the importance of the dry weight percent P content of the sludge as a potential process control parameter. Because daily sludge wastage from the aerobic zone is the principle method for removing P from the process, it follows that the P content of the sludge in the aerobic zone of the process is an excellent measure of the degree to which the process is exhibiting excess biological P removal. Processes not exhibiting any excess biological P removal are generally found to have a sludge dry weight percent P content of 1.5%, i.e. the P requirements for basic metabolic purposes (Hoffmann and Marais, 1977). Fig. 8.3 shows, on the basis of a P mass balance of the process, how the mean dry weight
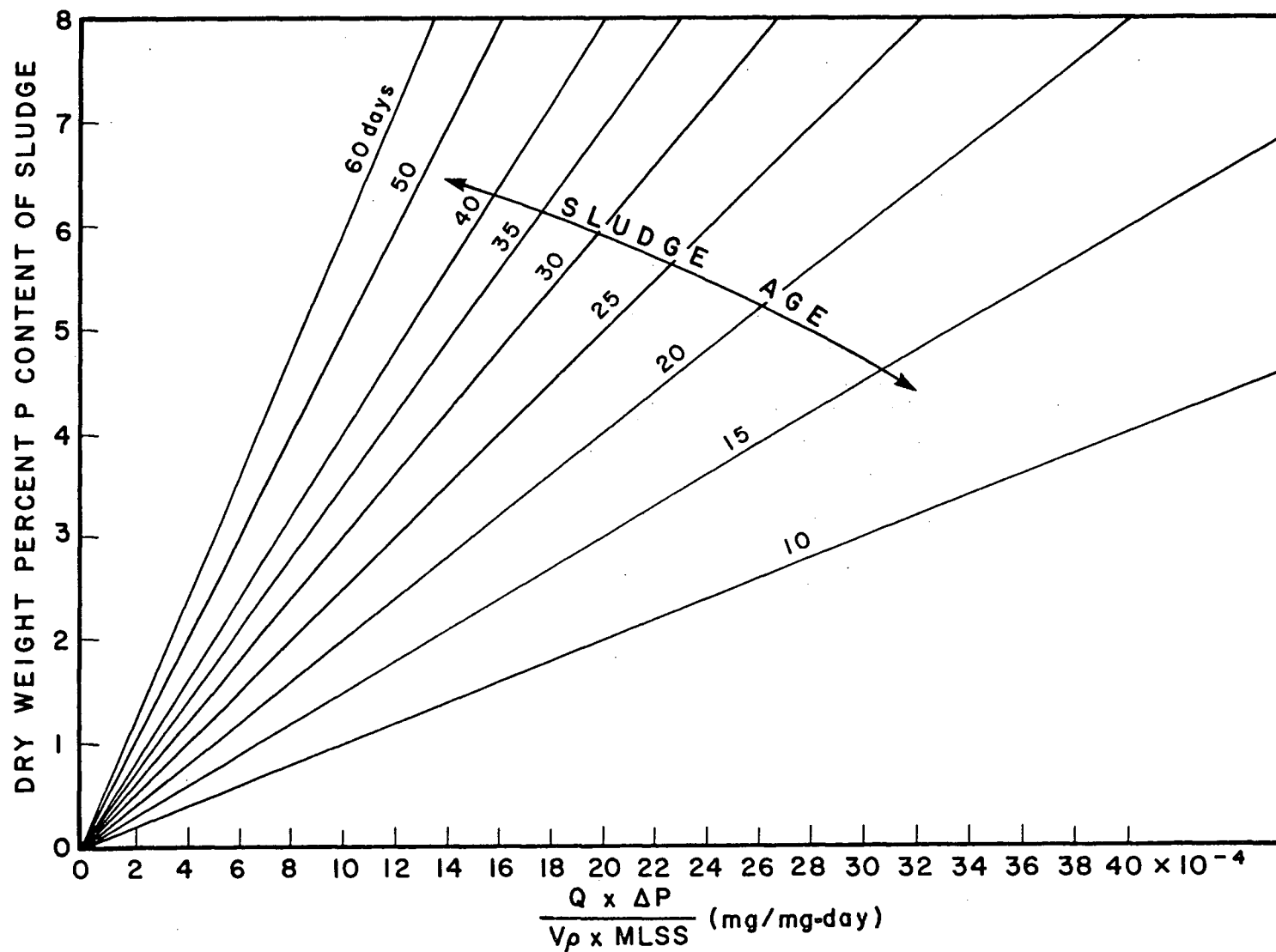
Fig. 8.3. Mass balance chart showing the dependence of the dry weight
percent P content of the sludge on various process parameters
and the sludge age, where:
Q=daily influent flow (L/D); ΔP=system P removal (mg/L);
Vp=process volume (L); MLSS=mixed liquor suspended solids (mg/L).

percent P content of the sludge varies with the mass of system P removal per unit mass of sludge in the process, as a function of the sludge age of the process. For example, for a process to exhibit a system P removal of 20 X 10^{-4} mg P per mg of MLSS per day in a process being operated at a sludge age of 15 days, the mean dry weight percent P of the sludge would have to be 3.0%. However, for a process to exhibit the same degree of system P removal per day per unit mass of sludge at a 25 day sludge age, the dry weight percent P content of the sludge would have to be 5.0%. It can thus be seen that, for a given set of operating conditions, the ability of a process to remove P from the wastewater is limited by the maximum attainable P content of the sludge. In general, reducing the sludge age of a process will have the following effects:

1) The mass of sludge in the process and, therefore, the MLSS concentration will decrease. However, the active sludge fraction will increase (Marais and Ekama, 1976).
2) The required P content of the sludge for a given system P removal will be reduced.

For these reasons, it is recommended that the sludge age of a process designed for excess biological P removal be kept as low as possible, in order to enhance the P removal characteristics of the process. The lower limit could be determined by the settling, dewatering and handling characteristics of the sludge (problems in sludge handling are often attributed to a high active sludge fraction), and the nitrifying characteristics of

the process, taking into account the lower growth rate of the nitrifiers and the negative effect of the unaerated sludge mass fraction.

CHAPTER NINE

CONCLUSIONS AND RECOMMENDATIONS

Significant progress has been made in biological P removal technology during the course of this study. The research reported in this thesis has contributed towards a greater understanding of the P removal mechanism and how it may be applied in the design of the nutrient removal activated sludge process. The conclusions of this research may be summarized as follows:

1) Anaerobic batch tests, using sludge from plants exhibiting excess biological P removal, have revealed that different soluble carbonaceous substrates exhibit a range of effectiveness in inducing anaerobic P release when COD is used as the control parameter. The effectiveness in inducing anaerobic P release appears to be a function of the complexity of the substrate used with the simpler substrates, sodium acetate and propionic acid, being the most effective and the more complex substrates, such as glucose and butyric acid, being significantly less effective.

2) The results of an anaerobic batch test using denitrified activated sludge and a range of initial sodium acetate concentrations between 0 and 100 mg/L (as COD) clearly show that P release took place in two distinct phases. Initally, there was a rapid release phase in which the release rate appeared to be independent of the initial substrate concentration but in which the mass of P released is a function of the mass of substrate

available. This was followed by a significantly lower release
rate which continued for the duration of the anaerobic phase.
Furthermore, there appears to be an upper limit to the mass of P
that a given sludge can release, beyond which additional
substrate will not cause any additional release. This upper P
release limit appears to be some function of the mass of
intracellularly stored poly-P that is available for release under
suitable anaerobic conditions, and the MLSS concentration of the
sludge used in the batch test.

3) The results of batch tests in which the disappearance of
acetate from the supernatant was monitored during the P release
phase clearly indicate that anaerobic P release and substrate
utilization are integral parts of the same mechanism. There
appears to be some form of a direct exchange phenomenon occurring
between the two molecules at a molar exchange ratio of 1.76 moles
of P released per mole of acetate utilized. (On the basis of
mass, this exchange ratio is equivalent to 0.91 mg P per mg of
acetate (as HAc)). This observation supports the proposed
biochemical kinetic model for the excess biological P removal
mechanism of Comeau (1985b). In this model, the function of P
release is to facilitate the transport of acetate across the cell
wall by maintaining the proton motive force of the cell. The
carbonaceous substrate that is taken up by the cells under
anaerobic conditions is stored as poly-B-hydroxybutyrate (PHB).

4) In monitoring the ORP of anaerobic batch tests, it was not
possible to define the prerequisite conditions for anaerobic P

release in terms of the mixed liquor ORP. In all cases where ORP was monitored, P release commenced immediately upon substrate addition and some time before a minimum level of ORP was established in the experimental flasks. This may, however, largely be a function of the probe response time and be peculiar to batch testing conditions. With regard to the effect of nitrate on the ORP level in anaerobic flasks, the disappearance of nitrate is marked by a significant increase in the rate of change of ORP, resulting in a "bend" in the ORP vs time curve. These two aspects of the biological P removal research program carried out at UBC, together with the potential of ORP as a monitoring and control parameter in continuous flow nutrient processes, are dealt with by Koch and Oldham (1984).

5) In batch tests designed to quantify the negative effect of nitrate on the anaerobic P release mechanism, it was shown that the effect was not as great as that predicted by Siebritz et al. (1982, 1983), i.e. that any available substrate is utilized preferentially in the denitrification reaction at a rate of 8.6 mg COD/mg NO_3-N and is thus rendered unavailable for the P release mechanism. In batch tests where excess substrate was available to the organisms, the presence of up to 9 mg/L of nitrate (as N) had no detrimental effect on P release. However, in a subsequent batch test in which the availability of substrate was more limited, the available substrate appeared to be utilized preferentially in the denitrification reaction at a rate of 3.6 mg COD/mg NO_3-N, calculated on the basis of the detrimental effect of nitrate on the P release mechanism.

6) In experiments where a sodium acetate solution was added to the unaerated zone of the pilot-scale activated sludge process, the P removal characteristics, as measured by the $\Delta P/\Delta COD$ ratio of the process and the dry weight percent P content of the sludge, showed significant improvement. For example, the addition of 86 mg/L of acetate (measured as mg of COD per litre of influent) to the influent end of a simplified nutrient removal process improved the $\Delta P/\Delta COD$ ratio from 0.0069 to 0.0102 and increased the dry weight percent P content of the sludge from 1.46% to 2.34%. Furthermore, when acetate was added to an unaerated zone that received no incoming nitrate concentration, significantly greater efficiency of the added substrate was made by the organisms of the process for the excess biological P removal mechanism. The addition of 39 mg COD/L of acetate to such a zone resulted in a $\Delta P/\Delta COD$ of 0.0142 and a dry weight percent P content of the sludge of 2.54%.

7) Operation of the pilot-scale primary sludge fermenter demonstrated that the two most effective substrates in inducing anaerobic P release in activated sludge, acetate and propionate, are also the principle products of primary sludge fermentation, making up more than 95% of the total short-chain VFA production. The acetate:propionate production ratio was found to be approximately 55:45 and this ratio appears to independent of sludge age, at least for sludge ages between 2.1 and 10 days. The best yield of total VFA produced by primary sludge fermentation was found to be approximately 0.09 mg VFA (as HAc) per mg of primary sludge (as COD). This optimum yield was

achieved at fermenter sludge ages of between 3.5 and 5.0 days with slightly lower yields being attained at both shorter and longer sludge ages. However, it may be possible to partially explain the lower VFA yields in terms of the lower fermenter operating temperatures and MLSS concentrations during some experimental periods. Decreasing the mean fermenter temperature from about $19^{\circ}C$ to about $13^{\circ}C$ resulted in a drop in VFA production of approximately 20%, a value significantly less than that predicted by the general theory of microbial activity. Increasing the mean fermenter pH from about 5.7 to near neutrality by the addition of sodium hydroxide did not result in an improvement in VFA production. However, these experiments need to be repeated under conditions that are more stringently controlled.

8) Volatile fatty acids can be produced by primary sludge fermentation on-site at an activated sludge treatment plant in sufficient quantities to significantly improve the P removal characteristics of the process. The incorporation of primary sludge fermentation into the design of the nutrient removal process shows great potential for both future plants and the retrofitting of existing plants. For the two pilot-scale activated sludge process configurations used in this research, the incorporation of primary sludge fermentation into the process design significantly improved the P removal characteristics of both processes. For example, the incorporation of primary sludge fermentation into a simplified nutrient removal process improved the $\Delta P/\Delta COD$ of the process from 0.0069 to 0.0141 and the dry

weight percent P content of the sludge from 1.46% to 3.20%.
Incorporation of primary sludge fermentation into the UCT process
improved the $\Delta P/\Delta COD$ ratio from 0.0086 to 0.0136 and the dry
weight percent P content of the sludge from 2.11% to 3.10%.

Further research work is recommended in the following areas:

1) A pilot-scale study of the proposed method of primary sludge
fermenter operation, with independent SRT and HRT and no
fermenter secondary clarifier, as outlined in Chapter Eight
(Section 8.6).

2) A comprehensive pilot-scale study of the kinetics of primary
sludge fermentation, with particular reference to the effects of,
and the accurate control of the sludge age, temperature, pH and
MLSS concentration.

3) An economic evaluation of the enhancement of the P removal
characteristics of the activated sludge process by primary sludge
fermentation, with particular reference to operation of the
fermenter in the mesophilic temperature range.

4) A series a batch testing, using such techniques such as
radioactive isotope labeling and PHB extraction, designed to
accurately determine the fate of the substrate in the excess
biological P removal mechanism and in the denitrification
reaction.

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APPENDIX A1

RAW DATA FROM CHAPTER FIVE

PILOT-SCALE SUBSTRATE ADDITION

COD, Total P and TKN Raw Data from Section 5.1.1

Day #	Inf. COD (mg/L)	Eff. COD (mg/L)	Inf. P (mg/L)	Eff. P (mg/L)	Inf. TKN (mg/L)	Eff. TKN (mg/L)	MLSS (mg/L)
1	-	-	-	-	-	-	-
2	-	-	-	-	-	-	-
3	-	-	3.54	2.12	-	-	-
4	-	-	3.48	2.12	-	-	-
5	183	-	3.33	2.30	21.6	-	-
6	214	-	3.32	2.32	19.4	-	2950
7	206	-	-	-	19.8	-	-
8	253	-	-	-	20.2	-	-
9	409	-	4.93	2.84	23.7	-	2900
10	152	-	4.63	2.87	21.6	-	2850
11	362	-	4.59	2.68	21.9	-	2880
12	206	-	3.44	2.70	21.9	-	2870
13	310	-	4.37	2.80	18.7	-	2730
14	302	-	-	-	23.4	-	-
15	226	-	-	-	20.5	-	-
16	282	-	4.35	3.12	23.8	-	2500
17	190	-	4.20	2.90	16.8	-	2580
Mean	253	-	4.02	2.62	21.02	-	2783

COD, Total P and TKN Raw Data from Section 5.1.2

Day #	Inf. COD (mg/L)	Eff. COD (mg/L)	Inf. P (mg/L)	Eff. P (mg/L)	Inf. TKN (mg/L)	Eff. TKN (mg/L)	MLSS (mg/L)
1	250	-	3.52	2.60	20.7	-	2575
2	218	-	2.94	2.02	18.9	-	2975
3	306	-	-	1.85	18.3	-	3200
4	-	-	-	-	20.0	-	-
5	-	-	-	-	-	-	-
6	204	67	3.30	2.20	20.0	1.8	2560
7	222	-	3.50	2.60	19.3	-	2960
8	214	-	3.70	1.90	19.4	-	3140
9	190	69	3.60	1.80	20.0	4.0	-
10	169	-	3.40	2.10	-	-	-
11	-	-	-	-	-	-	-
12	-	-	-	-	-	-	-
13	314	79	4.50	2.40	23.4	<1.0	2220
14	-	-	4.60	2.40	-	-	-
15	259	-	4.00	2.50	-	-	2830
16	371	71	5.50	2.40	28.6	2.6	2660
Mean	247	71	3.87	2.23	20.86	2.23	2791

COD, Total P and TKN Raw Data from Section 5.1.3

Day #	Inf. COD (mg/L)	Eff. COD (mg/L)	Inf. P (mg/L)	Eff. P (mg/L)	Inf. TKN (mg/L)	Eff. TKN (mg/L)	MLSS (mg/L)
1	320	42	4.70	<0.10	25.5	1.4	3474
2	173	-	3.00	<0.10	15.5	0.7	3284
3	121	-	2.40	<0.10	15.2	1.4	3188

COD, Total P and TKN Raw Data from Section 5.1.3 (cont.)

