AN INVESTIGATION OF THE FEASIBILITY OF NITRIFICATION AND DENITRIFICATION OF A COMPLEX INDUSTRIAL WASTEWATER WITH HIGH SEASONAL TEMPERATURES

by

Andrew R. Sabalowsky

Thesis submitted to the Faculty of

Virginia Polytechnic Institute and State University

in the partial fulfillment of the requirements for the degree of

Master of Science

in

Environmental Engineering

APPROVED:

Dr. Clifford W. Randall, Chairman
Dr. Nancy G. Love
Dr. John T. Novak

April, 1999

Blacksburg, VA

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(ABSTRACT)

The wastewater treated at the Hopewell Regional Wastewater Treatment Facility (HRWTF) is very unique both because it is comprised of effluents of seven different industries in the area in addition to the domestic wastewater in the area, and because it reaches high temperatures in the basins, often above 45^{o}C during the summer. Four different bench scale systems consisting of continuously stirred tank reactors (CSTRs) in series were operated during the summer of 1997 to quickly assess the feasibility of nitrifying and denitrifying the total flow at HRWTF down to a final effluent total nitrogen concentration of 10 mg-N/L or less. The four main treatment strategies tested were: aerobic/anoxic treatment of the final effluent of HRWTF at moderate temperatures (approximately 30^{o}C); anaerobic/anoxic/aerobic (A2/O) treatment of the primary effluent of HRWTF at moderate temperatures; treatment of the effluent of one of the industries which had a high ammonia wastewater and which was originally believed to contain nitrification inhibitors; and fully aerobic treatment of the primary effluent of HRWTF at high temperatures (of approximately 40 to 45^{o}C) with an activated sludge gradually acclimated to such temperatures over the course of two months. At the end of the study, a two-week high temperature study was conducted on the system which had been treating the secondary effluent all summer with the same activated sludge which was acclimated only to temperatures around 30^{o}C. The fully aerobic high temperature system which had been nitrifying the primary effluent all summer was converted to a modified Lutzack-Ettinger (MLE) process at the end of the study to test whether the primary effluent could be denitrified as well as nitrified at high temperatures with the sludge acclimated to high temperatures. All four of the main treatment strategies demonstrated that nitrification and denitrification of either the total flow or the high ammonia side stream could be achieved down to the desired total nitrogen concentrations. The high temperature study conducted on the system which had been treating the secondary effluent all summer indicated that the sudden increase from approximately 30^{o}C to approximately 40^{o}C over a twenty-four hour period, similar to the sudden temperature increase which occurs every spring at HRWTF, quickly ends nitrification in a system not acclimated to high temperatures, while denitrification and COD removal is hardly affected by such a temperature change. While the nitrification performance of the gradually acclimated system treating the primary effluent at high temperatures was adequate, problems maintaining a consistent MLVSS or ETSS concentration suggested that the high temperatures seen in the basins at HRWTF are likely to make consistent treatment difficult. As a result of considering both capital cost requirements and quality of treatment, the bench scale testing suggested that the most likely candidates for successful treatment of the total flow down to desired total nitrogen concentrations would involve either the A2/O treatment of the primary effluent of HRWTF, possibly with the addition of a cooling tower, or A2/O treatment of the high ammonia side stream, possibly involving the dilution of the wastewater with one of the other flows sent to HRWTF. It was concluded that pilot scale evaluation of the two options was required for a final design decision, and pilot scale evaluation was being performed when this thesis was completed.

ACKNOWLEDGEMENTS

This thesis was made possible only through the efforts of, literally, dozens of people, and I hope I am not failing to recognize any of them here (although, inevitably, I'm sure I will).

I would first like to thank those who served as my committee (Dr. Nancy Love, Dr. John Novak, and Dr. Clifford Randall) for their knowledge, patience, and understanding. Whether or not they were conscious of this fact, I have learned a great deal from them over the past few years about microbial processes, engineering, and myself. Special thanks are extended to Dr. Randall for the countless hours he has spent with me.

This project was initiated and funded by Malcolm Pirnie, Inc. I would like to thank all of those at Malcolm Pirnie involved for both the technical and the more tangible support provided which helped me continue my education. Special thanks to Alison, Erika, and Nikolai for their advice, input, and assistance.

Virtually all chemical analyses performed during this project were by the laboratory staff at HRWTF. This required their working extra hours for the duration of the project, to which I am very grateful. Special thanks must go to all those on the TPPEE staff for their efforts and ideas, and I am grateful to every person at HRWTF, both "up the hill" and "down the hill" for their moral support (there are too many folks here to name without guarenteeing that I leave someone out of the list). Although many of the guys "down the hill" were not directly involved in the project, I would like to thank all of them who took their time to talk with me and teach me a great deal about good engineering, the truths of wastewater treatment plant operation, how to laugh under almost any circumstance, how to find and avoid the hundreds (if not thousands) of black widow spiders hiding out at "the Regional", and about life in general.

I owe thanks to all of my comrades at VPI (many of whom have graduated and left already) who have provided me with valuable brainstorming, stimulating conversations and ideas, or just a good laugh.

Although I may have failed to provide them with a good understanding of what exactly it is I do, thanks to all my friends and family for their encouragement and just being there in general.

Finally, I would like to extend my sincerest gratitude to Dr. and Mrs. Randall for their spiritual guidance and support. Burdens are much easier to carry with the simple knowledge that someone cares.

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I. INTRODUCTION

Up until the late 1960s, the major goal of wastewater treatment in the United States was to achieve BOD removal, while avoiding nitrification due to the substantial savings in capital costs. The virtual impossibility of achieving nitrification without having to expand existing plants, as well as the problem of rising sludge in clarifiers due to unintended denitrification further complicated the issue (Wild *et al.*, 1971). The instability due to trace amounts of organics and susceptibility of biological nitrification to system upsets has led to considerable reluctance by the industrial sector to implement biological nitrification processes (Neufeld *et al.*, 1980). There is approximately an order of magnitude difference in sensitivities between *Nitrosomonas* (the organisms commonly accepted as being responsible for ammonia oxidation in wastewaters) and aerobic heterotrophs responsible for simple carbon oxidation, as well as substantially slower growth kinetics of nitrifiers (Blum and Speece, 1991). The slow growing nitrifiers tend to be more sensitive than heterotrophs to temperature, pH, sludge age, the quantity and kind of organics present, and known inhibitory substances (Churchwell *et al.*, 1980); making it obvious why, from a treatment plant operation perspective, the avoidance of nitrification would be preferred. Today it is understood, as reflected by increasingly stringent regulations, that there are numerous reasons which demand the nitrification, and possibly denitrification, of wastewaters, and that the above mentioned difficulties are obstacles to be overcome, not excuses to maneuver around.

The waste stream in this study is a complex mixture of industrial effluents and municipal wastewater which are all treated in a single mixed flow at the Hopewell Regional Wastewater Treatment Facility (HRWTF). The wastewater is comprised of approximately 15% domestic waste and 85% industrial waste. Industries contributing effluents to HRWTF include several chemical companies, a pulp mill, and a textile factory. HRWTF treats approximately 35 MGD, with a capacity to treat up to 50 MGD and has previously not observed substantial nitrification or denitrification in its existing configuration (consisting of primary treatment for solids removal and a high purity oxygen, UNOX secondary treatment). This study is devoted to investigating how to achieve a final effluent total nitrogen (TN) concentration of 10 mg-N/L or less to meet the requirements of future regulations to be imposed on HRWTF. Present final effluent TN concentrations are approximately 37 mg-N/L.

What are problems most likely to be encountered in this study?

Preliminary batch studies of the waste streams tested in this study determined that two major factors would make a biological nitrification/denitrification process difficult to maintain. The first problem investigated in this study was associated with the presence of a compound inhibitory to nitrification in one of the streams. Specifically, methylethyl ketoxime (MEKO) was found in the stream contributed by one of the industries, which was determined to be at levels inhibitory to nitrification (Malcolm Pirnie, Inc., 1996; Malcolm Pirnie, Inc., 1997). It was determined after data was collected from this study however, that processes had been changed, and the previously determined inhibitory stream no longer contained any MEKO. The treatment of that stream will still be discussed for the valuable information gained with regard to treating a stream with high influent ammonia concentrations (approximately 130 mg-N/L) and the viability of treating the nitrogen-containing waste streams separately from the other streams to achieve the above stated effluent quality goal.

The second difficulty in this study was associated with the high temperatures encountered at the plant, due primarily to the flow contributed by a pulp mill. Summer temperatures of the combined flows from all sources are typically 45 to 50^{o}C, with final effluent temperatures at HRWTF being above 40^{o}C generally from May to October. The problem of maintaining a high-temperature nitrifying population is compounded by the fact that every April the pulp mill has a shutdown which lasts one to three weeks, resulting in a drop in temperature of the combined flows from approximately 38^{o}C to 27^{o}C. Once the pulp mill is back on line, the temperatures rise from moderate conditions to above 40^{o}C within a few days (Mitchell, 1999).

As a result of previous batch testing to determine what the greatest threats to successful biological nitrification/denitrification at HRWTF were, four treatment options to be modeled as bench-scale continuously stirred tank reactor (CSTR) configurations were selected. Options tested entailed determining the treatability of the total flow at HRWTF (believed to have trace amounts of inhibitory compounds) and the high ammonia side stream (previously believed to be toxic). Their design will be discussed in the "Materials and Methods" section of this thesis. A general background to the issues surrounding this research will first be discussed in the following chapter.

II. LITERATURE REVIEW

Because this is a feasibility study, most of the discussion in the literature review is devoted to understanding what biological nitrification and denitrification is, and what factors are known to affect the process. Testing during this project revealed that there did not appear to be any compounds in the wastewaters which were inhibitory to nitrification or denitrification, and that substrate inhibition also was not a problem, even in the high ammonia side stream. Thus, discussion most pertinent to understanding the problems specific to this wastewater and how to overcome them pertains to high temperature studies, as well as the possible role of consortia dynamics. There is relatively little research pertaining to high temperature nitrification, however, primarily because the main concern of wastewater treatment plants is actually achieving nitrification at low, winter temperatures. What has been learned about high temperature effects is included in this review of literature, as are other general issues pertaining to nitrification and denitrification of industrial wastewaters. Because nitrification is known to be the most sensitive of the two processes, most discussion pertains to how nitrification is affected by various factors.

What is Biological Nitrification and Denitrification?

Nitrogen exists in many forms because of the high number of oxidation states it can take. The oxidation state for nitrogen is -III in its most reduced form, i.e., in ammonia and organic nitrogen compounds (the forms most closely associated with plants, animals, and domestic wastewaters). At the other extreme is the most oxidized form of nitrogen, i.e. nitrate, where the oxidation is +V (Sedlak, 1991). Nitrogen in untreated wastewaters is primarily in the form of ammonia and organic nitrogen, both soluble and particulate, with most particulate organic-nitrogen being transformed to ammonium and other organic forms during biological treatment. The major forms of organic-nitrogen are proteins and amino acids which yield ammonia upon biodegradation, while there is generally little to no nitrate or nitrite in untreated wastewaters (Metcalf and Eddy, 1991b; Barnes and Bliss, 1983). Additionally, especially in industrial wastewaters, there may be any of a variety of organic nitrogen-containing compounds which are not only compounds not produced biologically, but which can be inhibitory to either heterotrophic or autotrophic cells involved in biological wastewater treatment processes. In this study, the primary sources of nitrogen are due to the 15% domestic wastewater and the high ammonia wastewater of one of the industries.

While some of the ammonia-nitrogen is incorporated into cell mass, the majority of organic- and ammonia-nitrogen is oxidized by dissimilatory processes, ultimately to nitrate (NO_3^-). It is this energy-producing, dissimilatory process which refers to biological nitrification. Similarly, some nitrate-nitrogen may be reduced to ammonia-nitrogen and assimilated into cell mass, but the bulk of the nitrate-nitrogen is removed from wastewater via dissimilatory metabolism, ultimately to nitrogen gas (N_2). It is this dissimilatory process which is regarded as biological denitrification.

Why is Nitrification Important?

The three major reasons that at least nitrification and ideally the denitrification of effluents is desired are the avoidance of eutrophication of receiving bodies of water, reducing the likelihood of ammonia toxicity to higher organisms in the receiving bodies of water, and, especially in the case of nitrifying/denitrifying sludges, the reduction of sludge production and therefore handling costs.

Nitrification exerts an oxygen demand, thus, it is important to minimize, if not eliminate, the amount of ammonia being discharged (Barnes and Bliss, 1983; Churchwell *et al.*, 1980). It is not only ammonia-nitrogen which can lead to eutrophication in receiving bodies of water, however. Because other forms of nitrogen, such as nitrate, are nutrients, high loads of nitrogen in nearly any form are likely to lead to algal blooms or the excessive growth of higher aquatic plants, creating a heavy oxygen demand or possibly odor problems (Barnes and Bliss, 1983; Metcalf and Eddy, 1991b; Prosser, 1989; Sedlak, 1991). Thus, it is apparent that not only the nitrification of a wastewater is desired, but also the denitrification, in order to minimize the likelihood of eutrophication.

The main reason to nitrify a wastewater without denitrification is to simply remove ammonia-nitrogen. Such a goal is desired apparently to minimize ammonia toxicity to aquatic organisms (Churchwell *et al.*, 1980; Prosser, 1989; Metcalf and Eddy, 1991b; Sedlak, 1991). It has been shown that ammonia (NH_3) is toxic, especially to higher aquatic organisms, such as fish, at concentrations as low as 0.5 mg/L (Barnes and Bliss, 1983).

In addition to a cleaner effluent in terms of nitrogen discharged, better BOD removal can be realized as well when comparing a nitrifying/denitrifying sludge which undergoes alternating aerobic and anoxic environments to a conventional, fully aerobic, sludge (McClintock *et al.*, 1988). An even greater bonus realized is that up to 40% less sludge production has been observed in such a comparison, with the added benefit of less aeration costs (McClintock *et al.*, 1988). Other studies have also shown that the yield from nitrate respiration of denitrifying sludges is lower than conventional aerobic carbonaceous oxidation, resulting of course, in less sludge production

and handling costs for similar carbonaceous discharges, but with the added bonus of far superior effluent quality in terms of nitrogen concentrations (Barnes and Bliss, 1983).

There are other reasons why nitrification or combined nitrification/denitrification of a wastewater is desired. If the receiving bodies of water are to be used as a drinking water supply, high nitrogen concentrations can create a greater chlorine demand, and high nitrates can make the source a drinking hazard, especially for infants, resulting in methemoglobinemia (Barnes and Bliss, 1983; Metcalf and Eddy, 1991b; Sedlak, 1991). Nitrification occurring in receiving bodies of water can also result in corrosion of cement structures and natural stones as well (Prosser, 1989).

Aside from preventing negative effects, a biological nitrification/denitrification process can produce benefits over a process which does not nitrify at all, or which nitrifies without denitrifying. For instance, denitrification reduces aeration costs because organics are oxidized with nitrates as the terminal electron acceptor, requiring less oxygen to remove any remaining BOD. Biological denitrification occurring in a basin results in a better settling sludge and reduction or elimination of denitrification occurring in the clarifiers of a nitrified effluent, thereby reducing the problem of floating sludge. A denitrifying sludge tends to be less susceptible to bulking as well. Finally, biological denitrification results in the production of alkalinity, helping maintain a stable environment for a nitrifying sludge while saving on the cost of chemicals required for pH control (Gray, 1990).

Organisms Responsible for Nitrification and Denitrification

Determinations of what organisms were present or responsible for nitrification were not conducted during this study, but it is important to have a basic understanding of what organisms are known to carry out nitrification (as well as denitrification) and how it is possible for them to achieve it. Thus, discussion below will be devoted to what organisms are known to nitrify (and denitrify), how they achieve it, and what conditions are typically required. All such factors are important in understanding what conditions must be maintained and monitored, and how best to do so.