Day #	Inf. COD (mg/L)	Eff. COD (mg/L)	Inf. P (mg/L)	Eff. P (mg/L)	Inf. TKN (mg/L)	Eff. TKN (mg/L)	MLSS (mg/L)
4	175	44	2.60	<0.10	16.7	1.4	2998
5	184	–	2.40	<0.10	15.5	1.5	3000
6	200	–	2.60	<0.10	20.6	1.4	–
7	223	–	3.20	<0.10	22.5	1.3	–
8	247	51	3.60	<0.10	22.0	1.2	3028
9	257	–	3.40	<0.10	20.8	1.2	2895
10	273	–	3.40	<0.10	20.6	1.1	3059
11	206	55	2.90	<0.10	18.9	1.0	–
12	221	–	3.40	<0.10	20.1	1.2	–
13	220	–	2.90	<0.10	19.7	1.5	–
14	159	–	2.90	<0.10	22.6	1.2	–
15	347	92	3.60	<0.10	19.8	1.3	3013
16	214	–	3.00	<0.10	19.1	1.4	3037
17	269	–	3.00	<0.20	18.9	1.1	2968
18	221	73	2.80	<0.20	18.5	0.8	2919
19	167	–	2.50	<0.20	17.2	1.2	2988
20	202	–	2.70	<0.20	18.4	0.9	–
21	206	–	3.30	<0.20	20.2	1.0	–
22	255	60	4.30	<0.20	24.6	1.1	2829
23	253	–	3.80	0.20	22.9	1.5	2837
24	266	–	3.10	0.20	20.3	1.2	2925
25	268	55	3.70	0.20	21.8	1.8	2939
26	198	–	3.50	0.40	23.3	1.2	2839
27	268	–	3.60	<0.20	21.7	0.9	–
28	255	–	3.30	0.30	19.9	1.1	–
29	237	48	3.40	<0.20	20.2	1.2	2705
30	200	–	3.30	0.20	19.2	0.8	2803
31	176	–	2.80	<0.20	20.8	1.2	–
32	281	40	3.30	–	23.9	–	2775
33	192	–	3.30	0.40	26.2	1.2	2691
34	212	–	3.60	0.50	20.8	1.0	–
35	225	–	3.60	0.70	21.3	1.0	–
36	194	40	3.40	0.70	20.4	1.1	2582
37	185	–	3.00	0.90	20.4	1.1	2592
38	161	–	2.60	0.50	20.7	1.5	–
39	189	56	3.20	0.70	22.2	1.6	–
40	182	–	3.50	1.30	23.0	1.4	2524
41	194	–	3.90	1.80	24.3	1.5	–
42	220	–	4.20	1.90	22.7	1.5	–
43	225	53	3.80	1.90	23.2	1.5	2482
44	222	–	3.70	2.30	22.6	1.5	2413
45	227	–	3.20	2.20	20.7	1.7	2448
46	220	53	3.10	2.00	20.0	1.6	2375
47	207	–	2.90	1.90	20.3	1.8	2296
48	289	–	3.20	1.70	20.8	2.0	–
49	261	–	3.50	1.90	22.8	2.1	–
50	296	82	4.20	2.40	25.2	2.1	–
51	296	–	4.10	2.20	25.5	2.3	2314
52	244	–	3.80	2.10	26.7	2.6	2364
53	235	72	3.30	1.60	25.8	2.8	2187

COD, Total P and TKN Raw Data from Section 5.1.3 (cont.)

Day #	Inf. COD (mg/L)	Eff. COD (mg/L)	Inf. P (mg/L)	Eff. P (mg/L)	Inf. TKN (mg/L)	Eff. TKN (mg/L)	MLSS (mg/L)
54	217	-	3.60	1.60	20.8	1.8	-
55	215	-	3.40	1.40	21.3	2.0	-
56	227	-	2.10	1.30	21.7	2.4	-
57	245	82	3.70	1.40	24.2	2.7	-
58	239	-	3.40	1.20	24.6	2.8	2191
59	240	-	3.50	0.80	22.4	2.1	2164
60	218	-	3.40	0.70	23.2	1.6	2240
61	242	-	3.80	0.70	26.0	2.6	-
Mean	226	59	3.32	0.73	21.42	1.49	2732

COD, Total P and TKN Raw Data from Section 5.1.4

Day #	Inf. COD (mg/L)	Eff. COD (mg/L)	Inf. P (mg/L)	Eff. P (mg/L)	Inf. TKN (mg/L)	Eff. TKN (mg/L)	MLSS (mg/L)
1	204	-	2.90	0.83	18.4	4.3	-
2	255	51	3.80	0.41	23.8	3.6	2640
3	247	-	3.50	0.34	22.4	2.5	2830
4	204	51	3.20	0.30	19.2	<1.0	2410
5	231	-	3.40	0.18	18.7	2.0	2620
6	210	-	3.90	0.24	21.4	1.0	-
7	-	-	-	0.20	-	1.2	-
8	182	21	2.90	0.70	15.9	<1.0	2810
9	231	-	3.40	0.85	18.0	<1.0	2900
10	222	-	3.50	0.85	19.3	<1.0	3100
11	191	51	2.80	0.70	18.4	1.3	2950
12	207	-	2.70	0.62	21.3	1.4	2910
13	168	-	3.00	0.64	21.3	1.5	-
14	204	-	3.10	0.84	19.8	1.8	-
15	235	28	4.40	0.93	21.0	1.1	-
16	217	-	3.50	0.67	19.5	0.5	3000
17	181	-	2.70	0.38	17.7	0.8	3100
18	114	28	1.90	0.25	12.8	1.2	3110
19	284	-	2.70	0.23	20.5	1.0	-
20	165	-	2.40	0.23	17.7	1.3	-
21	198	-	3.20	0.30	18.2	1.8	-
22	189	35	3.30	0.20	11.3	1.3	3220
23	198	-	2.90	0.30	18.3	2.0	-
24	188	-	2.80	0.10	19.0	0.7	3290
25	163	72	2.60	0.10	16.4	0.6	3300
26	181	-	3.00	0.20	17.4	1.0	3200
27	170	-	2.90	0.10	16.7	1.5	-
28	170	-	3.40	0.10	19.9	0.6	-
29	237	35	3.90	0.10	21.5	0.8	3150
30	239	-	3.40	0.10	19.0	0.8	3270
31	222	-	3.10	<0.10	19.6	0.7	3080
32	181	31	2.30	-	17.4	-	3130
33	178	-	2.20	<0.10	15.5	0.5	3120
34	181	-	2.50	<0.10	12.6	0.7	-
35	229	-	3.10	<0.10	18.1	0.7	-

COD, Total P and TKN Raw Data from Section 5.1.4

Day #	Inf. COD (mg/L)	Eff. COD (mg/L)	Inf. P (mg/L)	Eff. P (mg/L)	Inf. TKN (mg/L)	Eff. TKN (mg/L)	MLSS (mg/L)
36	219	25	3.30	<0.05	17.3	0.5	–
37	175	–	3.40	<0.10	17.9	0.8	3210
38	210	–	2.90	<0.10	17.9	0.6	3190
39	239	36	2.70	<0.05	24.2	0.7	3260
40	205	–	2.80	<0.05	21.7	0.7	–
Mean	203	39	3.06	0.32	18.64	1.12	3033

COD, Total P and TKN Raw Data from Section 5.2

Day #	Inf. COD (mg/L)	Eff. COD (mg/L)	Inf. P (mg/L)	Eff. P (mg/L)	Inf. TKN (mg/L)	Eff. TKN (mg/L)	MLSS (mg/L)
1	309	–	4.10	0.78	30.3	2.3	2660
2	333	–	3.70	0.64	27.8	1.2	2790
3	279	56	4.40	0.78	27.7	1.3	2880
4	229	–	3.40	0.62	21.7	1.3	2540
5	245	–	4.00	–	27.7	–	–
6	276	–	4.00	–	27.5	–	–
7	266	54	4.40	0.30	29.6	0.9	2110
8	235	–	3.90	0.20	25.4	<0.5	–
9	284	–	4.40	0.20	27.8	0.8	2720
10	278	–	4.30	0.52	28.3	0.8	3170
11	276	–	–	0.14	–	0.7	–
12	260	–	4.70	–	28.2	–	–
13	264	–	4.90	–	28.8	–	–
14	266	47	4.70	0.30	28.9	1.3	2910
15	254	–	3.70	0.30	23.8	1.3	2880
16	223	–	2.97	0.27	23.0	1.4	–
17	207	–	2.56	0.66	19.3	1.0	2880
18	225	–	2.82	0.13	19.0	1.4	2870
19	211	–	3.06	–	19.8	–	–
20	338	–	4.43	–	26.8	–	–
21	264	49	3.82	0.29	23.6	1.4	3060
22	281	–	4.50	<0.20	24.8	1.3	3040
23	215	–	4.00	<0.20	23.5	0.9	3130
24	214	37	3.40	0.20	22.4	0.9	2960
25	195	–	3.50	<0.20	21.4	0.8	3040
26	216	–	3.50	<0.20	22.3	1.0	–
27	322	–	4.40	<0.20	26.2	0.5	–
28	216	39	3.90	<0.20	23.7	0.5	2700
29	261	–	3.90	<0.20	25.0	1.1	2512
30	234	–	3.60	0.40	22.8	0.8	2237
31	230	23	2.40	0.20	19.5	1.0	2100
32	203	–	2.80	0.40	21.7	1.0	2140
33	335	–	4.00	–	25.8	1.1	–
34	266	–	4.30	–	24.1	1.4	–
35	281	75	3.90	–	22.9	5.8	–
36	349	–	5.60	0.60	31.2	4.7	2627
37	298	–	4.00	0.90	25.3	5.6	3814
38	252	45	4.30	–	30.2	–	3900

COD, Total P and TKN Raw Data from Section 5.2 (cont.)

Day #	Inf. COD (mg/L)	Eff. COD (mg/L)	Inf. P (mg/L)	Eff. P (mg/L)	Inf. TKN (mg/L)	Eff. TKN (mg/L)	MLSS (mg/L)
39	291	–	3.70	–	24.5	3.4	3528
40	244	–	4.10	–	24.9	–	–
41	299	–	5.80	–	34.5	–	–
42	272	–	4.80	0.20	28.5	0.8	3857
43	256	–	4.30	<0.20	26.2	0.8	3625
44	209	–	3.70	0.24	26.6	2.1	3710
45	179	–	3.30	0.12	22.3	0.8	3771
46	175	–	3.00	<0.10	19.8	1.0	3457
47	185	–	3.20	–	20.8	–	–
48	229	–	3.90	–	23.2	–	–
49	388	20	4.00	<0.10	24.6	0.7	3428
50	227	–	5.40	<0.10	23.2	0.7	3514
51	277	–	3.90	<0.10	28.0	1.1	3628
52	324	–	4.50	0.50	30.2	1.0	3635
53	340	39	4.30	0.24	29.7	1.9	–
54	293	–	5.00	–	29.8	–	–
55	389	–	4.90	–	31.6	–	–
56	347	–	4.20	0.28	27.3	–	–
57	341	–	3.90	0.15	28.5	1.2	–
58	379	–	4.30	<0.10	35.5	1.1	3900
59	291	–	4.30	<0.10	40.3	1.0	4057
Mean	268	44	4.01	0.28	26.00	1.41	3105

Ortho-P and Nitrate Bioreactor Profiles from Section 5.1.1

	Ortho-P (mg/L)				NO$_3$ + NO$_2$ (mg/L)			
Day #	1st Unaer.	2nd Unaer.	Aerobic	Eff.	1st Unaer.	2nd Unaer.	Aerobic	Eff.
3	2.12	2.62	2.21	2.23	0.95	0.13	10.20	10.30
4	2.20	2.20	2.20	2.10	2.20	0.13	7.28	7.60
6	2.40	2.64	2.35	2.42	1.60	0.13	7.68	7.68
9	3.26	3.57	2.92	2.94	0.33	0.18	4.56	4.64
Mean	2.50	2.76	2.42	2.42	1.27	0.14	7.43	7.56

Ortho-P and Nitrate Bioreactor Profiles from Section 5.1.2

	Ortho-P (mg/L)				NO$_3$ + NO$_2$ (mg/L)			
Day #	1st Unaer.	2nd Unaer.	Aerobic	Eff.	1st Unaer.	2nd Unaer.	Aerobic	Eff.
2	2.40	2.70	2.40	2.50	–	–	–	–
4	2.40	2.80	1.80	1.80	–	–	–	–
7	2.60	3.20	2.30	2.10	–	–	–	–
9	2.80	3.80	1.80	1.70	<0.20	<0.20	4.70	2.60
16	3.42	4.58	2.34	2.25	0.21	0.08	5.28	4.84
Mean	2.72	3.42	2.13	2.07	0.15	0.09	4.99	3.72