For the most part, nitrification in wastewater treatment is commonly regarded as a two-step process. The first step is the conversion of ammonia to nitrite, generally considered to be the task of *Nitrosomonas*, and the second step is the further oxidation of nitrite to nitrate, which is commonly accepted as the process carried out by *Nitrobacter* (Antoniou *et al.*, 1990; Charley *et al.*, 1980; Sedlak, 1991; Wood et al, 1981). Both of these genera are autotrophic, meaning inorganic carbon, such as CO_2 and its aqueous forms act as the carbon supply for synthesis of new cells, while the oxidation of inorganic nitrogen serves as the energy source (Metcalf and Eddy, 1991b; Antoniou *et al.*, 1990). It must be pointed out, however, that *Nitrobacter* is not an obligate autotroph, and can actually grow using organic carbon, although at a slower rate than either its own autotrophic growth on nitrite or the growth rate of other heterotrophs (Churchwell *et al.*, 1980; Prosser, 1989).

In general, *Nitrosomonas* and *Nitrobacter* are assumed to be responsible for nitrification in wastewaters, and with fairly good reason. In addition to wastewater treatment kinetics and chemistry matching up well with the numerous studies conducted with the two isolates, *Nitrosomonas europaea* and *Nitrobacter agilis* are by far the most common isolates found in wastewater treatment systems, with *Nitrosomonas monocella* and *Nitrobacter winogradskyi* also being commonly isolated (Barnes and Bliss, 1983). Fairly recently, however, it has been discovered that there are, in fact, many genera and species of nitrifiers which carry out nitrification in wastewaters. This should come as no surprise, as each wastewater is fairly unique in its overall chemical composition, and there are several genera of autotrophic bacteria already known to be associated with nitrification. Among the autotrophic ammonia oxidizers are *Nitrosomonas*, *Nitrosococcus*, *Nitrosospira*, *Nitrosolobus*, *Nitrosovibrio*. Among the nitrite oxidizers are *Nitrobacter*, *Nitrococcus*, *Nitrospira*, and *Nitrospina*. Both groups of autotrophic nitrifiers consist solely of Gram negative bacteria (Prosser, 1989).

In addition to the variety of autotrophic microbes capable of nitrification, there are heterotrophs known to be capable of nitrification, most of which appear to be soil organisms. Truly solid evidence for heterotrophic nitrification has been hard to come by, however, as the evidence has been based upon indirect methods (Pennington and Ellis, 1993). Among the organisms believed to be responsible for heterotrophic nitrification are fungi, actinomycetes, and bacteria, with most research having been performed on aspergilli, streptomycetes, arthrobacters, and *Alcaligenes* species. Fungi are considered to be the most efficient and most common heterotrophic nitrifiers, but are found primarily in soil (Prosser, 1989). Many common denitrifiers appear to be capable of heterotrophic nitrification, which appears to occur simultaneously with denitrification, although the apparent nitrification rates can be quite poor (Pel *et al.*, 1997 citing Robertson *et al.*, 1988; Prosser, 1989 citing Castignetti and Hollocher, 1984).

The most promise for high temperature nitrification may involve methanotrophs. There appears to be a good deal of evidence that there are methanotrophic nitrifiers, as indicated by the three studies cited by Pel *et al.* (1997). Nitrification at temperatures as high as 53^{o}C was achieved, presumably by methanotrophs, although the

evidence was indirect (Pel *et al.*, 1997). In a study conducted by Prosser (1989), ammonia oxidation by methanotrophs appeared to occur, but at much slower rates than ammonia oxidation by *Nitrosomonas*. Slow kinetics can be compensated for with larger basins (within reason) if necessary however.

Another possible way for high temperature nitrification to occur may hinge upon consortia dynamics. For instance, an *Arthrobacter* sp. and *Corynebacterium* were isolated out of an estuarine environment in one study (Prosser, 1989 citing Rho, 1986). Little to no nitrite or nitrate production was observed when grown separately, but an order of magnitude increase in NO_x production was observed when the two organisms were grown in co-culture. Although the study did not involve high temperatures, it was shown that a consortium which does not include nitrifiers could yield nitrification. Such consortia dynamics may be a key to successful nitrification in a wastewater as hostile to nitrifiers as the one in this study.

Unlike nitrification, denitrification does not require specialist bacteria, and can be carried out by many facultative heterotrophs. Conversion of nitrate-nitrogen to readily removable forms of nitrogen, such as N_2, N_2O, or NO is carried out by a wide variety of bacteria, including the genera: *Achromobacter*, *Aerobacter*, *Alcaligenes*, *Bacillus*, *Brevibacterium*, *Denitrobacillus*, *Flavobacterium*, *Lactobacillus*, *Micrococcus*, *Proteus*, *Pseudomonas*, *Spirillum*, and *Xanthomonas* (Metcalf and Eddy, 1991a; Gray, 1990 citing Painter, 1970). These bacteria are all heterotrophs capable of dissimilatory nitrate reduction.

How is Biological Nitrification and Denitrification Achieved?

The below discussions will be based upon autotrophic nitrification, which is commonly accepted to be the means by which wastewater is nitrified, and which will be assumed to be the means of nitrification for this study, although it is understood that different organisms and biochemistries may have been at work.

Brief biological description. Nitrification is defined as the oxidation of nitrogen from the -III state to the +V oxidation state through chemical combination with oxygen (Quinlan, 1984 citing Focht and Verstraete, 1977). Nitrification is defined as the oxidation of reduced nitrogen compounds, as opposed to simply the oxidation of ammonium, in order to allow for the encompassing of heterotrophic nitrification (Prosser, 1989). In general, nitrification occurs at a detectable rate only in the presence a select group of chemoautotrophic and heterotrophic bacteria, with the majority of nitrification being attributed to two groups of chemoautotrophs. It is commonly accepted to be a two step process. The first step of nitrification, oxidizing ammonia-N(-III) to nitrite-N(+III), is attributed primarily to the autotrophic *Nitrosomonas europaea*; while the second step, oxidizing nitrite-N(+III) to nitrate-N(+V), is considered to be dominated be the species *Nitrobacter winogradskyi* (Metcalf and Eddy, 1991a; Quinlan, 1984 citing Focht and Verstraete, 1977).

The complete biochemistry of ammonia or nitrite oxidation is still not fully understood, however. There is still no solid evidence demonstrating whether it is the ammonium ion or free ammonia (or both) which gets transported into the cells of ammonia oxidizers, nor is it known how exactly the transport occurs (Prosser, 1989). It is generally believed, however, that unionized ammonia (NH_3), as opposed to the ammonium ion (NH_4^+), serves as the actual substrate for oxidation by *Nitrosomonas* (Prosser, 1989; Quinlan, 1984), and that free nitrous acid (HNO_2) appears to be the substrate of *Nitrobacter* instead of nitrite (Churchwell *et al.*, 1980 citing O'Kelly *et al.*, 1970). The form of the substrates are functions of pH and temperature equilibria. Because it has not been clearly determined if ammonia (NH_3) or ammonium (NH_4^+) is the actual substrate, the terms "ammonia" or "NH_x" will be used for both species of nitrogen from this point forward unless a distinction is otherwise specified. For the sake of this study, because no significant nitrite (NO_2^-) buildup was observed, nitrification was analyzed as a one step process, with the substrate ammonia being oxidized fully to nitrate (NO_3^-).

Nitrogen removal, as opposed to mere nitrogen oxidation, is achieved by either assimilation (followed by cell wastage) or by the conversion of nitrate ultimately to nitrogen gas (Metcalf and Eddy, 1991a; Sedlak, 1991). This second means, transforming nitrates to nitrogen gas, is what is meant by biological denitrification and is achieved by denitrifying organisms in the absence of molecular oxygen, although an organic carbon source is required (Sedlak, 1991). Like nitrification, denitrification is also regarded as a two step process, with step one of the process being the reduction of nitrate to nitrite. This step is followed by the production of nitric oxide (NO), nitrous oxide (N_2O), and nitrogen gas (N_2). All three products are gases and can be released into the atmosphere (Metcalf and Eddy, 1991a; Sedlak, 1991). Unlike, nitrification, however, there do not appear to be specialized organisms responsible for the process, and the two steps are not necessarily performed by two separate organisms.