Ortho-P and Nitrate Bioreactor Profiles from Section 5.1.3

| | Ortho-P (mg/L) | | | | NO$_3$ + NO$_2$ (mg/L) | | | |
| | 1st | 2nd | | | 1st | 2nd | | |
Day #	Unaer.	Unaer.	Aerobic	Eff.	Unaer.	Unaer.	Aerobic	Eff.
1	6.20	7.40	1.20	0.03	<0.01	0.05	3.10	3.30
4	3.80	5.00	0.03	0.02	0.02	0.03	4.00	4.00
8	3.90	4.60	0.04	0.06	0.12	0.10	2.80	3.10
11	4.50	5.40	0.06	0.05	0.07	0.05	2.70	2.70
15	4.10	5.10	0.03	0.04	0.01	0.01	3.90	3.80
18	5.10	5.80	0.14	0.08	0.05	0.07	3.40	3.40
22	3.90	5.70	0.30	0.08	0.10	0.03	3.90	3.90
25	3.07	4.17	0.24	0.10	0.03	0.02	3.56	4.27
30	6.00	7.70	0.40	0.20	0.10	0.05	2.50	2.70
32	6.50	8.50	0.25	0.14	0.09	0.04	4.30	4.60
36	7.20	9.80	1.90	0.52	0.29	0.20	3.80	4.50
43	6.17	8.95	1.87	1.45	0.18	0.17	4.60	4.90
46	6.30	8.80	1.60	1.70	0.20	0.05	3.90	4.10
50	7.00	9.90	1.80	1.80	0.05	0.18	4.50	4.50
53	5.50	7.90	0.97	1.30	0.31	0.06	4.60	3.90
60	6.60	11.40	0.28	0.32	0.32	0.36	6.30	5.30
Mean	5.37	7.26	0.69	0.49	0.12	0.09	3.87	3.94

Ortho-P and Nitrate Bioreactor Profiles from Section 5.1.4

| | Ortho-P (mg/L) | | | | NO$_3$ + NO$_2$ (mg/L) | | | |
| | 1st | 2nd | | | 1st | 2nd | | |
Day #	Unaer.	Unaer.	Aerobic	Eff.	Unaer.	Unaer.	Aerobic	Eff.
2	4.40	3.30	0.33	0.37	0.10	0.04	4.40	4.30
4	4.20	5.90	0.30	0.20	0.05	0.07	4.40	4.40
8	5.30	7.70	0.87	0.60	0.05	0.17	3.90	3.81
11	6.00	7.60	0.88	0.61	<0.01	<0.01	3.90	3.50
15	7.30	8.70	0.97	0.97	0.02	0.02	4.10	4.00
18	5.50	6.20	0.18	0.21	0.03	0.03	2.90	2.40
22	8.42	9.32	0.20	0.15	0.02	0.06	3.70	3.70
25	7.70	8.90	0.06	0.07	0.08	0.14	3.70	2.70
29	7.90	9.10	0.04	0.05	0.01	0.24	3.30	3.10
32	5.80	7.50	0.02	0.05	<0.01	0.03	4.90	3.30
39	8.00	8.40	0.74	<0.02	<0.02	0.08	3.50	3.10
Mean	6.41	7.51	0.42	0.30	0.03	0.08	3.88	3.48

Ortho-P and Nitrate Bioreactor Profiles from Section 5.2

| | Ortho-P (mg/L) | | | | $NO_3 + NO_2$ (mg/L) | | | |
| | 1st | 2nd | | | 1st | 2nd | | |
Day #	Unaer.	Unaer.	Aerobic	Eff.	Unaer.	Unaer.	Aerobic	Eff.
3	3.74	8.00	0.76	0.75	0.19	0.32	6.30	5.40
7	2.70	7.40	0.36	0.35	0.16	0.05	5.80	5.40
14	2.00	11.00	0.42	0.34	0.26	0.15	6.90	6.60
17	0.51	12.00	0.12	0.69	0.12	0.06	6.20	5.20
21	0.50	15.50	0.06	0.06	0.34	0.26	8.00	7.70
24	0.48	16.50	0.07	0.06	0.25	0.08	6.83	7.10
28	0.22	12.00	0.20	0.14	0.23	0.11	6.30	6.30
31	0.21	10.40	0.34	0.24	0.67	0.04	5.80	6.00
38	0.40	8.00	0.03	0.13	0.10	0.13	4.00	3.30
43	0.48	9.10	0.02	0.04	0.30	0.21	5.74	5.62
45	0.10	9.20	0.05	0.05	0.06	0.15	2.94	4.40
49	0.19	8.70	0.03	0.04	0.57	0.03	6.20	6.40
52	0.75	10.70	1.10	0.41	0.06	0.03	6.10	7.20
Mean	0.94	10.65	0.27	0.25	0.25	0.12	5.93	5.89

APPENDIX A2

RAW DATA FROM CHAPTER SIX

PRIMARY SLUDGE FERMENTATION RESULTS

COD, pH and MLSS Raw Data from Section 6.1.1

	COD (mg/L)		pH		MLSS (mg/L)	
Day #	Raw Inf.	Sett.Sewage	Ferm.1	Ferm.2	Ferm.1	Ferm.2
1	214	115	5.90	5.70	−	−
2	285	139	6.15	5.80	−	−
3	297	139	−	−	−	−
4	256	152	6.20	5.95	−	−
5	297	139	6.05	5.95	−	−
6	275	188	6.20	5.95	1230	2305
7	269	179	−	−	−	−
8	−	181	6.15	5.95	1300	2470
9	290	187	6.10	5.90	−	−
10	251	173	6.10	5.90	−	−
11	303	177	6.10	5.90	−	−
12	320	223	6.25	5.95	1950	3210
13	303	179	−	−	−	−
14	307	202	−	−	−	−
15	322	243	6.05	5.85	1510	2120
16	305	220	6.05	5.85	−	−
17	262	195	−	−	−	−
18	322	205	−	−	−	−
19	303	232	6.25	6.05	1060	2360
20	287	172	−	−	−	−
21	−	166	−	−	−	−
22	289	208	6.30	6.00	1160	2170
23	275	176	6.05	5.85	−	−
24	271	259	6.15	5.90	−	−
25	330	−	6.00	5.80	−	−
26	297	185	5.75	6.00	1090	2110
27	309	206	6.00	5.75	−	−
28	333	210	5.95	5.75	−	−
29	279	212	6.05	5.85	1180	2270
30	229	146	−	−	−	−
31	245	158	6.00	5.75	−	−
32	276	174	6.05	5.85	−	−
33	266	180	6.10	5.80	1330	2810
34	235	162	−	−	−	−
35	284	158	6.00	5.70	−	−
36	278	180	5.95	5.65	−	−
37	276	184	6.00	5.70	−	−
38	260	172	6.00	5.70	−	−
39	264	181	5.95	5.75	−	−
40	266	181	6.10	5.80	1830	2710
41	254	152	6.15	5.90	−	−
42	223	133	6.10	5.85	−	−
43	207	129	6.00	5.75	1140	2520
44	225	153	5.90	5.75	−	−
Mean	277	179	6.05	5.85	1344	2460

COD, pH and MLSS Raw Data from Section 6.1.2

Day #	COD (mg/L)		pH		MLSS (mg/L)	
	Raw Inf.	Sett.Sewage	Ferm.1	Ferm.2	Ferm.1	Ferm.2
1	198	110	5.65	5.45	-	-
2	248	116	5.70	5.50	-	-
3	225	146	5.75	5.55	2769	2396
4	247	141	5.75	5.45	-	-
5	210	141	5.80	5.50	-	-
6	178	100	5.65	5.45	2664	2297
7	178	112	5.75	5.50	-	-
8	-	133	5.70	5.45	-	-
9	223	140	5.60	5.30	-	-
10	225	162	5.70	5.45	2093	2232
11	219	140	5.80	5.55	-	-
12	220	137	5.85	5.45	-	-
13	237	141	5.80	5.45	2120	2318
14	208	137	5.90	5.50	-	-
15	225	-	6.00	5.55	-	-
16	235	-	6.05	5.65	-	-
17	266	-	6.05	5.65	1970	2044
18	333	-	6.05	5.70	1800	1853
19	299	145	6.00	5.65	-	-
20	213	84	6.00	5.65	1764	2667
21	204	118	5.95	5.60	-	-
22	223	141	5.95	5.60	-	-
23	383	255	5.95	5.60	-	-
24	257	166	6.00	5.65	1700	1880
25	221	156	5.95	5.55	-	-
26	244	154	6.05	5.65	-	-
27	238	162	6.05	5.80	1640	1880
28	233	94	6.00	5.65	-	-
29	213	102	6.00	5.65	-	-
30	291	141	6.10	5.80	-	-
31	287	129	5.55	5.20	1530	1940
32	217	90	5.95	5.65	-	-
33	221	143	5.95	5.60	-	-
34	254	102	5.90	5.55	-	-
35	204	125	5.90	5.60	1930	2165
36	217	150	5.90	5.60	-	-
37	303	125	5.95	5.60	-	-
38	303	156	5.95	5.60	2030	2020
39	295	160	5.95	5.60	-	-
40	258	139	5.95	5.60	-	-
41	-	164	5.90	5.50	1940	2090
42	272	158	-	-	-	-
43	317	137	5.90	5.55	-	-
44	393	176	6.35	6.20	-	-
45	327	176	5.80	5.45	1410	2110
46	242	142	5.90	5.50	-	-
47	234	138	5.90	5.50	-	-
48	254	156	5.90	5.60	2050	2190
Mean	250	140	5.90	5.60	1961	2139

COD, pH and MLSS Raw Data from Section 6.1.3

Day #	COD (mg/L)		pH		MLSS (mg/L)	
	Raw Inf.	Sett.Sewage	Ferm.1	Ferm.2	Ferm.1	Ferm.2
1	288	-	5.75	5.60	750	810
2	279	171	5.75	5.45	-	-
3	238	156	5.70	5.40	-	-
4	271	171	-	-	-	-
5	240	175	-	-	-	-
6	247	133	-	-	1240	705
7	221	152	-	-	-	-
8	224	152	6.10	5.90	830	320
9	215	136	5.85	5.75	-	-
10	226	151	-	-	-	-
11	245	155	-	-	-	-
12	253	192	5.85	5.65	515	180
13	280	156	5.70	5.45	-	-
14	245	175	5.65	5.45	-	-
15	237	156	5.85	5.50	1240	260
16	296	119	5.70	5.40	-	-
17	222	146	-	-	-	-
18	199	197	5.70	5.45	-	-
19	214	156	5.65	5.30	190	195
20	-	123	5.55	5.10	-	-
21	283	121	5.60	5.15	-	-
22	224	143	5.65	5.20	156	260
23	252	121	5.80	5.30	-	-
24	243	119	5.65	5.15	-	-
25	231	139	-	-	-	-
26	237	154	5.60	5.15	-	-
27	233	146	-	-	-	-
28	241	168	-	-	-	-
29	239	172	5.60	5.10	1550	2810
30	244	139	5.55	5.15	-	-
31	215	162	5.55	5.00	-	-
32	250	-	5.40	4.95	-	-
33	228	-	5.45	5.00	1770	3260
34	230	-	5.50	5.00	-	-
35	230	140	-	-	-	-
36	225	110	5.30	4.90	1760	3380
37	176	114	5.30	4.95	1570	3560
38	-	130	5.45	4.90	-	-
39	219	158	5.45	5.00	-	-
40	233	164	5.50	5.00	-	-
41	285	168	5.50	5.00	1840	4130
42	306	149	5.50	5.00	1850	4590
43	269	149	5.50	5.00	2430	4010
44	207	103	5.50	5.00	1900	4600
45	248	124	-	-	-	-
46	261	153	-	-	-	-
47	277	162	5.60	5.10	-	-
48	-	-	5.55	5.05	1430	4030
49	377	195	5.50	5.00	1740	3310
50	203	166	-	-	1840	3230

COD, pH and MLSS Raw Data from Section 6.1.3 (cont.)

Day #	COD (mg/L)		pH		MLSS (mg/L)	
	Raw Inf.	Sett.Sewage	Ferm.1	Ferm.2	Ferm.1	Ferm.2
51	219	165	–	–	–	–
52	229	165	–	–	–	–
53	300	184	–	–	–	–
54	248	149	5.65	5.15	1200	2580
Mean	245	151	5.60	5.15	1358	2433

COD, pH and MLSS Raw Data from Section 6.1.4

Day #	COD (mg/L)		pH		MLSS (mg/L)	
	Raw Inf.	Sett.Sewage	Ferm.1	Ferm.2	Ferm.1	Ferm.2
1	360	179	6.05	5.90	–	–
2	411	208	5.85	5.70	–	–
3	370	216	5.85	5.65	–	–
4	366	–	5.85	5.65	1888	1588
5	160	156	5.85	5.65	–	–
6	146	117	5.75	5.65	–	–
7	135	115	5.70	5.60	–	–
8	168	129	5.70	5.60	2155	2400
9	175	146	5.70	5.60	–	–
10	222	173	6.30	6.20	–	–
11	239	181	6.40	6.30	2395	2015
12	339	204	5.75	5.65	–	–
13	376	189	5.70	5.60	–	–
14	323	184	5.90	5.80	–	–
15	303	199	5.70	5.60	2135	2453
16	313	226	–	–	–	–
17	330	177	5.90	5.95	–	–
18	301	187	5.90	5.70	–	–
19	368	182	5.90	5.80	1337	1485
20	–	–	–	–	–	–
21	–	–	–	–	–	–
22	325	199	5.85	5.75	–	–
23	362	195	5.60	5.50	880	1150
24	357	195	5.70	5.60	–	–
25	374	183	5.75	5.55	–	–
26	323	172	5.65	5.55	1685	1545
27	307	200	5.75	5.65	–	–
28	325	186	5.75	5.65	–	–
29	337	179	6.00	5.80	–	–
30	291	171	5.85	5.65	1255	1400
31	291	158	5.85	5.70	–	–
32	300	220	5.75	5.65	–	–
33	351	168	5.85	5.60	1205	1380
34	302	181	5.70	5.65	–	–
35	310	159	5.75	5.65	–	–
36	336	108	5.70	5.65	–	–
37	338	172	5.85	5.65	–	–
38	304	185	5.80	5.70	–	–
39	295	185	6.00	5.75	–	–

COD, pH and MLSS Raw Data from Section 6.1.4 (cont.)

Day #	COD (mg/L) Raw Inf.	Sett.Sewage	pH Ferm.1	Ferm.2	MLSS (mg/L) Ferm.1	Ferm.2
40	367	157	5.90	5.65	2085	1665
41	338	192	5.70	5.70	-	-
42	383	194	5.90	5.80	-	-
43	410	151	5.95	5.90	-	-
44	293	-	5.90	5.75	1795	-
45	291	174	5.90	5.70	-	-
46	307	192	6.15	5.90	-	-
47	263	148	5.95	5.70	1805	2353
48	328	163	5.80	5.90	-	-
49	368	-	5.80	5.90	-	-
50	463	127	6.05	6.00	-	-
51	501	169	5.95	5.95	1935	1545
52	516	184	5.90	5.85	-	-
53	-	-	-	-	-	-
54	401	144	5.90	5.80	-	-
55	397	222	5.95	5.90	-	-
56	351	157	5.85	5.80	-	-
57	551	145	5.90	5.85	-	-
58	-	169	-	-	-	-
59	293	165	5.85	5.80	2165	1915
60	348	185	6.00	5.95	-	-
61	-	149	5.90	5.90	-	-
62	-	159	-	-	1755	1875
63	320	-	-	-	-	-
64	348	172	-	-	-	-
65	340	208	6.10	6.00	-	-
66	389	190	6.00	5.95	-	-
67	-	190	6.00	6.00	-	-
68	332	196	5.90	5.90	-	-
69	366	200	-	-	-	-
70	-	-	-	-	-	-
71	-	233	6.30	6.20	-	-
72	383	218	6.00	5.95	-	-
73	311	210	6.10	6.00	-	-
74·	325	210	6.10	6.00	-	-
75	335	223	6.10	6.00	2234	2338
76	339	208	6.25	6.10	-	-
77	343	226	6.55	6.50	-	-
78	351	251	6.20	6.10	1686	1966
79	341	257	6.20	6.10	-	-
80	374	216	6.00	5.90	-	-
81	435	220	6.00	5.95	1422	1866
82	336	182	6.30	6.10	-	-
83	311	191	6.30	6.20	-	-
84	423	241	6.25	6.20	-	-
85	622	245	6.30	6.25	1928	2040
86	382	195	6.30	6.20	-	-
87	602	216	6.40	6.30	-	-
88	519	187	-	-	-	-
89	718	178	-	-	-	-

COD, pH and MLSS Raw Data from Section 6.1.4 (cont.)