Alkalinity production and consumption. Note that there is alkalinity consumption during the nitrification process, and alkalinity production during the denitrification process. In practice, approximately 7.14 g of alkalinity as $CaCO_3$ are consumed for every g of NH_3-N oxidized to NO_3^--N (Azevedo *et al.*, 1995; Sedlak, 1991). The hydroxide ions produced during the denitrification process result in the replacement of 50% of the alkalinity consumed during nitrification. Thus, for a nitrification/denitrification process, the overall theoretical

alkalinity consumption is only 3.57 g of alkalinity as $CaCO_3$ per g of NH_3-N oxidized to NO_3^- (Azevedo *et al.*, 1995; Barnes and Bliss, 1983; Sedlak, 1991). In the case where CO_2 is not stripped, such as high purity oxygen systems which are covered, a change in alkalinity as much as 10 g for every g of nitrogen oxidized may result, however (Sedlak, 1991).

Heterotrophic vs. autotrophic nitrification. Both *Nitrosomonas* and *Nitrobacter* are autotrophic, using inorganic carbon (carbon dioxide, bicarbonate, or carbonate) for synthesis (instead of organic carbon) and ammonia or nitrite to derive energy. The whole process of nitrification and growth lies in a very delicate balance, as both groups of nitrifiers are inhibited by high concentrations of their own substrates and have little energy to spare for high affinity (i.e. low substrate concentration) uptake systems (Prosser, 1989). The vast majority of energy derived from their respective inorganic-nitrogen oxidation must be used to produce reducing power for the fixation of carbon for growth via the Calvin cycle (Barnes and Bliss, 1983; Prosser, 1989). It has been estimated that 80% of the energy generated by autotrophs is used to fix CO_2 (Prosser, 1989 citing Kelly, 1978); and that both groups of autotrophic nitrifiers must convert approximately ten times their own cell weight of ammonia-N or nitrite-N in order to double in mass (Prosser, 1989).

Unlike autotrophic nitrification, where nitrification is required in order to generate energy necessary for growth, it is generally accepted that heterotrophic nitrification is *not* linked to cellular growth, and the majority of nitrification appears to occur in heterotrophs in the stationary phase of growth (Pennington and Ellis, 1993 citing Focht and Verstraete, 1977). It is during the stationary growth phase that most products of apparent heterotrophic nitrification have been formed, and (as of 1989), no heterotrophic nitrification has been demonstrated to be associated with energy production or growth, but rather, is considered to be endogenous or secondary metabolism (Prosser, 1989). This would help explain why nitrification occurring at significant rates is generally attributed to autotrophic nitrification. Unlike autotrophic nitrification which is associated primarily with ammonia, nitrite, and to a minor extent hydroxylamine oxidation, heterotrophic nitrification is associated with ammonia, nitrite, hydroxylamine, hydroxamic acids, amino or oxime nitrogen, and aliphatic and aromatic nitro compounds. Products include NO2-, NO3-, and various nitrogenous organics (Prosser, 1989).

Electron donors required for denitrification. Biological reduction of nitrate to nitrite and N_2 gas requires a suitable electron donor, which is usually an organic compound, such as acetic acid, citric acid, acetone, and methanol, with methanol being the most preferred compound commonly used at plants because it is inexpensive, easy to get, and very effective (Barnes and Bliss, 1983 citing McCarty *et al.*, 1969 and U.S. EPA, 1975). Industrial and agricultural wastes can be an inexpensive option, providing they are easy to obtain consistently and contain no compounds which are inhibitory to the denitrification process (Barnes and Bliss, 1983). Additionally, methane (Barnes and Bliss, 1983 citing Rhee and Fuhs, 1978) and elemental sulfur (Barnes and Bliss, 1983 citing Batchelor and Lawrence, 1978) are known to be acceptable electron donors for denitrification. The wastewater itself may provide a sufficient carbon source, and endogenous respiration can even be used for denitrification, but at a much reduced rate (Barnes and Bliss, 1983; Gray, 1990; Metcalf and Eddy, 1991b). In the case of using a good exogenous carbon, such as methanol, roughly a 3:1 ratio of available carbon to oxidized nitrogen is typically required for zero-order denitrification (i.e., for denitrification to proceed at a rate independent of the amount of organics present) (Barnes and Bliss, 1983). In practice, 25% to 30% of methanol required for denitrification is used for cell synthesis (Metcalf and Eddy, 1991b).

The rate of denitrification depends primarily upon the nature and concentration of the organic matter present. It is commonly accepted that denitrification is zero-order with respect to nitrate concentration down to very low levels (Sedlak, 1991). If there is excess electron donor present for denitrification requirements, denitrification will occur at a rate independent [zero order] of the electron donor concentration (Barnes and Bliss, 1983 citing McCarty *et al.*, 1969 and Dawson and Murphy, 1972). However, using readily biodegradable electron donors such as methanol can yield denitrification rates 10 times those obtained from sludges using wastewater or endogenous respiration (Barnes and Bliss, 1983 citing U. S. EPA, 1975). Furthermore, *complete* denitrification in either fixed film or suspended growth processes is generally feasible only with the use of a readily degradable carbon source such as methanol (Barnes and Bliss, 1983).

Electron acceptors which may interfere with denitrification. An alternative electron acceptor is required under conditions where low or zero dissolved oxygen is present. Inorganic anions such as nitrate, phosphate, and sulfate generally serve this purpose. Under aerobic conditions, oxygen is the preferred electron acceptor, and aerobic oxidation will be the predominant reaction. Nitrate is the next-most favored electron acceptor, offering much more energy gain than anaerobic pathways (Barnes and Bliss, 1983). Since less energy is gained using NO_3^- as the electron acceptor, O_2 is used preferentially and denitrification will occur only at low to zero DO concentrations (Barnes and Bliss, 1983). A denitrification rate of 0.006 mgNO_3^--N/mgVSS/day has been reported under aerobic conditions (Sedlak, 1991 citing Christensen, 1975), but for all intents and purposes, the rate is zero if

the DO exceeds 1.0 mg/L. Although other inorganic anions are known to serve as electron acceptors in the absence of dissolved oxygen, in a wastewater which has undergone nitrification, the concentration of nitrate ions is likely to be present in much greater concentrations than phosphate or sulfate ions; thus, under conditions of low dissolved oxygen, denitrification can be expected to occur (Barnes and Bliss, 1983). The rate of denitrification depends primarily upon the nature and concentration of the organic matter present. Thus, it is commonly accepted that denitrification is zero-order with respect to nitrate concentration down to very low levels (approximately 1 mg/L) (Barnes and Bliss, 1983 citing various sources; Sedlak, 1991).

Consortia dynamics. The role of consortia or properties of flocs cannot be stressed enough in the activated sludge process, especially when treating industrial wastewaters. A consortium of various species may lead to difficulty in achieving nitrification, but more likely, toxic or inhibitory effects will be buffered, or possibly species not known for nitrification will be induced to nitrification.

The proportion of a given microbial species present in a mixed culture will depend upon the relative abundance and type of electron donor (generally an organic substance), electron acceptor (oxygen, nitrate, phosphate, etc.), and amount of energy gained by using the particular electron acceptor (Barnes and Bliss, 1983). Generally, nitrifiers are only a small percent of the biomass, which is typically mostly heterotrophic (Barnes and Bliss, 1983). This should come as no surprise, as nitrifier cell yield is lower than heterotrophic cell yield (Metcalf and Eddy, 1991b; Sedlak, 1991). Because of this fact, the sludge age must be carefully controlled. If the biomass growth rate (and therefore the wastage rate) is greater than the nitrifier growth rate, the percentage of nitrifiers in the biomass will taper off until nitrification is lost (i.e., the nitrifiers are washed out). The net biomass growth rate can be reduced by reducing the organic loading rate per unit of biomass however (i.e., by lowering the F:M ratio). Optimizing the DO and pH for nitrifiers is relatively simple, but generally more costly than carbonaceous removal prior to nitrification (Barnes and Bliss, 1983).

Flocs and fixed films are obviously going to create microenvironments which will not necessarily have the same temperature, pH, DO, or even substrate type and availability as the bulk solution. This would help explain why nitrifiers have been found to grow in environments which lab studies suggest are completely unsuitable (for example, at low pH). Interaction with other organisms or modification of the local environment for protection may be what causes such phenomena (Prosser, 1989). It is well known that flocs and films do not distribute oxygen evenly and that interactions which are more complex than pure culture interactions are occurring. As a result, the saturation constants for oxygen appear to be higher for the nitrifiers in activated sludge than in pure cultures. Furthermore, the constants tend to increase with temperature suggesting that good oxygenation is crucial in this study, where activated sludge is used at high temperatures. Variations in reported saturation values are likely to be due to oxygen diffusion limitations in larger floc particles, differences in dynamic and steady-state techniques used, and double substrate limitations (oxygen plus ammonia or nitrite) (Barnes and Bliss, 1983 citing Stenstrom and Poduska, 1980).