Day #	COD (mg/L)		pH		MLSS (mg/L)	
	Raw Inf.	Sett.Sewage	Ferm.1	Ferm.2	Ferm.1	Ferm.2
90	627	224	6.40	6.30	–	–
91	643	216	–	–	–	–
92	369	207	6.40	6.35	2012	2198
93	444	199	6.30	6.20	–	–
94	336	203	6.25	6.15	–	–
95	353	220	6.25	6.15	1665	1710
96	301	168	6.25	6.15	–	–
97	501	174	–	–	–	–
98	431	174	6.40	6.30	–	–
99	354	198	6.35	6.25	–	–
100	366	202	6.25	6.25	1786	1906
101	401	229	6.30	6.25	–	–
102	328	158	–	–	–	–
103	332	158	6.50	6.40	–	–
104	291	150	6.70	6.65	–	–
105	279	158	6.70	6.60	–	–
106	411	174	–	–	2434	2286
107	380	157	6.75	6.70	–	–
108	440	172	6.65	6.55	–	–
109	331	176	6.55	6.50	2244	2378
110	339	184	6.50	6.40	–	–
111	525	200	6.50	6.35	–	–
112	362	215	6.55	6.45	–	–
113	331	231	6.50	6.40	–	–
114	351	234	6.40	6.30	2516	3054
115	332	198	6.40	6.25	–	–
116	339	210	6.30	6.20	–	–
117	465	217	6.40	6.20	2912	3054
118	367	205	6.30	6.10	–	–
119	381	220	6.15	5.95	–	–
120	444	224	6.25	5.95	2560	3704
121	374	203	6.35	6.20	–	–
122	386	199	6.35	6.10	–	–
123	443	232	6.25	6.10	–	–
124	415	242	6.25	6.10	3038	3152
125	343	244	6.20	6.05	–	–
126	380	234	6.20	6.10	–	–
127	528	230	6.15	6.10	–	–
128	333	211	6.15	6.05	3286	3534
129	296	185	6.15	6.05	–	–
130	296	248	6.20	6.00	–	–
131	325	195	6.20	6.05	3236	2896
132	420	191	6.20	5.95	–	–
133	329	210	6.10	5.95	–	–
134	420	222	6.10	5.95	–	–
135	319	181	–	–	–	–
136	332	181	6.15	5.90	3004	2692
137	282	151	6.10	5.90	–	–
138	290	193	6.10	5.90	2676	2898
139	351	218	6.20	6.00	–	–

COD, pH and MLSS Raw Data from Section 6.1.4 (cont.)

Day #	COD (mg/L) Raw Inf.	Sett.Sewage	pH Ferm.1	Ferm.2	MLSS (mg/L) Ferm.1	Ferm.2
140	347	197	6.15	5.95	–	–
141	376	224	6.15	6.00	–	–
142	324	212	6.30	6.10	2608	2918
143	320	201	6.15	6.05	–	–
144	346	197	6.20	6.10	–	–
145	309	180	6.20	6.10	2554	2602
146	284	172	6.10	5.95	–	–
147	258	148	6.10	5.90	–	–
148	252	176	6.05	5.95	2304	3040
149	349	145	6.05	6.00	–	–
150	243	137	6.05	6.05	–	–
151	259	147	–	–	–	–
152	280	157	6.10	6.10	–	–
153	306	145	6.10	6.05	2706	2778
154	337	186	6.15	6.15	–	–
155	300	165	6.15	6.05	–	–
156	308	186	6.10	6.05	3576	2922
157	321	147	6.10	6.05	–	–
158	–	112	6.15	6.15	–	–
159	323	167	6.20	6.25	3748	4202
160	275	131	–	–	–	–
161	285	159	6.05	6.10	–	–
162	327	169	6.00	6.05	–	–
163	291	167	6.05	6.10	3872	4334
164	339	185	6.10	6.10	–	–
165	283	181	6.10	6.15	–	–
166	299	187	6.15	6.15	3924	3626
167	267	179	6.15	6.25	–	–
168	263	158	6.05	6.10	–	–
169	325	193	–	–	–	–
Mean	351	187	6.10	6.00	2310	2432

VFA Raw Data from Section 6.1.1

Day #	Fermenter 1 (mg/L as HAc) Acetic	Propion.	Butyric	Total	Fermenter 2 (mg/L as HAc) Acetic	Propion.	Butyric	Total
1	30.3	18.8	<2.0	49.1	86.3	84.1	8.8	179.2
2	35.9	20.5	<2.0	56.4	41.3	31.0	3.5	75.8
3	16.2	8.0	<2.0	24.2	17.0	11.4	<2.0	28.4
4	15.7	7.3	<2.0	23.0	29.9	17.9	2.2	50.0
5	28.5	11.7	<2.0	40.2	45.6	26.5	2.5	74.6
6	26.1	16.2	<2.0	42.3	35.1	28.2	2.9	66.2
7	–	–	–	–	–	–	–	–
8	31.1	13.7	<2.0	44.8	43.9	32.0	2.9	78.8
9	25.5	14.5	2.3	42.3	44.5	32.3	<2.0	76.7
10	22.0	14.4	<2.0	36.5	49.6	39.5	2.4	91.4
11	38.0	22.2	<2.0	60.2	56.1	45.3	2.4	103.7
12	36.7	23.2	5.0	64.9	58.2	49.4	2.5	107.9
13	38.7	23.3	<2.0	62.0	50.6	41.3	4.4	96.3

VFA Raw Data from Section 6.1.1 (cont.)

Day #	Fermenter 1 (mg/L as HAc)				Fermenter 2 (mg/L as HAc)			
	Acetic	Propion.	Butyric	Total	Acetic	Propion.	Butyric	Total
14	39.8	23.5	<2.0	63.3	63.1	49.9	<2.0	113.0
15	62.6	38.2	4.9	105.7	115.0	86.3	5.4	206.7
16	49.3	25.7	<2.0	75.0	18.6	11.6	<2.0	30.2
17	-	-	-	-	34.2	21.1	<2.0	55.3
18	-	-	-	-	44.9	28.5	<2.0	73.4
19	-	-	-	-	27.5	16.9	<2.0	44.4
20	14.0	4.8	<2.0	18.8	64.9	40.7	2.5	108.1
21	-	-	-	-	40.7	26.7	3.4	70.8
22	22.7	10.6	<2.0	33.3	28.0	17.7	<2.0	45.7
23	19.6	8.2	<2.0	27.8	68.0	45.8	2.7	116.5
24	37.8	18.2	<2.0	56.0	48.8	32.6	<2.0	81.4
25	15.2	6.3	<2.0	21.5	27.0	17.1	<2.0	44.1
26	17.7	8.3	<2.0	26.0	-	-	-	-
27	20.0	9.2	<2.0	29.2	59.4	42.8	2.3	105.0
28	14.0	6.2	<2.0	20.2	40.5	27.9	<2.0	68.4
29	37.2	18.5	<2.0	55.7	47.0	34.3	<2.0	81.3
30	21.5	9.7	<2.0	31.2	-	-	-	-
31	14.2	7.6	<2.0	21.8	46.7	36.9	<2.0	83.6
32	39.6	21.5	<2.0	61.1	34.8	27.4	<2.0	62.2
33	29.9	14.5	<2.0	44.4	67.6	50.2	<2.0	117.8
34	26.9	12.9	<2.0	39.8	61.3	44.3	2.2	107.8
35	33.0	16.6	<2.0	49.6	62.1	44.6	2.3	109.0
36	29.4	14.1	<2.0	43.5	55.6	40.8	2.1	98.5
37	29.5	14.3	<2.0	46.3	60.7	43.5	2.6	106.8
38	23.1	11.9	<2.0	35.0	60.3	44.3	<2.0	104.7
39	23.6	13.3	<2.0	36.9	63.4	47.0	2.7	113.1
40	20.3	10.6	<2.0	30.9	65.7	47.7	3.0	116.4
41	32.1	16.9	2.5	51.5	63.1	45.1	3.0	111.2
42	15.8	10.4	<2.0	26.2	59.6	43.5	3.5	106.6
43	21.9	13.3	<2.0	35.2	51.2	38.7	2.2	92.1
44	22.7	15.9	<2.0	38.6	53.1	41.1	2.6	96.8
Mean	27.6	14.7	0.5	42.8	51.0	37.4	1.8	90.2

VFA Raw Data from Section 6.1.2

Day #	Fermenter 1 (mg/L as HAc)				Fermenter 2 (mg/L as HAc)			
	Acetic	Propion.	Butyric	Total	Acetic	Propion.	Butyric	Total
1	31.7	22.1	7.9	61.7	14.1	10.7	2.0	26.7
2	17.2	13.7	<2.0	30.9	19.9	20.3	2.3	42.5
3	32.5	31.8	<2.0	64.3	79.7	68.9	3.7	152.3
4	33.7	23.1	<2.0	56.8	30.7	27.3	<2.0	59.8
5	37.8	33.6	<2.0	71.4	51.9	43.3	<2.0	95.3
6	65.1	55.9	3.2	124.2	54.0	47.2	<2.0	102.3
7	10.8	11.8	<2.0	22.6	20.2	26.8	<2.0	47.0
8	3.8	9.4	<2.0	13.2	27.9	31.9	2.2	61.9
9	21.6	23.8	2.8	48.1	25.5	26.9	<2.0	52.4
10	53.2	44.8	2.6	100.6	36.3	38.3	2.1	76.7
11	25.4	27.2	6.0	58.6	48.4	43.0	3.6	97.0
12	72.7	36.5	<2.0	109.2				

VFA Raw Data from Section 6.1.2 (cont.)

Day #	Fermenter 1 (mg/L as HAc)				Fermenter 2 (mg/L as HAc)			
	Acetic	Propion.	Butyric	Total	Acetic	Propion.	Butyric	Total
13	66.6	39.4	4.4	110.7	73.3	71.6	4.5	149.4
14	61.9	36.8	<2.0	99.4	97.1	71.3	2.3	170.7
15	79.6	35.4	3.5	118.4	66.5	63.6	3.2	133.3
16	73.1	50.3	<2.0	123.4	121.0	86.3	<2.0	207.3
17	71.7	46.1	<2.0	117.8	105.4	75.3	<2.0	180.7
18	61.6	48.3	<2.0	109.9	97.6	72.5	<2.0	170.2
19	73.4	57.7	<2.0	131.1	115.1	90.0	<2.0	205.1
20	65.9	54.3	<2.0	120.2	114.5	94.8	<2.0	209.3
21	62.7	51.5	<2.0	114.2	117.2	99.6	<2.0	216.8
22	71.9	50.6	<2.0	122.5	129.1	101.0	<2.0	230.1
23	60.3	47.7	<2.0	108.0	-	-	-	-
24	62.5	48.9	<2.0	111.4	110.4	96.6	<2.0	207.0
25	62.5	56.4	<2.0	118.9	118.5	98.7	<2.0	217.2
26	91.5	34.0	<2.0	125.6	159.8	80.1	<2.0	239.9
27	43.2	25.0	<2.0	68.2	160.2	63.7	<2.0	223.9
28	159.3	54.1	<2.0	213.4	192.2	94.3	<2.0	286.5
29	68.8	51.8	<2.0	120.6	111.3	88.2	<2.0	199.5
30	59.6	43.6	<2.0	103.2	129.2	86.4	<2.0	215.6
31	63.6	46.2	<2.0	109.8	108.7	86.3	<2.0	195.0
32	79.3	54.2	<2.0	133.5	132.8	105.7	<2.0	238.5
33	-	-	-	-	-	-	-	-
34	65.1	51.7	<2.0	116.8	97.8	88.2	<2.0	186.0
35	69.5	47.0	<2.0	116.5	97.0	84.1	<2.0	181.0
36	78.0	41.2	<2.0	119.2	94.0	72.5	10.8	177.3
37	76.2	50.4	<2.0	126.6	95.8	82.5	<2.0	178.3
38	79.3	52.7	<2.0	132.0	97.1	81.5	<2.0	178.6
39	81.8	45.5	<2.0	127.3	101.2	85.1	<2.0	186.3
40	94.5	89.3	<2.0	183.9	93.0	78.8	<2.0	173.3
41	64.0	58.0	<2.0	122.0	72.0	59.0	<2.0	131.0
42	-	-	-	-	-	-	-	-
43	58.0	45.0	<2.0	103.0	100.0	84.0	6.0	190.0
44	73.0	59.0	<2.0	132.0	94.0	57.0	<2.0	151.0
45	63.0	51.0	<2.0	114.0	77.0	57.0	<2.0	132.0
46	63.0	56.0	<2.0	126.0	96.0	80.0	<2.0	182.0
47	64.0	54.0	<2.0	108.0	100.0	85.0	6.0	191.0
48	70.0	54.0	<2.0	124.0	100.0	85.0	7.0	192.0
Mean	61.8	43.9	0.7	106.4	90.9	70.1	1.5	162.5

VFA Raw Data from Section 6.1.3

Day #	Fermenter 1 (mg/L as HAc)				Fermenter 2 (mg/L as HAc)			
	Acetic	Propion.	Butyric	Total	Acetic	Propion.	Butyric	Total
1	59.9	35.9	<2.0	95.8	61.9	42.6	<2.0	104.5
2	57.5	32.9	<2.0	90.4	65.8	44.0	<2.0	109.8
3	58.4	34.9	<2.0	93.3	66.1	44.9	<2.0	111.0
4	33.6	11.7	<2.0	45.3	43.2	18.1	<2.0	61.3
5	39.4	16.9	<2.0	56.3	44.0	19.9	<2.0	63.9
6	53.3	26.7	<2.0	80.0	55.7	31.1	<2.0	86.6
7	41.9	29.4	<2.0	71.3	33.2	23.3	<2.0	56.5

VFA Raw Data from Section 6.1.3 (cont.)