Such different microenvironments can either inhibit or induce nitrification, depending on an array of factors. In at least one case, however, it was shown that nitrification was dependent upon a consortium, as an *Arthrobacter* sp. and *Corynebacterium* sp. were unable to nitrify when grown separately, but nitrification did occur when the two species were grown in co-culture (Prosser, 1989 citing Rho, 1986). Although no investigations were made to determine what species were responsible for nitrification during this study, it must be understood that complex consortia might have played a critical role for sustained nitrification in the wastewater tested, especially given the conditions such as: a complex industrial wastewater (with the potential for occasional spikes of inhibitory compounds), high ammonia (including possibly high free ammonia) concentrations, and high temperatures.

In a study by Hockenbury (1977) cited in Neufeld *et al.* (1980), heterotrophic activity in domestic and industrial wastes did not appear to inhibit nitrifiers, and it may have enhanced nitrification rates. The presence of heterotrophic organisms has been shown to actually slightly stimulate nitrification because of the release of growth factors by the heterotrophs (Barnes and Bliss, 1983 citing Hockenbury *et al.*, 1977). Consortia involved in activated sludge processes may help to dampen inhibitory effects as well.

Single sludge vs. multiple sludge systems. A separate sludge system is one where each sludge has its own settling and recycling (Barnes and Bliss, 1983). In other words, the anoxic reactor has its own anoxic clarifier, and the aerobic reactor has its own aerobic clarifier, and so on. Thus, for a process which includes nitrification, predominantly nitrifiers will grow and remain in the nitrifying reactor, while predominantly heterotrophs will grow and remain in reactors where carbonaceous removal occurs.

In a single-sludge system, only one clarifier is used for the entire treatment process. In the single-sludge systems, the mixed liquor contains both heterotrophs and autotrophs. The autotrophs will grow only in the aerobic basin, while the heterotrophs will grow in the aerobic, anoxic, or anaerobic basins as long as metabolizable organic matter is present (Sedlak, 1991). While this may result in a lower percent of nitrifying biomass, there are many

economic benefits to using a single-sludge process instead of a separate-sludge process. For instance, only one clarification step is used (saving on size and pumping costs), an external carbon source is less likely to be required, pH control chemical requirements are lower, and oxygen requirements are lower as well (Sedlak, 1991). Additionally, the problem of poor-settling nitrifiers is greatly reduced, and a more stable sludge resistant to shock loads is likely (Barnes and Bliss, 1983; Prosser, 1989). Denitrification rates in single-sludge systems are approximately half the rates of separate-sludge systems (Metcalf and Eddy, 1991b), but since nitrification is generally the rate limiting step, such a fact will likely be of little concern, although it should be kept in mind. Because of the many benefits, single sludge processes were selected for nitrifying and denitrifying this wastewater.

Physical/chemical processes. Physical and chemical processes have been used for nitrogen removal, such as air stripping, breakpoint chlorination, and ion exchange. Only very few treatment facilities use such techniques, however, due to cost, inconsistent performance, and operation and maintenance problems (Metcalf and Eddy, 1991b; Sedlak, 1991). Biological nitrification-denitrification is generally best because of: high potential removal efficiency, high process stability and reliability, relatively easy process control, low land area requirements, and moderate costs, but physical/chemical means of removal may be viable options in certain circumstances (Metcalf and Eddy, 1991b; Sedlak, 1991).

Temperature Effects

Temperature affects wastewater treatment in many ways, both directly and indirectly affecting the biomass. There is an apparent optimal temperature or range of temperatures for any organism, and the optimal temperature for growth will not necessarily be the same as the optimal temperature for substrate oxidation/reduction (Charley *et al.*, 1980). What defines the optimal temperature is not necessarily dependent upon the organism of interest itself, but a wide array of factors which are affected by temperature, such as electron donor or acceptor availability, the chemical form of the substrate at given temperatures and pHs, sensitivities to inhibitors at different concentrations, and efficiency of enzymes involved.

Numerous studies have yielded various ranges for optimal temperatures. There are many factors involved in such studies, which will obviously affect the results. For instance, pure culture versus activated sludge studies, the biomass concentration, the substrate concentration, the DO concentration, sludge age, the pH, short term versus long term effects, the growth conditions prior to testing, and the difference between growth and test conditions. For the most part, however, nitrifier growth and activity tends to increase with temperature up to a threshold value, as is the case for any microorganism.

Temperature effects on nitrifier growth. Temperature has a strong effect on the growth of nitrifiers, but quantifying the effect has been difficult (Metcalf and Eddy, 1991a). Part of the difficulty in quantifying temperature effects is the fact that the optimum temperature and pH are not fixed, but vary as the total ammonia-N concentration changes (Quinlan, 1984). The trend with optimal temperature for growth appears to be that the growth rate increases as temperature increases up to approximately 35^{o}C with an overall range for growth between 4^{o}C and 45 to 50^{o}C (Antoniou *et al.*, 1990 citing Loveless and Painter, 1968; Barnes and Bliss, 1983 citing Painter, 1970 and Focht and Chang, 1975; Churchwell *et al.*, 1980 citing Nelson, 1931 and Deppe and Engel, 1960). This upper limit of 45 to 50^{o}C is specifically the problem associated with the wastewater tested in this study, as its summer temperatures are exactly in that range.

Temperature effects on nitrifier activity. The optimal temperature for nitrifier activity has been reported as low as 15^{o}C (Charley *et al.*, 1980), but more typically appears to increase with increasing temperature up to approximately 30^{o}C, slowing down as the temperature increases beyond that (Fdz-Polanco *et al.*, 1994; Groeneweg *et al.*, 1994; Wild *et al.*, 1971). Especially at lower temperatures, nitrification appears to be more severely affected than denitrification by temperature changes (Azevedo *et al.*, 1995). Although temperature affects many variables, among them, the concentration of free ammonia and free nitrous acid, one study revealed that temperature had a much greater effect on the rate of ammonia oxidation than can be accounted for by the temperature dependent shift in free ammonia availability (Groeneweg *et al.*, 1994). Obviously, other factors, such as concentration and activity of the organisms involved must be taken into account when considering the relationship between temperature and nitrification rates (Fdz-Polanco *et al.*, 1994).

Possible means of high temperature nitrification. To date, there has been no success isolating autotrophic nitrifiers from thermophilic environments, and it is believed that autotrophic nitrifiers are unable to survive in 50-60^{o}C operating temperatures, although such studies have dealt with composts and not aqueous environments such as activated sludge processes (Pel *et al.*, 1997 citing various sources). In one study, nitrification was achieved at 53^{o}C in a suspended growth culture. Failure to successfully enrich for autotrophic nitrifiers, combined with the decline in nitrification upon removal of methane from the feed led the authors to conclude that the observed ammonia oxidation was attributable to thermophilic methanotrophs (Pel *et al.*, 1997).

Acceptable temperatures for denitrification. As with nitrification, temperature affects both the activity (the nitrate removal rate) and the growth rate of the organisms involved in denitrification (Metcalf and Eddy, 1991a). Temperature dependence for denitrification is similar to other biological processes, occurring between 0°C and 50°C, with optimum reaction rates occurring at 35-50°C (Barnes and Bliss, 1983). The higher threshold makes it apparent that high temperatures is a greater concern to the nitrification process than the denitrification process.

Other environmental factors affecting nitrification and denitrification

pH effects. The pH will obviously affect several different factors, which in turn will affect the growth and activity of organisms involved. Because this study focused on the feasibility of nitrification of a particular wastewater, and not pH effects on nitrification, discussion will be reserved to understanding what acceptable ranges of pH are for nitrification, with the understanding that the pH is, in fact, tied in to a lot of factors important to successful nitrification. In brief, the pH is known to affect enzymes of interest, affinity for the substrate, substrate availability, effects of inhibitory compounds, and substrate or product inhibition (Antoniou *et al.*, 1990; Groeneweg *et al.*, 1994; Prosser, 1989).