Day #	Fermenter 1 (mg/L as HAc)				Fermenter 2 (mg/L as HAc)			
	Acetic	Propion.	Butyric	Total	Acetic	Propion.	Butyric	Total
8	43.5	32.6	<2.0	76.1	40.3	30.2	<2.0	70.5
9	64.4	49.4	<2.0	113.0	61.8	47.4	<2.0	109.2
10	39.4	33.1	<2.0	72.5	67.1	57.8	<2.0	124.9
11	35.1	32.5	<2.0	67.6	75.5	60.3	<2.0	136.0
12	60.5	46.8	<2.0	107.0	69.8	55.6	<2.0	125.4
13	82.1	62.7	<2.0	149.0	79.4	63.0	<2.0	142.5
14	54.5	41.1	<2.0	95.6	43.4	36.8	<2.0	80.2
15	47.0	34.1	<2.0	81.3	87.0	67.0	3.8	157.8
16	64.5	46.8	<2.0	111.0	113.8	90.2	4.7	208.7
17	-	-	-	-	-	-	-	-
18	70.4	49.7	<2.0	120.1	99.2	76.6	3.5	179.3
19	71.3	51.9	<2.0	123.2	124.5	99.1	6.3	229.9
20	66.1	47.5	4.5	118.1	66.3	50.3	6.0	122.6
21	68.5	49.3	3.7	121.5	80.1	60.4	<2.0	140.5
22	45.7	29.6	6.5	81.8	61.0	42.5	<2.0	103.5
23	69.6	51.0	4.7	125.3	76.5	58.1	7.1	141.7
24	53.5	36.8	2.8	93.1	64.9	49.8	3.3	118.0
25	78.3	57.7	5.4	141.4	109.0	85.0	6.7	200.7
26	66.2	49.0	8.5	123.7	86.8	66.6	11.1	164.5
27	76.0	56.8	5.0	138.0	92.5	72.8	6.0	171.3
28	-	-	-	-	125.0	97.3	7.2	229.5
29	-	-	-	-	105.0	82.1	11.3	198.4
30	-	-	-	-	111.0	86.5	11.8	209.3
31	-	-	-	-	92.4	72.6	6.1	171.1
32	-	-	-	-	68.9	53.2	3.8	125.9
33	87.7	66.3	7.4	161.4	73.2	57.8	7.0	138.0
34	-	-	-	-	121.0	92.9	12.5	226.4
35	86.6	64.0	6.8	157.4	133.0	102.0	15.5	250.5
36	47.7	38.2	8.5	94.4	70.0	69.2	11.6	150.8
37	43.1	28.5	5.8	77.4	73.0	66.0	8.2	147.2
38	75.6	54.6	<2.0	130.2	87.9	82.8	9.4	180.0
39	43.7	55.1	<2.0	98.9	56.6	71.7	8.6	136.9
40	58.6	44.4	<2.0	103.0	98.4	89.3	9.2	196.9
41	53.5	37.6	<2.0	91.1	122.0	111.0	5.7	238.8
42	82.2	67.4	4.2	153.8	89.3	82.1	8.5	179.8
43	69.9	56.9	6.5	133.3	77.7	73.4	8.2	159.3
44	59.5	62.2	6.7	128.4	75.4	77.1	7.4	159.9
45	59.9	46.9	2.4	109.2	120.0	112.0	9.4	241.4
46	71.5	48.9	2.8	123.2	84.5	78.6	6.2	169.3
47	71.7	53.6	3.9	129.2	102.8	82.9	7.9	193.6
48	57.1	43.7	4.0	104.8	94.8	82.9	8.3	186.0
49	54.3	40.8	10.6	105.7	110.0	92.0	7.7	209.7
50	57.5	43.6	4.1	105.2	123.1	106.2	15.9	245.2
51	81.5	62.8	5.1	149.4	119.1	99.9	6.3	225.3
52	98.5	69.3	6.2	174.0	137.3	116.3	8.0	262.6
53	29.2	20.2	1.5	50.9	79.3	64.7	4.4	148.4
54	-	-	-	-	-	-	-	-
Mean	60.7	44.6	2.8	108.1	85.0	69.2	5.5	159.7

VFA Raw Data from Section 6.1.4

	Fermenter 1 (mg/L as HAc)				Fermentor 2 (mg/L as HAc)				Clar.
Day#	Acetic	Propion	Butyric	Total	Acetic	Propion	Butyric	Total	Total
1	60.3	44.7	<2.0	105.0	79.0	67.6	6.0	152.6	164.4
2	76.4	59.0	<2.0	135.4	80.2	63.4	6.0	149.6	132.6
3	85.9	68.1	5.3	159.3	85.9	72.4	5.3	163.6	179.1
4	79.4	68.1	4.8	152.3	84.5	73.0	5.3	162.8	189.8
5	81.1	70.4	3.9	155.4	92.0	80.9	5.3	178.2	175.4
6	82.3	71.4	5.3	196.7	101.9	89.5	5.3	196.7	198.6
7	82.1	70.5	5.3	171.9	104.4	98.4	5.3	195.1	177.4
8	87.9	73.2	5.3	166.4	96.2	88.9	5.3	190.4	207.0
9	100.3	85.3	<2.0	185.6	107.0	97.5	<2.0	194.5	226.0
10	109.0	92.9	<2.0	201.9	119.2	105.2	<2.0	224.4	222.4
11	92.0	77.2	8.2	177.4	73.9	65.0	6.1	145.0	188.3
12	91.3	75.2	8.2	174.7	100.7	89.6	8.2	198.5	210.2
13	94.0	78.1	8.2	180.3	106.0	89.0	8.2	203.2	227.0
14	92.7	75.1	8.2	176.0	93.8	77.3	8.2	179.3	197.3
15	88.1	72.2	8.2	168.5	95.8	83.7	8.2	187.7	219.0
16	-	-	-	-	-	-	-	-	-
17	75.0	57.9	5.2	138.1	76.1	72.4	5.2	153.7	177.3
18	63.3	46.1	5.2	114.6	76.9	66.8	5.2	148.9	135.4
19	60.3	42.5	5.2	108.0	71.8	57.0	5.2	134.0	169.2
20	-	-	-	-	-	-	-	-	-
21	-	-	-	-	-	-	-	-	-
22	46.4	29.8	5.2	81.4	73.2	58.3	5.2	136.7	184.7
23	65.2	48.9	4.0	118.1	77.8	63.9	6.0	147.7	159.0
24	77.2	59.4	4.0	140.6	80.4	67.5	5.0	152.9	164.7
25	64.3	50.9	2.0	117.2	76.5	70.9	4.0	151.4	160.9
26	67.0	55.9	4.0	127.3	88.9	76.0	7.5	172.4	215.9
27	66.9	54.1	4.0	125.0	85.4	76.3	6.0	167.7	255.9
28	67.3	51.8	4.0	123.1	84.0	75.4	4.0	163.4	197.7
29	55.2	40.5	4.0	99.7	79.5	67.1	6.0	152.6	-
30	54.4	39.2	4.0	97.6	59.9	51.5	4.0	115.4	175.5
31	53.5	40.5	4.0	98.0	64.5	56.8	4.0	125.3	158.4
32	56.1	43.2	4.0	101.3	66.8	57.1	3.0	126.9	-
33	53.8	41.2	2.0	97.0	67.7	54.2	4.0	125.9	154.8
34	69.1	49.1	4.0	122.2	81.5	62.0	6.0	149.5	154.4
35	65.6	42.5	2.0	110.1	85.0	62.3	5.0	152.3	157.7
36	61.8	42.5	2.0	106.3	67.0	56.3	2.0	125.3	145.5
37	46.5	36.0	2.0	84.5	73.2	57.1	5.0	135.3	166.3
38	67.2	51.3	3.0	121.5	67.8	56.4	3.0	127.2	135.2
39	62.0	49.1	3.0	114.1	72.2	63.8	3.0	139.0	145.1
40	62.0	52.5	3.0	117.5	75.9	68.8	3.0	147.7	153.2
41	73.5	61.0	4.0	138.5	93.0	83.1	5.0	181.1	176.4
42	68.5	52.0	2.0	122.5	85.3	77.0	4.5	166.8	170.6
43	87.8	72.5	2.0	162.3	87.8	73.3	2.0	163.1	128.9
44	76.0	64.1	2.0	142.1	89.0	78.9	2.0	169.9	170.9
45	69.8	53.2	2.0	125.0	96.2	91.2	4.5	191.9	191.3
46	77.6	57.0	2.0	136.6	96.5	78.5	2.0	177.0	188.6
47	79.5	54.8	2.0	136.3	102.1	85.0	4.5	191.6	201.7
48	86.0	76.2	6.0	168.2	90.2	89.5	6.0	185.7	196.5
49	84.9	70.0	6.0	160.9	87.6	83.8	6.0	177.0	195.6
50	83.6	65.3	5.0	153.9	82.9	74.0	6.0	162.9	177.8

VFA Raw Data from Section 6.1.4 (cont.)

Day#	Fermenter 1 (mg/L as HAc)				Fermenter 2 (mg/L as HAc)				Clar.
	Acetic	Propion	Butyric	Total	Acetic	Propion	Butyric	Total	Total
51	94.8	74.3	6.0	175.1	112.0	92.6	6.0	210.6	192.1
52	93.9	76.1	6.0	176.0	106.0	96.8	7.5	210.3	221.5
53	-	-	-	-	-	-	-	-	-
54	110.0	95.2	6.0	211.2	124.6	115.4	6.0	246.0	238.4
55	113.8	100.0	6.0	219.8	126.2	115.6	9.0	250.8	264.8
56	114.0	83.3	4.0	201.3	121.4	96.8	6.0	224.2	235.2
57	107.0	82.5	4.0	193.5	110.6	91.0	6.0	207.6	226.8
58	-	-	-	-	-	-	-	-	-
59	98.5	70.6	5.5	174.6	115.2	90.0	5.5	210.7	203.8
60	94.4	72.0	3.5	169.9	102.0	90.0	5.5	197.5	191.5
61	85.8	63.4	3.5	152.7	115.2	93.2	5.5	213.9	222.2
62	-	-	-	-	-	-	-	-	-
63	-	-	-	-	-	-	-	-	-
64	-	-	-	-	-	-	-	-	-
65	86.5	61.5	5.5	155.3	102.8	76.0	5.5	184.3	182.7
66	80.6	55.1	3.5	139.2	104.2	80.6	5.5	190.7	189.4
67	79.3	57.1	3.5	139.9	93.8	68.3	5.5	167.6	168.7
68	61.4	45.8	4.0	111.2	63.1	51.0	4.0	118.1	205.9
69	-	-	-	-	-	-	-	-	-
70	-	-	-	-	-	-	-	-	-
71	75.4	51.9	5.5	132.8	100.0	83.7	8.5	192.5	181.1
72	90.0	66.3	4.0	160.3	95.9	76.7	6.0	178.9	160.7
73	79.1	62.3	6.0	147.4	88.4	78.5	6.0	172.9	173.2
74	70.5	53.3	4.0	127.8	90.5	83.3	6.0	179.8	189.9
75	74.6	52.8	3.5	130.9	80.9	66.0	3.5	150.4	154.5
76	60.5	42.2	2.0	104.7	71.2	61.3	3.5	136.0	147.5
77	59.1	41.5	2.0	104.1	76.4	69.4	3.5	149.3	156.9
78	57.1	40.3	2.0	99.4	78.0	66.0	3.5	147.5	180.2
79	59.3	43.4	3.0	105.7	65.9	57.5	3.0	126.4	136.2
80	61.5	48.9	3.0	113.4	78.2	61.5	5.0	144.7	135.5
81	53.0	41.1	2.0	96.1	60.1	53.3	4.0	117.4	130.4
82	51.4	36.0	2.0	89.4	68.1	51.8	4.0	123.9	125.1
83	59.3	36.0	2.0	97.3	65.9	46.3	4.0	116.2	119.8
84	52.2	28.5	2.0	82.5	64.0	42.1	2.0	108.1	119.2
85	52.8	26.3	2.0	81.1	56.0	36.0	2.0	94.0	113.2
86	50.2	25.2	2.0	77.4	61.0	38.4	2.0	101.4	106.7
87	63.5	29.1	2.0	94.6	68.2	40.5	2.0	112.7	121.5
88	-	-	-	-	-	-	-	-	-
89	50.3	11.1	2.0	63.4	70.2	35.3	2.0	107.5	132.2
90	63.6	22.9	2.0	88.5	67.8	28.5	2.0	98.3	101.0
91	58.7	25.3	2.0	86.0	70.2	34.8	2.0	107.0	112.6
92	59.3	29.6	2.0	90.4	69.6	38.1	2.0	109.7	111.3
93	51.5	30.3	2.0	83.8	62.2	36.2	2.0	100.4	109.4
94	65.0	33.3	2.0	100.3	63.3	37.2	2.0	102.5	111.8
95	57.5	32.9	2.0	92.4	61.2	38.8	2.0	102.0	101.3
96	55.2	30.4	2.0	87.6	57.6	35.9	2.0	95.8	95.8
97	46.9	24.3	2.0	73.2	64.3	37.9	2.0	104.2	105.2
98	40.3	20.0	2.0	62.3	51.5	32.0	2.0	85.5	110.6
99	44.1	23.2	2.0	69.3	56.0	32.0	2.0	90.0	95.7
100	54.2	28.9	2.0	85.1	57.9	29.9	2.0	89.8	87.9

VFA Raw Data from Section 6.1.4 (cont.)

Day#	Fermenter 1 (mg/L as HAc)				Fermenter 2 (mg/L as HAc)				Clar.
	Acetic	Propion	Butyric	Total	Acetic	Propion	Butyric	Total	Total
101	63.6	28.3	4.0	95.9	65.0	31.3	3.0	99.3	90.2
102	-	-	-	-	-	-	-	-	-
103	38.4	15.4	2.0	55.8	46.1	20.9	3.0	70.0	75.7
104	33.1	12.4	2.0	44.5	43.2	17.9	3.0	64.1	72.1
105	39.7	6.9	2.0	48.6	40.8	11.8	2.0	54.6	60.4
106	-	-	-	-	-	-	-	-	-
107	33.7	7.9	3.0	44.6	35.4	9.9	3.0	48.3	53.8
108	35.1	11.1	<2.0	46.2	37.3	13.7	<2.0	51.0	43.8
109	33.6	11.9	<2.0	45.5	39.2	14.1	<2.0	53.3	57.6
110	30.9	11.2	2.0	44.2	35.1	12.0	2.0	49.1	49.4
111	28.6	10.9	2.0	41.5	34.7	14.8	2.0	51.5	52.7
112	22.4	13.0	2.0	37.4	36.2	16.4	2.0	54.6	55.7
113	35.1	13.7	2.0	50.8	41.1	16.4	2.0	59.5	60.3
114	47.2	23.2	3.0	73.4	43.4	23.2	3.0	69.6	74.5
115	45.9	23.8	3.0	72.7	50.7	27.7	3.0	81.4	81.2
116	49.3	28.2	3.0	80.5	53.5	32.5	3.0	89.0	92.3
117	45.2	25.0	3.0	73.2	59.0	38.1	3.0	100.1	101.6
118	41.9	25.0	2.0	68.9	67.3	41.2	3.0	111.5	106.1
119	62.6	40.0	3.0	105.6	97.0	66.3	4.5	167.8	152.2
120	67.2	39.1	3.0	109.3	79.0	53.3	4.5	136.8	138.4
121	65.2	41.3	3.0	109.5	74.1	45.9	3.0	123.0	120.0
122	64.0	35.9	3.0	102.9	74.2	50.0	4.5	128.7	136.4
123	68.9	43.5	2.5	114.9	74.3	51.0	3.5	128.8	-
124	68.2	45.3	2.5	116.0	73.1	50.4	3.5	127.0	132.0
125	61.1	40.6	2.5	104.2	76.4	54.0	3.5	133.9	145.4
126	72.3	51.2	2.5	126.0	67.1	44.3	3.5	114.9	154.2
127	65.5	44.3	2.5	112.3	71.6	49.5	3.5	124.6	140.0
128	66.0	45.8	2.5	114.3	74.1	54.3	3.5	131.9	138.2
129	73.3	50.9	2.0	126.2	83.0	58.0	2.0	143.0	151.0
130	66.5	44.9	2.0	113.4	79.4	59.8	2.0	141.2	146.9
131	72.4	49.4	2.0	123.8	81.8	57.5	2.0	141.3	148.7
132	66.0	42.3	2.0	110.3	82.7	58.8	4.5	146.0	153.9
133	72.4	50.6	2.0	125.0	76.1	57.0	2.0	135.1	145.9
134	73.3	48.0	2.0	123.3	81.6	58.3	2.0	143.4	150.1
135	74.1	43.1	2.0	119.2	78.8	52.9	3.5	135.2	124.3
136	76.3	45.6	3.5	125.4	95.0	66.3	5.5	166.8	168.8
137	71.0	45.6	2.0	118.6	84.0	65.0	5.5	154.5	158.8
138	80.3	52.0	3.5	135.8	86.1	64.0	5.5	155.6	167.8
139	75.0	45.2	3.5	123.7	86.1	65.3	3.5	154.8	184.7
140	68.4	44.8	2.0	115.2	83.4	63.1	5.5	151.9	174.1
141	79.1	51.0	2.0	132.1	88.9	59.4	3.5	151.8	157.8
142	60.6	39.0	2.0	102.6	72.0	52.0	3.0	127.0	133.7
143	57.2	40.8	2.0	100.0	70.4	48.9	3.0	122.3	119.8
144	56.1	41.0	3.0	100.1	62.2	49.6	3.0	114.8	118.6
145	54.0	39.2	2.0	95.2	66.8	53.5	3.0	123.3	128.7
146	58.5	48.8	3.0	110.3	69.0	55.0	3.0	127.0	126.0
147	62.2	44.6	3.0	109.8	70.4	62.6	3.0	136.0	153.8
148	63.9	49.6	2.0	115.5	71.2	57.2	3.0	131.4	129.2
149	67.5	50.9	2.0	120.4	74.7	55.7	3.0	133.4	135.1
150	69.0	48.9	3.0	120.9	74.7	54.4	3.0	132.1	142.1

VFA Raw Data from Section 6.1.4 (cont.)

Day#	Fermenter 1 (mg/L as HAc)				Fermenter 2 (mg/L as HAc)				Clar.
	Acetic	Propion	Butyric	Total	Acetic	Propion	Butyric	Total	Total
151	63.0	45.0	2.0	110.0	70.4	50.3	3.0	123.7	128.4
152	68.9	47.2	3.0	119.1	70.4	52.0	2.0	124.4	138.2
153	76.0	48.3	4.0	128.3	91.0	62.4	4.0	157.4	177.9
154	78.2	50.1	2.0	130.3	85.0	55.9	4.0	144.9	152.1
155	77.9	47.0	2.0	126.9	83.3	55.9	4.0	143.1	158.8
156	71.4	44.5	2.0	117.9	79.7	49.3	2.0	131.0	143.3
157	79.7	41.9	4.0	125.6	91.0	53.3	4.0	148.3	155.1
158	78.7	43.8	4.0	126.5	72.2	41.9	2.0	116.1	121.0
159	77.9	42.3	2.0	122.2	82.4	43.6	2.0	128.0	123.6
160	83.4	47.0	2.0	132.4	85.3	47.8	2.0	135.1	135.1
161	83.3	48.3	2.0	133.6	101.3	58.3	2.0	161.5	173.9
162	93.0	56.8	2.0	151.8	96.8	57.6	2.0	156.4	166.7
163	89.1	48.0	2.0	139.1	95.0	57.0	2.0	154.0	161.2
164	87.1	50.3	2.0	139.4	86.3	51.1	2.0	139.4	150.6
165	85.0	51.1	2.0	138.1	97.0	54.2	2.0	153.2	159.0
166	93.0	49.9	2.0	144.4	94.3	53.2	2.0	149.5	157.2
167	101.3	60.1	2.0	163.4	112.0	62.1	2.0	176.1	165.0
168	117.5	75.2	2.0	194.5	120.0	74.8	2.0	196.8	205.8
169	97.8	58.7	2.0	158.5	98.2	57.9	2.0	158.1	178.5
Mean	69.0	47.3	3.4	119.7	79.0	59.1	4.2	141.7	157.0

COD, pH and MLSS Raw Data from Section 6.2

Day #	COD (mg/L)		pH		MLSS (mg/L)	
	Raw Inf.	Sett.Sewage	Ferm.1	Ferm.2	Ferm.1	Ferm.2
1	203	120	5.85	5.80	2310	2437
2	335	153	-	-	-	-
3	266	159	5.85	5.80	-	-
4	281	153	6.00	5.85	1900	3539
5	349	157	6.00	5.90	-	-
6	298	172	5.85	5.75	-	-
7	252	153	5.80	5.70	3136	2830
8	291	155	5.90	5.80	-	-
9	244	151	5.80	5.60	-	-
10	299	173	5.85	5.60	-	-
11	272	196	5.85	5.65	3070	3210
12	256	179	5.70	5.50	-	-
13	209	124	5.50	5.35	-	-
14	179	121	5.50	5.50	3407	3000
15	175	139	5.70	5.50	-	-
16	185	131	5.70	5.60	-	-
17	229	161	5.75	5.65	-	-
18	388	169	5.85	5.70	2680	3870
19	227	133	-	-	-	-
20	277	157	-	-	-	-
21	324	189	6.30	5.80	2575	5611
22	340	189	6.35	5.90	-	-
23	293	201	6.40	5.95	-	-

COD, pH and MLSS Raw Data from Section 6.2 (cont.)

Day #	COD (mg/L)		pH		MLSS (mg/L)	
	Raw Inf.	Sett.Sewage	Ferm.1	Ferm.2	Ferm.1	Ferm.2
24	389	159	6.35	5.90	–	–
25	347	171	6.20	5.85	–	–
26	341	161	6.20	5.75	–	–
27	379	183	6.25	5.75	–	–
28	291	171	6.25	5.70	2600	7538
29	–	–	–	–	–	–
30	–	–	–	–	–	–
31	–	–	5.60	5.50	–	–
32	271	175	–	–	–	–
33	287	163	5.60	5.45	5413	6350
34	–	–	5.70	5.45	–	–
35	318	122	5.65	5.40		
36	234	128	5.60	5.40	5161	4250
37	244	148	5.65	5.45	–	–
38	244	144	5.70	5.50	–	–
39	248	144	5.20	5.10	3593	3400
40	186	118	5.75	5.60	–	–
41	199	107	5.80	5.60	–	–
42	190	98	5.90	5.65	3325	2672
43	184	109	5.90	5.65	–	–
44	201	93	5.95	5.75	–	–
45	199	107	6.00	5.85	–	–
46	197	116	5.65	5.45	2253	2809
47	211	97	6.00	5.75	–	–
48	211	100	6.00	5.90	–	–
49	211	104	5.60	5.45	4161	2659
50	197	95	6.05	5.90	–	–
51	199	88	6.10	5.90	–	–
52	247	–	6.05	5.80	–	–
53	249	139	6.10	5.85	2260	2790
54	247	146	6.00	5.80	–	–
55	323	142	5.90	5.80	–	–
56	290	137	5.40	5.30	2643	2361
57	243	172	5.90	5.60	–	–
58	361	144	5.85	5.65	–	–
59	304	–	–	–	–	–
60	296	211	5.95	5.65	3228	3019
61	384	197	5.90	5.60	–	–
62	250	216	5.85	5.55	3277	3296
Mean	265	147	5.80	5.65	3166	3647

VFA Raw Data from Section 6.2

Day #	Fermenter 1 (mg HAc/L)				Fermenter 2 (mg HAc/L)			
	Acetic	Propion.	Butyric	Total	Acetic	Propion.	Butyric	Total
1	44.7	35.9	<2.0	80.6	69.1	59.0	3.6	131.7
2	46.9	35.5	2.2	84.6	47.8	42.0	2.6	92.4
3	39.2	28.3	<2.0	67.5	65.7	55.2	3.3	124.2
4	45.5	31.5	<2.0	77.0	66.9	53.0	3.0	122.9
5	48.3	13.3	<2.0	61.6	80.0	69.0	<2.0	149.0
6	80.0	72.5	<2.0	152.5	85.0	82.5	<2.0	167.5
7	47.8	26.5	<2.0	74.3	-	-	-	-
8	49.2	26.7	<2.0	75.9	63.4	37.1	<2.0	100.5
9	37.6	17.5	<2.0	55.1	82.2	39.0	3.1	124.3
10	-	-	-	-	57.3	44.4	<2.0	101.7
11	-	-	-	-	43.4	25.8	<2.0	69.2
12	70.1	28.9	<2.0	99.0	85.2	49.8	4.1	139.1
13	88.0	20.2	<2.0	108.2	-	-	-	-
14	42.2	30.6	<2.0	72.8	86.7	63.9	<2.0	150.6
15	43.7	25.6	<2.0	69.3	91.4	59.5	<2.0	150.9
16	80.8	50.8	2.1	133.7	101.0	56.1	<2.0	157.1
17	48.0	18.3	3.1	69.4	70.5	67.1	<2.0	137.6
18	36.7	16.2	<2.0	52.9	51.8	48.2	2.4	102.4
19	22.5	20.0	<2.0	42.5	73.1	55.1	<2.0	128.2
20	-	-	-	-	-	-	-	-
21	19.2	8.7	<2.0	27.9	40.0	35.0	<2.0	75.0
22	17.8	8.6	<2.0	26.4	77.2	59.4	<2.0	136.6
23	22.4	14.0	<2.0	36.4	82.0	55.5	<2.0	137.5
24	22.0	10.6	<2.0	32.6	81.2	63.0	<2.0	144.2
25	29.5	23.0	<2.0	52.5	81.1	64.9	<2.0	146.0
26	32.1	16.5	<2.0	48.6	62.0	50.0	<2.0	112.0
27	32.0	21.2	<2.0	53.2	-	-	-	-
28	54.8	42.4	<2.0	97.2	62.5	57.5	<2.0	120.0
29	-	-	-	-	-	-	-	-
30	-	-	-	-	-	-	-	-
31	-	-	-	-	-	-	-	-
32	47.1	52.3	<2.0	99.4	78.8	64.0	<2.0	142.8
33	83.7	80.0	<2.0	163.7	90.0	91.2	<2.0	181.2
34	70.0	66.3	<2.0	136.3	86.0	87.0	3.0	176.0
35	50.0	47.5	<2.0	97.5	98.0	91.0	16.0	205.0
36	65.0	65.0	<2.0	130.0	85.0	89.0	10.0	184.0
37	80.0	75.0	5.0	160.0	83.0	85.0	<2.0	168.0
38	65.0	54.0	5.0	124.0	85.0	76.0	7.0	168.0
39	68.0	61.0	5.0	134.0	96.0	93.0	<2.0	189.0
40	83.1	43.4	2.5	129.0	78.1	72.0	<2.0	150.1
41	68.5	50.4	<2.0	118.9	71.5	61.4	<2.0	132.9
42	52.8	28.1	<2.0	80.9	70.0	60.0	<2.0	130.0
43	59.7	44.0	<2.0	103.7	87.5	75.5	<2.0	163.0
44	56.3	40.1	<2.0	96.4	90.0	70.0	<2.0	160.0
45	57.7	29.0	<2.0	86.7	59.0	47.0	<2.0	106.0
46	49.3	20.1	<2.0	69.4	78.1	62.4	<2.0	140.5
47	63.7	25.2	6.2	95.1	85.8	59.9	<2.0	145.7
48	43.7	28.1	<2.0	71.8	58.2	40.6	<2.0	98.8
49	64.0	33.5	<2.0	97.5	87.3	46.7	<2.0	134.0
50	34.0	19.6	<2.0	53.6	104.5	65.7	2.7	172.9

VFA Raw Data from Section 6.2 (cont.)

Day #	Fermenter 1 (mg HAC/L)				Fermenter 2 (mg HAc/L)			
	Acetic	Propion.	Butyric	Total	Acetic	Propion.	Butyric	Total
51	56.6	19.3	<2.0	75.9	80.6	62.2	3.2	146.0
52	34.9	20.9	<2.0	55.8	101.8	63.3	<2.0	165.1
53	66.4	27.9	<2.0	94.3	80.2	55.9	<2.0	136.1
54	47.7	21.1	2.0	70.8	89.6	71.7	<2.0	161.3
55	75.9	38.1	<2.0	114.0	91.1	53.6	<2.0	144.7
56	66.1	50.4	2.5	119.0	104.1	83.2	<2.0	187.3
57	64.4	43.9	<2.0	108.3	77.0	47.4	<2.0	124.4
58	-	-	-	-	69.9	34.3	<2.0	104.2
59	-	-	-	-	-	-	-	-
60	47.8	33.7	<2.0	81.5	78.8	57.4	<2.0	136.2
61	58.4	45.8	5.2	109.4	-	-	-	-
62	-	-	-	-	71.2	61.1	<2.0	132.3
Mean	52.5	34.1	0.7	87.3	77.8	60.7	1.2	139.7

COD, pH and MLSS Raw Data from Section 6.3

Day #	COD (mg/L)		pH		MLSS (mg/L)	
	Raw Influent	Settled Sewage	Ferm.1 Low/High	Ferm.2 Low/High	Ferm.1	Ferm.2
1	254	156	6.40/6.80	6.45/6.90	2050	2190
2	234	143	6.55/7.20	6.65/7.40	-	-
3	-	114	6.30/7.20	6.70/7.00	-	-
4	302	167	6.85/6.85	7.05/7.05	-	-
5	306	163	6.35/7.55	6.45/7.50	2170	2810
6	200	122	6.60/7.00	6.85/7.15	-	-
7	212	118	6.45/7.15	6.60/7.30	2510	2500
8	392	143	6.70/6.80	6.80/6.90	-	-
9	285	143	6.60/ -	6.70/ -	-	-
10	249	155	- / -	- / -	-	-
11	265	186	6.30/7.00	6.25/6.85	1800	2650
12	249	180	6.55/6.80	6.65/6.85	-	-
13	261	167	6.50/7.00	6.65/6.90	-	-
14	276	176	6.60/7.50	6.60/7.15	2070	1890
15	270	-	6.45/6.90	6.55/6.80	-	-
16	277	147	6.45/7.30	6.35/6.90	-	-
17	302	180	6.90/ -	6.85/ -	-	-
18	270	182	6.90/7.20	6.90/7.15	1250	1590
19	306	194	6.75/6.90	6.80/7.00	-	-
20	270	188	6.60/ -	6.70/ -	-	-
21	318	176	6.70/6.90	6.80/7.00	1160	1260
22	265	176	6.55/6.90	6.60/7.20	-	-
23	445	172	- / -	- / -	-	-
24	307	176	6.60/7.00	6.55/7.60	-	-
25	311	184	6.75/7.35	7.00/7.70	1290	1160
26	282	139	6.80/7.20	6.90/7.80	-	-
27	315	176	6.90/ -	7.50/ -	-	-
28	-	157	6.40/ -	6.80/ -	1460	1520
29	243	161	- /7.20	- /7.60	-	-
30	247	180	6.50/7.35	6.70/7.50	-	-
31	267	175	6.95/6.95	7.15/7.15	-	-

COD, pH and MLSS Raw Data from Section 6.3 (cont.)