Most nitrifiers have an optimum pH at approximately 7.5-8.0 and will grow within a pH range of approximately 2 pH units (Painter and Loveless, 1983 citing Loveless and Painter, 1968; Prosser, 1989). Growth for *Nitrosomonas europaea* occurs in the pH range of 5.8 to 8.5, suggesting there is a diversity of strains within the species. Optimal pH ranges are similar for mixed cultures and pure cultures (Prosser, 1989), thus, similar optimal pH ranges can be expected for activated sludge processes.

As with temperature, the optimum pH for growth will not necessarily be the same as the optimum pH for activity (Barnes and Bliss, 1983 citing Loveless and Painter, 1968 and Painter, 1970). For nitrification activity, the optimum pH range is commonly accepted to be 7.5 to 8.5 (Metcalf and Eddy, 1991a; Painter and Loveless, 1983 citing Downing *et al.*, 1964; Sedlak, 1991), with little to no nitrification occurring below approximately pH 6 to 6.5 or above 10 (Groeneweg et al, 1994; Painter and Loveless, 1983; Painter and Loveless, 1983 citing Downing et al, 1964). More specifically, the optimum pH range, defined as oxidation rates being at least 90% of the maximum rate, for *Nitrosomonas* activity tends to fall within the range of 6.7 to 9.2 while the optimum range for *Nitrobacter* tends to be approximately 8.0 to 9.5 (Churchwell *et al.*, 1980 citing several references; Wild *et al.*, 1971 citing several references).

Acclimation to pH values outside the accepted ranges has been shown possible for nitrifiers. Within reason, acclimation should be expected if changes are made gradually enough, and it must further be kept in mind that the majority of studies for determining optimal pH ranges dealt only with short-term effects on the oxidation of substrate nitrogen with cultures of pre-grown cells, ignoring the ability of nitrifiers to adapt to environmental changes (Barnes and Bliss, 1983). Acclimation to pH values as high as 11.2 (Groeneweg *et al.*, 1994 citing Focht and Verstraete, 1977) and as low as 3.4 (Pennington and Ellis, 1993) by autotrophic nitrifiers has been achieved. Because the wastewaters in this study tended to have relatively neutral pH values, acclimation to pH extremes was not an issue as much as maintaining the pH in the optimal, neutral range.

As with nitrification, the optimal pH for denitrification is neutral to slightly alkaline, with optimum activity occurring roughly in the range of 6 to 8 but generally in the range of 7 to 8 (Azevedo *et al.*, 1995 citing U.S. EPA 1975; Barnes and Bliss citing Moore and Schroeder, 1971; Metcalf and Eddy, 1991a). Maintaining the pH is generally enhanced using biological denitrification, as alkalinity is produced by the reduction of nitrate to nitrogen gas (Metcalf and Eddy, 1991a). It is possible, however, for the generation of alkalinity to raise the localized pH to inhibitory levels, especially in fixed film processes (Barnes and Bliss, 1983 citing Riemer and Harremoes, 1978 and Arvin and Kristensen, 1982). Such a problem did not arise in this study, however.

Aeration effects on nitrification and denitrification. Dissolved oxygen is an "absolute requirement for growth of both *Nitrosomonas* and *Nitrobacter*" (Barnes and Bliss, 1983). While *Nitrosomonas* and *Nitrobacter* are obligate aerobes for growth, prolonged lack of O_2 is *not* lethal (Barnes and Bliss, 1983 citing Painter, 1970). This helps explain how single-sludge systems are possible. A critical value for DO, below which nitrification does not occur, has been reported at 0.2 mg/L for pure cultures of both *Nitrosomonas* and *Nitrobacter* (Barnes and Bliss, 1983 citing Schoberl and Engel, 1964) and activated sludge (Barnes and Bliss, 1983 citing Downing *et al.*, 1964). In general, nitrite oxidizers appear to be more sensitive to low DO concentrations than ammonia oxidizers (Prosser, 1989 citing Helder and deVries, 1983). While a DO of at least 1 mg/L appears to be a requirement to prevent oxygen from becoming the limiting nutrient in nitrification (Metcalf and Eddy, 1991a), 2 mg/L of dissolved oxygen is typically considered the cutoff value to fully ensure nitrification is not oxygen limited (Wild *et al.*, 1971). Saturation constants for oxygen for pure cultures of ammonia and nitrite oxidizers are in the range of 0.25 to 2.5 mg/L of DO (Prosser, 1989 citing Painter, 1986). Similar oxygen saturation constant values are reported for mixed

cultures. Saturation values (K_s) for nitrifiers are generally higher for nitrifiers than for heterotrophs, meaning the nitrifiers are likely to get out-competed by the heterotrophs at low DO concentrations (Prosser, 1989).

Note, the dissolved oxygen in the bulk solution is not going to be the same as the DO within flocs or films (Sedlak, 1991). A dissolved oxygen gradient has been shown to exist in flocs (suggesting that nitrification can be occurring on the outside of the floc while denitrification can be occurring within the floc) (Wood *et al.*, 1981). Such a phenomenon will depend upon sludge age, floc size or film thickness, type of aeration, wastewater makeup, and many other environmental factors. For this study, the aerobic DO was simply monitored and maintained above 2.0 mg/L and regarded as adequate if above 2.0 mg/L. Optimization of DO was to be determined at a later stage during pilot scale testing.

Low to zero DO is required for denitrification to occur, with optimum results occurring at zero DO (Barnes and Bliss, 1983; Metcalf and Eddy 1991a). It is believed that if dissolved oxygen is present, the enzyme system needed for denitrification will be suppressed (Metcalf and Eddy, 1991a). A denitrification rate of 0.006 mgNO3-N/mgVSS/day has been reported under aerobic conditions (Sedlak, 1991 citing Christensen, 1975), and it has been shown that denitrification can occur in the presence of molecular oxygen under acidic pH conditions (Sedlak, 1991), but for all intents and purposes, the rate is zero if the DO exceeds 1.0 mg/L.

Sludge age (SRT effect on both nitrification and denitrification). Sludges with a long sludge age can be expected to have a smaller fraction of viable biomass than sludges with a short sludge age (Barnes and Bliss, 1983). However, a longer sludge age is generally required in order to have a sustainable nitrifier population (Metcalf and Eddy, 1991b). Although nitrifiers are generally regarded as the rate limiters to a nitrification/denitrification process, a careful balance must be achieved, since several studies have shown that rates of denitrification are lower at higher SRTs (Barnes and Bliss, 1983 citing various sources). Changing from a conventional sludge age of 3 days to an extended aeration sludge age of over 15 days has been shown to produce a decrease in the rate of denitrification by a factor of 2 to 3 (Barnes and Bliss, 1983). It was determined in this study that longer sludge ages (12 to 24 days) tended to be required for complete nitrification of the streams tested at reasonable rates, and that denitrification rates were adequate even at the higher sludge ages.

Inhibitory trends. Nitrifying autotrophs are commonly accepted as being more sensitive to toxins than the wide range of heterotrophs responsible for denitrification. Thus, little inhibition of denitrification in actual treatment systems is expected (Barnes and Bliss, 1983). The oxidation of ammonia to nitrite by *Nitrosomonas* is considered to be the more sensitive of the two steps in nitrification, and *Nitrosomonas* is generally more susceptible to inhibition from other compounds than *Nitrobacter* (Barnes and Bliss, 1983; Blum and Speece, 1991). Most inhibitors stop ammonia oxidation, but they often inhibit nitrite oxidation as well in high enough concentrations (Prosser, 1989). Most compounds inhibitory toward nitrification inhibit ammonia oxidizers, however, with only a small percentage of compounds tested being more toxic to *Nitrobacter* than *Nitrosomonas* (Tomlinson *et al.*, 1966).