Day #	COD (mg/L)		pH		MLSS (mg/L)	
	Raw	Settled	Ferm.1	Ferm.2	Ferm.1	Ferm.2
	Influent	Sewage	Low/High	Low/High		
32	278	186	6.85/7.00	7.15/7.50	–	–
33	249	184	6.75/6.95	6.90/7.30	–	–
34	247	–	6.85/ –	7.10/ –	–	–
35	263	–	6.90/ –	7.10/ –	810	950
36	254	202	6.90/ –	7.00/ –	–	–
37	242	162	– / –	– / –	–	–
38	276	174	6.70/6.90	6.65/6.90	–	–
39	259	179	6.70/6.90	6.75/7.00	882	819
40	246	182	6.70/6.95	6.75/7.05	–	–
41	283	153	6.75/7.05	6.80/7.15	–	–
42	286	149	6.85/7.05	6.95/7.65	–	–
43	372	137	6.75/7.00	6.95/7.35	–	–
44	251	141	6.85/7.10	6.75/7.85	–	–
45	253	157	6.90/7.15	7.05/7.65	–	–
46	286	178	6.80/7.30	6.9597.40	–	–
47	272	182	6.75/7.20	6.85/7.30	–	–
48	260	176	6.85/7.10	6.95/7.35	–	–
49	254	178	6.75/7.10	6.85/7.30	576	1198
50	255	150	6.75/7.00	6.90/7.50	–	–
51	247	144	6.75/6.95	6.90/7.55	–	–
52	405	130	6.80/ –	6.90/ –	–	–
53	482	166	6.85/ –	7.20/7.20	724	620
54	251	174	– / –	– / –	–	–
55	364	190	7.05/7.05	7.35/7.35	–	–
56	247	162	6.60/ –	6.75/ –	799	740
57	225	152	6.75/7.20	6.80/7.55	–	–
58	266	152	6.95/7.30	6.85/7.75	–	–
59	279	202	6.90/ –	7.10/ –	–	–
60	380	165	6.70/ –	6.85/ –	–	–
61	236	158	6.85/7.50	6.95/7.60	–	–
62	250	170	6.95/ –	7.05/ –	808	800
Mean	282	165	6.75/7.05	6.85/7.30	1357	1513

VFA Raw Data from Section 6.3

Day #	Fermenter 1 (mg HAc/L)				Fermenter 2 (mg HAc/L)			
	Acetic	Propion.	Butyric	Total	Acetic	Propion.	Butyric	Total
1	67.0	59.0	7.0	133.0	102.0	87.0	7.0	196.0
2	64.0	47.0	<2.0	111.0	96.0	76.0	8.0	180.0
3	60.0	45.0	<2.0	105.0	92.0	75.0	5.0	172.0
4	29.0	20.0	<2.0	49.0	107.0	75.0	13.0	195.0
5	60.0	44.0	<2.0	104.0	72.0	56.0	<2.0	128.0
6	56.0	43.0	<2.0	99.0	61.0	45.0	<2.0	106.0
7	53.0	36.0	<2.0	89.0	84.0	64.0	13.0	161.0
8	58.0	42.0	<2.0	100.0	84.0	62.0	<2.0	146.0
9	46.0	34.0	<2.0	80.0	72.0	54.0	<2.0	126.0
10	–	–	–	–	–	–	–	–
11	55.0	42.0	<2.0	97.0	62.0	61.0	<2.0	123.0
12	54.0	45.0	<2.0	99.0	79.0	69.0	7.0	155.0

VFA Raw Data from Section 6.3 (cont.)

	Fermenter 1 (mg HAc/L)				Fermenter 2 (mg HAc/L)			
Day #	Acetic	Propion.	Butyric	Total	Acetic	Propion.	Butyric	Total
13	70.0	57.0	<2.0	127.0	104.0	78.0	10.0	192.0
14	53.0	44.0	<2.0	97.0	102.0	96.0	5.0	203.0
15	62.0	52.0	<2.0	114.0	98.0	96.0	6.0	200.0
16	74.1	58.0	<2.0	132.1	111.0	104.0	7.3	222.3
17	66.3	52.0	3.2	121.5	117.0	107.0	7.6	231.6
18	70.6	47.5	3.4	121.5	106.0	80.9	5.0	191.9
19	65.0	44.1	3.2	112.3	98.8	77.6	5.0	181.4
20	62.5	37.0	3.3	102.8	97.7	76.5	6.4	180.6
21	56.2	34.1	<2.0	90.3	92.8	71.9	6.5	171.2
22	55.4	31.8	<2.0	87.2	82.5	59.2	5.0	146.7
23	54.8	30.6	3.5	88.9	79.1	58.5	5.0	142.6
24	53.5	28.3	<2.0	81.8	79.9	55.1	3.5	138.5
25	50.7	30.4	3.5	84.6	69.8	52.3	5.4	127.5
26	-	-	-	-	-	-	-	-
27	50.6	32.1	<2.0	82.7	73.6	53.7	10.0	137.3
28	46.2	32.8	<2.0	79.0	74.3	52.7	6.6	133.6
29	50.0	31.0	3.5	84.5	77.6	55.5	6.0	139.1
30	46.6	27.7	3.4	77.7	71.0	50.0	6.0	127.0
31	50.6	30.2	3.5	84.3	62.5	44.5	5.0	112.0
32	47.4	29.5	3.5	80.4	69.8	51.0	7.1	127.9
33	45.1	25.3	<2.0	70.4	64.0	45.3	5.0	114.3
34	37.6	20.5	<2.0	58.1	47.2	31.2	4.6	82.8
35	38.0	20.6	<2.0	58.6	49.2	33.0	5.0	87.2
36	33.2	18.7	<2.0	51.9	46.8	32.5	4.0	83.3
37	26.5	13.8	<2.0	40.3	45.1	32.5	<2.0	77.6
38	36.0	20.8	<2.0	56.8	47.8	31.2	<2.0	79.0
39	-	-	-	-	38.5	27.2	<2.0	65.7
40	31.7	17.2	<2.0	48.9	55.4	38.4	4.5	98.3
41	32.2	17.8	<2.0	50.0	40.7	28.1	<2.0	68.8
42	30.6	17.1	<2.0	47.7	48.5	34.2	3.0	85.7
43	33.8	17.2	<2.0	51.0	49.9	33.3	4.5	87.1
44	33.8	17.8	<2.0	51.6	43.6	29.0	<2.0	72.6
45	22.8	12.6	<2.0	35.4	43.8	29.7	<2.0	73.5
46	33.5	18.2	<2.0	51.7	39.7	28.0	<2.0	67.7
47	54.9	26.0	<2.0	80.9	69.7	54.6	3.0	127.3
48	36.7	16.0	<2.0	52.7	49.5	34.6	<2.0	84.1
49	35.1	15.5	<2.0	50.6	54.9	33.8	<2.0	88.7
50	44.4	22.2	<2.0	66.6	51.6	39.4	<2.0	91.0
51	32.2	14.5	<2.0	46.7	59.8	41.4	<2.0	101.2
52	34.7	16.0	<2.0	50.7	30.2	43.1	4.0	77.3
53	50.6	26.4	<2.0	77.0	47.2	31.3	3.0	81.5
54	38.3	22.3	4.0	64.6	47.5	32.2	<2.0	79.7
55	32.8	21.1	<2.0	53.9	51.9	33.9	<2.0	85.8
56	38.3	21.3	<2.0	59.6	47.3	30.3	<2.0	77.6
57	30.6	18.5	4.0	53.1	38.4	28.2	5.0	71.6
58	30.5	15.7	5.0	51.2	47.1	30.8	5.0	82.9
59	34.5	17.4	4.0	55.9	46.3	31.1	5.0	82.4
60	31.9	16.1	4.0	52.0	33.2	25.6	5.0	63.8
61	36.6	18.8	<2.0	55.4	48.9	30.0	<2.0	78.9
62	39.1	19.9	<2.0	59.0	51.3	27.7	<2.0	79.0
Mean	46.2	29.4	1.0	76.6	66.7	50.8	4.0	121.5

APPENDIX A3

RAW DATA FROM CHAPTER SEVEN

THE USE OF PRIMARY SLUDGE FERMENTATION

IN THE ACTIVATED SLUDGE PROCESS

COD, VFA, Total P, TKN and MLSS Raw Data from Section 7.1.2

Day#	COD (mg/L) Raw Inf.	COD (mg/L) Sett. Sewage	COD (mg/L) Eff.	VFA (mg HAc/L) Total	Total P (mg/L) Inf.	Total P (mg/L) Eff.	TKN (mg/L) Inf.	TKN (mg/L) Eff.	MLSS (mg/L) Aerobic	SVI
1	360	179	–	164.4	5.3	2.0	31.8	2.3	–	53.4
2	411	208	–	132.6	6.7	2.1	33.1	1.7	–	53.4
3	370	216	–	179.1	5.1	2.0	31.7	2.3	–	55.3
4	366	–	43	189.8	5.0	2.1	32.7	2.5	2530	51.4
5	160	156	–	175.4	4.9	1.6	31.7	1.8	–	51.1
6	146	117	–	198.6	5.4	–	33.3	–	–	–
7	135	115	–	177.4	5.9	–	37.3	–	–	48.5
8	168	129	26	207.0	4.7	1.5	35.7	2.3	2590	46.3
9	175	146	–	226.0	5.7	1.9	34.2	2.3	–	41.9
10	222	173	–	222.4	5.6	2.2	34.4	2.6	–	50.8
11	239	181	55	188.3	5.7	1.8	33.8	1.6	2690	48.3
12	339	204	–	210.0	6.1	2.8	35.4	2.4	–	47.6
13	376	189	–	227.0	6.0	–	36.7	–	–	52.4
14	323	184	–	197.3	5.8	–	33.7	–	–	51.7
15	303	199	32	219.0	5.8	1.9	34.5	2.0	2845	51.0
16	313	226	–	–	5.4	2.1	30.6	1.7	–	–
17	330	177	–	177.3	4.6	3.7	35.8	1.9	–	53.4
18	301	187	60	135.4	5.4	2.3	32.4	1.9	2795	55.5
19	368	182	–	169.2	5.9	2.0	32.4	1.5	–	52.7
20	–	–	–	–	–	–	–	–	–	–
21	–	–	–	–	–	–	–	–	–	–
22	325	199	–	184.7	5.5	2.6	33.9	1.9	3000	56.7
23	362	195	–	159.0	6.8	2.7	36.2	2.3	–	60.0
24	357	195	–	164.7	5.8	2.6	37.2	2.6	–	60.0
25	374	183	60	217.2	5.3	2.5	34.3	2.2	2500	64.0
26	323	172	–	215.9	6.0	1.9	37.7	2.2	–	63.6
27	307	200	–	255.9	6.0	–	38.7	–	–	65.3
28	325	186	58	197.7	6.4	–	39.0	–	–	63.0
29	337	179	–	152.6	6.3	1.6	40.8	2.2	2555	66.5
30	291	171	–	175.5	4.1	1.1	24.7	1.7	–	61.8
31	291	188	–	158.4	4.9	1.6	27.7	1.7	–	61.1
32	300	220	60	–	4.9	1.6	28.7	1.9	2650	67.9
33	351	168	–	154.8	5.4	1.4	31.3	1.8	–	64.2
34	302	181	–	154.4	5.1	–	30.4	–	–	–
35	310	159	–	157.7	4.9	–	31.1	–	–	–
36	336	108	52	145.5	5.5	–	36.8	–	–	–
37	338	172	–	166.3	5.3	1.5	33.2	2.1	–	66.1
38	304	185	–	135.2	4.7	1.6	30.6	1.6	–	73.7
39	295	185	70	145.1	4.8	2.2	30.3	1.8	2645	75.6
40	367	157	–	153.2	5.6	2.1	33.3	1.3	–	71.6
41	338	192	–	176.4	6.3	–	21.8	–	–	–
42	383	194	–	170.6	6.1	–	36.5	–	–	–
43	410	151	32	128.9	5.6	1.8	33.5	1.7	2125	89.4
44	293	–	–	170.9	5.3	1.6	30.1	1.8	–	90.0
45	291	174	–	191.3	7.4	1.9	28.0	–	–	92.9
46	307	192	57	188.6	7.4	2.0	27.9	1.4	2085	86.3
47	263	148	–	201.7	5.1	2.2	27.7	1.7	–	97.6
48	328	163	–	196.5	6.0	–	31.1	–	–	–
49	386	–	–	195.6	6.8	–	31.0	–	–	–

COD, VFA, Total P, TKN and MLSS Raw Data from Sect. 7.1.2 (cont.)

	COD (mg/L)			VFA	Total P		TKN		MLSS	SVI
	Raw	Sett.		(mg HAc/L)	(mg/L)		(mg/L)		(mg/L)	
Day#	Inf.	Sewage	Eff.	Total	Inf.	Eff.	Inf.	Eff.	Aerobic	
50	463	137	71	177.8	6.9	2.1	32.0	1.6	1945	92.5
51	501	169	–	192.1	6.3	2.2	37.2	1.6	–	94.2
52	516	184	–	221.5	6.3	0.5	36.3	1.7	–	88.6
53	–	–	–	–	–	–	–	–	–	85.6
54	401	144	–	238.4	6.8	1.3	42.0	1.6	–	76.0
55	397	222	–	264.8	6.2	–	34.8	–	–	–
56	351	157	–	235.2	5.8	1.3	33.8	–	–	–
57	551	145	56	226.8	6.1	0.2	36.3	1.6	2450	87.8
58	–	169	–	–	5.3	<0.2	21.2	1.8	–	86.7
59	293	165	–	203.8	4.5	<0.2	32.7	1.6	–	85.5
60	348	185	–	191.5	5.2	0.6	32.2	1.9	–	86.5
61	–	149	–	222.2	–	–	–	–	2395	85.6
62	–	159	–	–	6.6	–	43.6	–	–	–
63	320	–	–	–	5.6	–	37.2	–	–	–
64	348	172	–	–	5.7	–	34.2	–	–	85.9
65	340	208	–	182.7	5.3	1.4	32.8	1.4	–	80.2
66	389	190	–	189.4	5.5	1.0	35.6	1.4	–	83.3
67	–	190	–	168.7	8.0	0.8	47.6	1.4	–	84.5
68	332	196	61	205.9	5.5	0.9	28.8	1.3	–	83.6
69	366	200	–	–	6.1	2.0	35.6	–	–	–
70	–	–	–	–	–	–	–	–	–	–
71	–	233	41	181.1	5.5	–	32.8	1.3	–	83.1
72	383	218	–	160.7	4.7	2.1	30.8	1.0	–	82.4
73	311	210	–	173.2	4.9	1.4	28.8	1.3	–	85.3
74	325	210	57	189.9	5.0	1.7	29.6	1.3	–	77.2
75	335	223	–	154.5	5.6	1.8	32.4	1.3	2746	61.9
76	339	208	–	147.5	4.6	–	26.0	–	–	79.2
77	343	226	–	156.9	4.9	–	28.4	–	–	70.0
78	351	251	56	180.2	4.7	1.3	29.4	1.4	3982	62.8
79	341	257	–	136.1	4.9	1.0	29.6	1.8	–	62.8
Mean	332	183	53	184.0	5.64	1.72	33.06	1.79	2619	69.6

Ortho-P and Nitrate Bioreactor Profiles for Section 7.1.2.

	Ortho-P (mg/L)				NO$_3$ + NO$_2$ (mg/L)			
	1st	2nd			1st	2nd		
Day#	Unaer.	Unaer.	Aerobic	Eff.	Unaer.	Unaer.	Aerobic	Eff.
4	2.50	13.50	2.00	2.20	0.24	0.34	9.60	9.20
8	2.20	13.80	2.10	1.60	0.53	0.09	13.40	12.50
11	5.30	11.90	2.20	1.80	0.43	0.64	13.10	12.70
15	2.80	15.00	2.20	2.00	0.06	0.07	14.30	13.60
18	2.00	12.00	2.20	2.30	0.40	0.26	15.10	15.30
22	2.60	9.20	2.60	2.80	0.23	0.21	14.20	13.10
25	3.10	13.70	2.40	2.60	0.06	0.26	14.10	14.10
29	2.00	13.90	1.70	1.60	0.56	0.36	13.90	13.20
32	1.80	11.90	1.40	1.90	0.17	0.10	11.30	12.60
43	2.40	12.10	1.60	1.90	0.30	0.11	14.10	13.70
46	2.10	11.00	1.90	2.00	0.20	0.02	12.40	11.40

Ortho-P and Nitrate Bioreactor Profiles for Sect. 7.1.2. (cont.)

| | Ortho-P (mg/L) | | | | NO$_3$ + NO$_2$ (mg/L) | | | |
| | 1st | 2nd | | | 1st | 2nd | | |
Day#	Unaer.	Unaer.	Aerobic	Eff.	Unaer.	Unaer.	Aerobic	Eff.
50	2.80	11.50	1.90	2.10	0.27	0.08	13.20	12.60
57	3.70	14.50	0.07	0.24	0.04	0.25	12.60	12.00
60	1.20	11.60	0.51	0.50	0.17	0.03	15.30	12.70
67	2.10	11.40	0.83	1.03	-	-	-	-
74	2.67	12.80	1.62	1.82	0.27	0.28	12.00	11.80
Mean	2.58	12.49	1.70	1.77	0.26	0.21	13.24	12.70

COD, Total P, TKN and MLSS Raw Data from Section 7.2.1

| | COD (mg/L) | | Total P (mg/L) | | TKN (mg/L) | | MLSS (mg/L) | SVI |
Day#	Inf.	Eff.	Inf.	Eff.	Inf.	Eff.	Aerobic	
1	281	-	-	-	25.8	-	4089	215.2
2	338	-	-	-	26.6	-	4238	215.9
3	261	21	3.73	1.19	23.9	-	4210	191.1
4	257	-	-	-	23.0	-	3609	252.1
5	245	-	-	-	23.2	-	-	-
6	253	-	-	-	23.5	-	-	-
7	276	19	4.32	2.79	24.8	-	4106	222.8
8	246	-	3.75	2.58	23.8	-	3270	226.3
9	242	-	3.68	1.74	22.4	-	4150	215.7
10	253	26	3.64	1.83	21.2	-	3435	250.4
11	231	-	3.50	1.89	20.4	-	-	253.6
12	256	-	-	-	22.7	-	-	-
13	273	-	-	-	23.8	-	3835	-
14	330	24	4.33	1.90	24.6	-	3968	209.2
15	346	-	3.75	1.72	23.6	-	3995	215.3
16	217	-	3.50	1.55	21.8	-	3457	167.8
17	222	24	3.55	2.08	20.2	-	2982	278.3
18	193	-	3.25	1.25	19.6	-	-	233.8
19	136	-	-	-	14.4	-	-	-
20	146	-	-	-	11.9	-	-	-
21	180	26	2.92	1.95	14.9	-	4647	182.9
22	158	-	3.05	1.56	14.5	-	4409	182.6
23	169	-	3.20	-	17.1	-	4443	191.3
24	196	22	3.50	2.07	16.9	-	3873	213.0
25	254	-	3.52	1.87	17.4	-	-	221.9
26	-	-	-	-	-	-	-	-
27	254	-	-	-	27.9	-	-	-
28	281	25	3.99	2.70	21.2	-	3167	145.2
29	199	-	3.84	2.62	17.9	-	-	252.3
30	203	-	3.60	1.93	19.1	-	-	226.1
31	172	25	3.02	1.09	19.0	-	-	-
32	629	-	5.60	0.65	32.6	-	3821	212.0
33	185	-	-	-	19.3	-	-	216.5
34	-	-	-	-	-	-	3845	206.8
35	-	30	3.67	1.84	-	-	-	207.5
36	194	-	4.12	2.01	22.2	-	-	197.8

COD, Total P, TKN and MLSS Raw Data from Section 7.2.1 (cont.)

Day#	COD (mg/L) Inf.	COD (mg/L) Eff.	Total P (mg/L) Inf.	Total P (mg/L) Eff.	TKN (mg/L) Inf.	TKN (mg/L) Eff.	MLSS (mg/L) Aerobic	SVI
37	211	-	3.67	1.62	20.5	-	3880	208.8
38	185	22	3.56	1.73	18.8	-	3037	123.5
39	140	-	3.30	2.10	18.3	-	3816	197.9
40	224	-	-	-	18.4	-	-	-
41	197	-	-	-	17.0	-	-	-
42	244	18	3.20	2.20	18.7	-	4003	174.9
43	204	-	3.10	1.50	17.1	-	3000	140.0
44	201	-	2.89	1.54	16.6	-	3719	200.3
45	-	-	-	1.30	-	-	3604	216.4
46	278	-	3.14	1.77	19.6	-	3616	217.1
47	173	-	-	-	18.6	-	-	-
48	220	-	-	-	19.5	-	-	-
49	235	20	3.00	2.10	19.1	-	3561	205.0
50	404	-	4.50	1.40	20.6	-	3518	204.7
51	250	-	3.10	1.40	19.8	-	-	201.8
52	209	25	3.00	1.30	19.4	-	-	190.4
53	186	-	2.90	1.70	17.9	-	-	181.9
54	194	-	-	-	18.5	-	-	-
55	-	-	-	-	10.5	-	-	-
56	225	21	3.60	1.70	22.0	-	-	213.2
57	-	-	3.47	1.56	21.2	-	-	213.2
58	-	-	3.18	0.83	20.2	-	-	218.9
59	231	26	3.25	1.00	-	-	-	207.5
60	203	-	3.52	1.10	-	-	-	207.5
Mean	236	24	3.54	1.72	20.2		3777	207.3

Ortho-P and Nitrate Bioreactor Profiles for Section 7.2.1

Day#	Ortho-P (mg/L) Anaer.	Anoxic	Aerobic	Eff.	$NO_3 + NO_2$ (mg/L) Anaer.	Anoxic	Aerobic	Eff.
10	5.48	2.85	2.27	2.12	0.04	0.35	3.36	2.20
17	3.85	2.39	2.21	2.34	0.20	2.58	-	2.82
24	4.50	2.65	2.15	2.15	0.03	0.11	3.10	2.20
38	2.73	2.13	1.99	2.10	0.09	2.20	6.04	2.94
49	4.11	1.94	1.42	1.97	0.14	0.13	4.02	6.42
56	2.62	1.31	0.90	0.98	0.03	0.31	3.24	1.84
Mean	3.88	2.21	1.82	1.94	0.08	0.94	3.95	3.07

COD, VFA, Total P, TKN and MLSS Raw Data from Section 7.2.1

Day#	COD (mg/L) Raw Inf.Sewage	COD (mg/L) Sett. Eff.	VFA (mg HAc/L) Total	Total P (mg/L) Inf.	Total P (mg/L) Eff.	TKN (mg/L) Inf.	TKN (mg/L) Eff.	MLSS (mg/L) Aerobic	SVI
1	381 220	-	152.2	6.90	-	43.8	-	-	89.8
2	444 224	51	138.4	7.30	0.92	40.4	1.9	3368	102.4
3	374 203	30	120.0	5.90	1.70	-	-	-	142.8

COD, VFA, Total P, TKN and MLSS Raw Data from Sect. 7.2.2 (cont.)

Day#	COD (mg/L) Raw Inf.	COD (mg/L) Sett. Sewage	COD (mg/L) Eff.	VFA (mg HAc/L) Total	Total P (mg/L) Inf.	Total P (mg/L) Eff.	TKN (mg/L) Inf.	TKN (mg/L) Eff.	MLSS (mg/L) Aerobic	SVI
4	386	199	–	136.4	6.10	2.10	–	–	–	134.5
5	443	232	–	128.8	6.00	2.20	34.0	1.4	–	160.2
6	415	242	–	132.1	6.80	2.10	33.2	1.6	3620	154.7
7	343	244	–	145.4	6.40	–	32.4	–	–	123.6
8	380	234	–	154.2	6.20	–	36.0	–	–	144.8
9	528	230	45	140.0	5.50	1.70	32.8	1.9	–	111.4
10	333	211	–	138.2	5.50	1.20	32.6	1.2	3701	108.1
11	296	185	–	151.1	7.40	0.92	40.4	1.4	–	101.8
12	296	218	47	146.9	5.30	0.89	30.4	1.6	–	111.6
13	325	195	–	148.7	5.20	0.89	28.4	1.7	3796	106.7
14	420	191	–	153.9	6.00	–	35.6	–	–	–
15	329	210	–	145.9	7.20	–	43.8	–	3895	125.8
16	420	222	51	150.1	5.80	1.00	37.4	1.3	–	126.7
17	319	181	–	124.3	5.50	1.20	36.2	1.8	–	130.2
18	332	181	–	168.8	5.80	1.40	33.5	2.2	4044	138.5
19	282	151	48	158.8	5.30	1.80	34.4	1.7	–	140.0
20	290	193	–	167.8	6.10	0.91	31.5	1.3	4099	131.7
21	351	218	–	184.7	6.20	1.90	36.7	1.7	–	139.1
22	347	197	–	174.1	6.30	2.40	36.3	1.4	–	114.6
23	376	224	32	157.8	6.30	1.80	36.2	1.7	–	109.7
24	324	212	–	133.7	5.40	2.20	31.2	1.7	4101	–
25	320	201	–	119.8	5.50	2.00	31.0	1.5	–	102.4
26	346	197	38	118.6	5.50	2.60	31.0	1.6	–	105.3
27	309	180	–	128.7	5.30	2.20	29.4	1.6	3931	–
28	284	172	–	126.0	4.70	2.10	24.8	1.4	–	–
29	258	148	–	153.8	4.30	2.30	26.4	1.3	–	91.3
30	252	176	32	129.2	4.30	2.10	26.4	1.3	3769	90.2
31	349	145	–	135.1	3.80	2.20	25.2	1.4	–	93.8
32	243	137	–	142.1	3.80	1.90	23.8	1.1	–	94.8
33	259	147	31	128.4	4.00	1.80	24.6	1.4	–	93.0
34	280	157	–	138.2	4.50	2.80	25.8	1.1	–	–
35	306	145	–	177.9	5.00	–	29.2	–	3577	91.3
36	337	186	–	152.1	5.50	1.70	34.4	1.1	–	92.3
37	300	165	39	158.8	5.30	1.30	32.8	1.4	–	94.9
38	308	186	–	143.3	4.60	0.91	29.2	1.5	3589	94.8
39	321	147	–	155.1	4.50	0.82	28.8	2.2	–	94.7
40	–	112	33	121.0	4.80	0.14	32.0	2.0	–	–
41	323	167	–	123.6	4.80	1.16	30.8	2.2	3571	92.4
42	275	131	–	135.2	5.60	0.78	34.8	2.1	–	–
43	285	159	–	173.9	5.30	0.94	32.6	1.3	–	90.3
44	327	169	37	166.7	5.60	0.99	35.6	1.2	–	89.3
45	291	167	–	161.2	4.60	1.00	31.4	1.5	3738	88.3
46	339	185	–	150.6	4.80	0.68	31.6	1.3	–	86.3
47	283	181	33	159.0	4.60	0.62	29.6	1.3	–	87.1
48	299	187	–	157.2	5.00	0.72	30.6	1.6	3641	90.6
49	267	179	–	165.0	4.20	0.78	29.6	1.3	–	89.3
50	263	158	–	205.8	5.10	1.10	36.8	1.3	–	89.3
Mean	330	186	39	147.6	5.43	1.47	32.4	1.5	3791	109.1

Ortho-P and Nitrate Bioreactor Profiles for Section 7.2.1

Day #	Ortho-P (mg/L)				NO$_3$ + NO$_2$ (mg/L)			
	Anaer.	Anoxic	Aerobic	Eff.	Anaer.	Anoxic	Aerobic	Eff.
2	9.10	5.40	2.20	2.20	0.35	5.80	15.70	15.80
5	11.70	6.50	2.80	2.20	0.20	0.67	13.30	13.10
9	14.90	6.00	1.60	3.60	0.22	2.50	11.50	11.60
12	14.50	3.40	1.00	0.85	0.43	3.50	12.90	12.50
19	10.30	5.40	1.70	1.70	0.05	3.80	13.10	12.10
23	9.00	4.50	1.20	1.20	0.12	0.34	12.10	11.30
26	10.70	5.70	2.50	2.60	0.16	3.10	11.30	11.50
30	7.70	4.60	2.40	2.20	0.24	4.40	10.60	10.30
33	9.10	5.40	1.46	1.70	0.16	-	9.70	9.00
37	11.10	5.90	1.20	1.20	0.19	3.10	13.00	12.40
40	11.40	6.00	1.30	1.00	0.23	1.90	11.10	11.10
44	13.30	7.40	0.96	0.98	0.05	2.30	12.70	12.20
47	7.30	3.00	0.40	0.76	0.29	2.60	11.50	11.50
Mean	10.78	5.27	1.59	1.71	0.35	2.74	12.19	11.88