Excessive COD/BOD loading tends to cause inhibition of nitrification. Some compounds may be directly inhibitory in high enough concentrations even though they are not necessarily specific inhibitors. For instance, methanol toxicity to ammonia oxidation has been reported (Azevedo *et al.*, 1995 citing Hooper and Terry, 1973). More commonly, however, nitrification inhibition by the presence of organic matter can be attributed to DO depletion caused by heterotrophic organisms utilizing the organics present (Barnes and Bliss, 1983 citing various sources). Although BOD levels up to 40-50 mg/L can be tolerated in nitrifying reactors (Wild *et al.*, 1971), it has been shown that BOD_5 levels over 40 mg/L can lead to as little as 50% nitrification. Such a phenomenon was postulated to be due to low levels of DO due to outcompetition by heterotrophs, even though DO meter values indicated sufficient levels of oxygen were present in the bulk solution (Azevedo *et al.*, 1995). Such problems may be indicative of consortia dynamics and the physics of flocs or biofilms, where microenvironments will develop. The outcompetition for oxygen by heterotrophs is one key reason that aerobic nitrifying reactors are often placed last in a sequence, in order to minimize the amount of organic substrate present.

Acclimation. Long term effects of inhibitors are often different from the immediate effects observed in activated sludge, with one of the main reasons for this fact being that sludges can often acclimate to inhibitory conditions and compounds. For instance, in long term studies, evidence for acclimation to thiourea concentrations of 0.76 mg/L was observed; a level which would have been completely inhibitory to unadapted *Nitrosomonas* species (Tomlinson *et al.*, 1966). In their study, Tomlinson *et al.* determined that short term effects of inhibitors were not good estimates of long term effects of a continuous activated sludge operating under steady conditions. One reason for a difference in short term and long term effects is that, for instance, other organisms in activated sludge can develop the ability to decompose an inhibitor (Tomlinson *et al.*, 1966). In the case of acclimation in pure culture studies, Tomlinson *et al.* (1966) postulated that either there are strains of *Nitrosomonas* which are resistant to certain inhibitors and are gradually selected out under inhibitory conditions, or that normally susceptible strains can develop resistance. Occasionally, long term effects can actually be more inhibitory than short term effects. This is

perhaps the result of compounds producing no measurable effects during the duration of short tests, but which do suppress growth or yield to a measurable degree over longer periods (Tomlinson *et al.*, 1966). What is important about either acclimation or greater inhibition resulting from prolonged exposure to compounds is the understanding that long term studies, and not merely batch tests are important to determine the feasibility of nitrification (and/or denitrification) of a wastewater, especially one as complex as that of HRWTF.

Substrate/product toxicity. Both nitrite and ammonia concentrations were examined and evaluated in this study to determine if free ammonia (FA) or free nitrous acid (FNA) toxicity were likely to be occurring in the treatment trains tested. Because nitrification never appeared to be inhibited for any of the treatment trains tested, discussion of substrate and product toxicity has been restricted to briefly defining it and pointing out that it is a concern when testing the feasibility of nitrification of a previously non-nitrified wastewater.

Both groups of nitrifiers are inhibited by high concentrations of their own substrates and have little energy to spare for high affinity (i.e. low substrate concentration) uptake systems (Prosser, 1989). Furthermore, both groups of nitrifiers are much more susceptible to the others' substrate (Anthonisen *et al.*, 1976; Barnes and Bliss, 1983). Thus, a somewhat delicate and precise balance must be maintained for successful nitrification. The form of the products and substrates also plays a major role in the inhibition of nitrifiers. Factors such as acclimation to FA and FNA, temperature, pH, and the number of active nitrifiers cannot be ignored when considering inhibitory concentrations of FA and FNA however (Anthonisen *et al.*, 1976), as all such factors have effects on each other and on the organisms of interest. Important to maintaining a nitrification process in a real wastewater treatment facility is having a culture resilient enough to recover from occasional spikes. Studies have shown that FA and FNA inhibition is not permanent if conditions are alleviated soon enough, and that nitrification will resume (Anthonisen *et al.*, 1976; Turk and Mavinic, 1989; Churchwell *et al.*, 1980).

III. MATERIALS AND METHODS

System Hardware

Four different systems were tested in this study using CSTRs in series. The purpose, design, sampling, and analysis techniques of each are described below.

Where indicated, phosphorus addition was used. All phosphorus added was pumped from a 20 liter plastic carboy, which contained a 1:1000 dilution of 85 percent phosphoric acid (H_3PO_4) and distilled water. In a similar fashion, where indicated, alkalinity addition was used. Alkalinity was supplied in the form of sodium bicarbonate ($NaHCO_3$) dissolved in distilled water in a 20 liter plastic carboy. Because of the various tubing diameters and alkalinity requirements, the concentration of the stock solution was not kept constant throughout the experiment. Methanol was supplied in a similar fashion, diluting HPLC grade methanol into a 20 L carboy from which the stock solution was pumped directly into the anoxic reactors. The stock solutions were generally made up in the range of 2 to 6 mL of methanol per liter of distilled water.

All recirculation, recycle, and supplemental chemicals were supplied to their respective reactors via rubber tubing and Cole Parmer "Economy Drive" adjustable speed pumps. All mixers used were Lightnin G2U05R mixers. All reactors in series were set on platforms of different heights such that the mixed liquor would flow via gravity and the half inch Tygon tubing connecting them from the exit port at the top of the mixed liquor in one reactor to inlet port located approximately one inch from the bottom of the next reactor.

Secondary effluent treatment train. The secondary effluent treatment train was designed to determine if the present secondary (i.e., final) effluent of the Hopewell Regional Wastewater Treatment Facility (HRWTF) could successfully be nitrified and denitrified, and to determine the maximum rate to which the system could be pushed in order to estimate basin size requirements. The secondary treatment system consisted of an aerobic/anoxic treatment scheme for the final effluent of HRWTF which already had the majority of organics removed. A third reactor of the system was a small aerobic reactor for conditioning of the sludge leaving the anoxic reactor, in order to strip nitrogen bubbles from the flocs and to ensure any remaining organics present in the anoxic reactor were utilized prior to settling, to avoid denitrification and floating sludge in the clarifier. A schematic diagram of the treatment train is provided in Figure 1.

To best simulate treating the secondary effluent at the plant, the secondary (final) effluent of HRWTF was continuously pumped into the bottom of an open plastic 55 gallon drum which had an overflow pipe located near the top of the drum. The intake screen for the system was located approximately midway down in the drum and was pumped to the first (aerobic) reactor via a peristaltic pump.

All reactors were connected in series by half-inch Tygon tubing at varying heights such that gravity would feed the effluent from one reactor to the next reactor in the series. All internal recirculation and chemical addition was performed via peristaltic pumps (Cole Parmer), including the return activated sludge (RAS) from the final clarifier of the system and the alkalinity, phosphorus, and methanol addition.

The first (aerobic) reactor used up front was constructed of a rectangular plastic tub divided into four CSTRs in series by three plastic baffles (to create a serpentine flow), with 6 inch aeration stones set in the bottom of each reactor to provide both oxygen and mixing. The level of the outlet was set such that the mixed liquor volume was 18 liters. Air was used for aeration, and the nitrate-rich effluent flowed by gravity to the anoxic reactor. At the front of the aerobic reactor, three stainless steel tubes were fastened just above the surface of the mixed liquor for continuous pumping of phosphorus, alkalinity, and RAS addition. Once denitrification was established and stabilized, alkalinity addition was stopped, as it was determined that the pH in the aerobic reactor of the system was sufficiently high, as was the alkalinity and pH of the final effluent, without supplemental alkalinity.

The second (anoxic) reactor of the series was constructed of 6 inch PVC pipe, with an inlet located at the bottom of the cylinder, and an outlet fitting located at a height such that the mixed liquor volume was 8.5 liters. A sampling port was located at mid depth of the mixed liquor, and a port to allow the escape of gases generated was located in the side of the reactor above the mixed liquor. On top of the reactor was a cap into which a Lightnin G2U05R mixer was mounted as well as one 1/4 inch stainless steel tube. The stainless steel tube was inserted approximately three fourths of the way down into the mixed liquor for methanol addition. The methanol was continuously pumped into the reactor from the carboy of stock solution via a peristaltic pump and rubber tubing.

The third reactor was a small aerobic reactor constructed of a one gallon plastic bucket with an aeration stone for mixing and aeration (mixed liquor volume of 4.1 liters). Air was used as the oxygen supply in this aerobic reactor as well. From this reactor, the effluent flowed into the final clarifier.

The final clarifier of the system was a 15 gallon plastic drum with a 60^o slope in the bottom portion to enhance settling and compaction for the RAS which was pumped from the bottom. The outlet fitting was at the

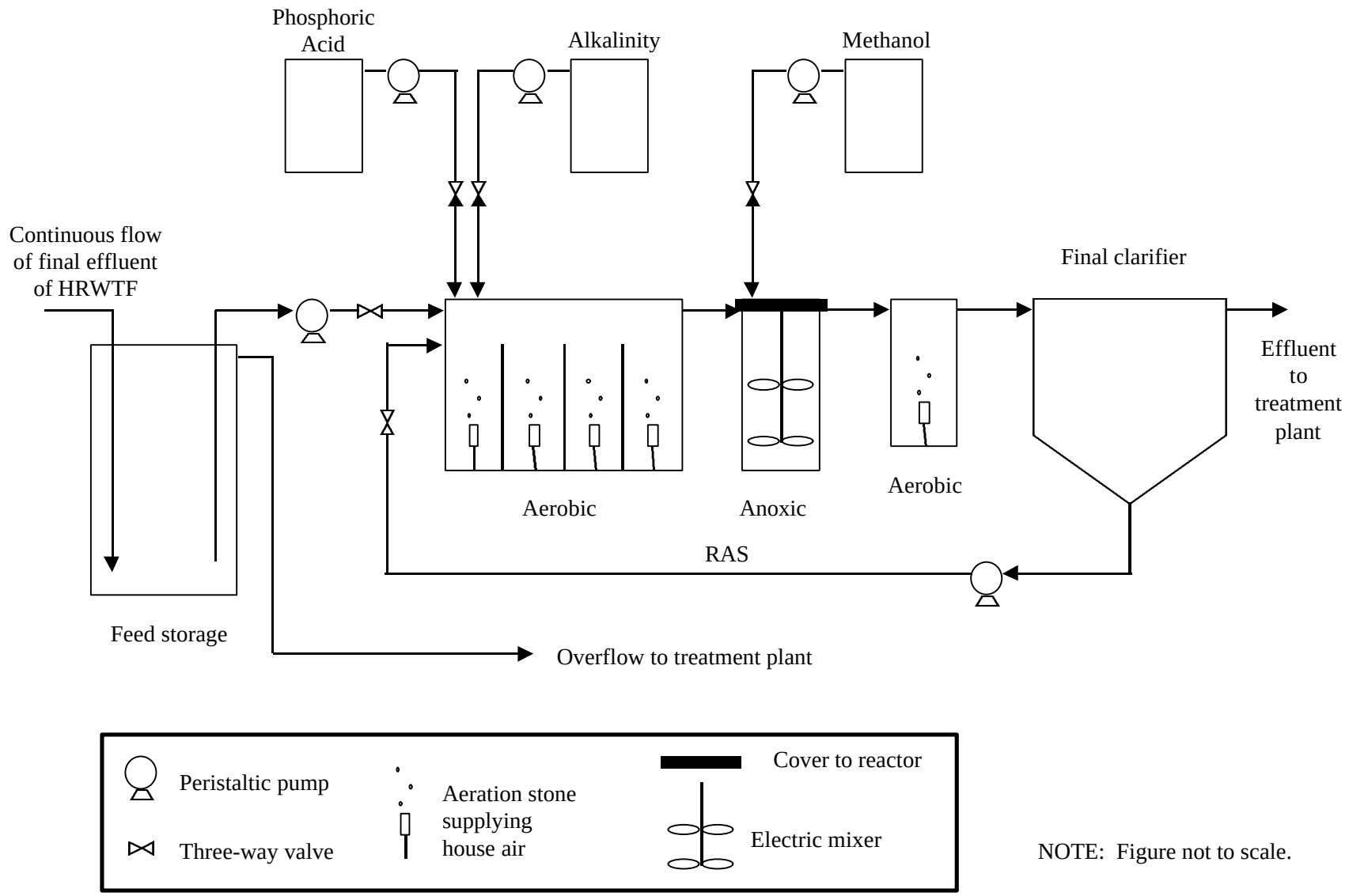

Figure 1. Schematic diagram of the aerobic/anoxic treatment of the secondary effluent of HRWTF.

opposite side of the clarifier as the inlet fitting and was set at a height such that the total volume in the clarifier was approximately 10.5 gallons. From the outlet, the effluent flowed from half-inch Tygon tubing down a drain which returned the effluent to the head of the HRWTF treatment plant. To prevent channeling through the sludge blanket and prevent the blanket from sticking to the sides of the clarifier, a 1 rpm motor with squeegee blades ran continuously in the clarifier.

A high temperature experiment was performed on the system at the end of the study in order to determine if the secondary effluent of HRWTF could be nitrified and denitrified at existing high temperatures. To accomplish this, the same system was used, but the reactors were insulated with fiberglass duct insulation, and fish tank heaters were inserted into the aerobic reactors. Heaters were not inserted into the anoxic reactor because the mixer blades would have broken them. As a result, the anoxic reactor tended to be approximately 5^{o}C cooler than the aerobic reactors. An industrial sized water heater with a thermostat control was inserted into the feed drum for additional temperature increase and control. The heaters were adjusted to obtain a temperature of approximately 45^{o}C, and the temperature rose from approximately 28^{o}C to 38^{o}C within a 24 hour span. This was somewhat faster than the existing process, where the temperature rises to a similar condition over the span of a few days, but it was believed to be a reasonable approximation of what occurs at HRWTF every spring.

Primary effluent treatment train. The primary effluent treatment train was set up to determine if the present primary effluent of HRWTF could successfully be nitrified and denitrified at ambient temperatures and to determine the maximum rates for estimating basin size requirements. The design selected to treat the primary effluent was an anaerobic/anoxic/oxic (A2/O) process. Such a configuration is commonly used for biological nutrient removal (BNR), which entails the removal of both nitrogen and phosphorus, but this stream is actually phosphorus deficient, and the process was selected primarily to offer better buffering against possible toxicity via the anaerobic reactor up front, although theoretically only an anoxic/oxic configuration should have been required. A schematic diagram of the treatment train is provided in Figure 2.

As with the secondary effluent treatment system, the primary effluent from the plant was continuously pumped into the bottom of an open plastic 55 gallon drum which had an overflow pipe located near the top of the drum. The intake screen for the system was located approximately midway down in the drum, and the influent was pumped to the first (anaerobic) reactor via a peristaltic pump.

The first (anaerobic) reactor of the series was constructed of a 5 inch plexiglass cylinder with an inlet located at the bottom of the cylinder, and an outlet fitting located at a height such that the mixed liquor volume was 4 liters. Similar to the anoxic reactor of the secondary effluent treatment train, a sampling port was located at mid depth, and a port to allow the escape of gases generated was located in the side of the reactor above the mixed liquor. On top of the reactor was a cap into which a Lightnin G2U05R mixer was mounted, as well as three 1/4 inch stainless steel tubes. The stainless steel tubes were inserted approximately three fourths of the way down into the mixed liquor and were attached to appropriate pumps for continuous RAS addition, phosphoric acid, and sodium bicarbonate (alkalinity) addition.

The second (anoxic) reactor in the series was constructed in a similar fashion, but with 6 inch PVC pipe for the reactor, and a volume of 8.5 liters. Only one stainless steel inlet was inserted in the cap, however, for mixed liquor recirculation from the aerobic (third) reactor as a nitrate supply. A second inlet was added later for occasional continuous methanol addition.

The third (aerobic) reactor was constructed of a covered rectangular plastic tub made by Rubbermaid, with three plastic baffles installed to create a serpentine flow and, in essence, four CSTRs in series. A 10 inch aeration stone was attached to the bottom of each of the four reactors, and pure oxygen was supplied to simulate the high purity oxygen (UNOX) process used at HRWTF for secondary treatment. The aeration supplied both oxygen and mixing for the reactor. The level of the outlet (from which the mixed liquor flowed into the final clarifier via half inch Tygon tubing) was set for an activated sludge volume of 27.4 liters. One-quarter inch stainless steel tubing was inserted through the cover of the reactor at the end of the aerobic reactor for the nitrate-rich mixed liquor recirculation from the aerobic rector back to the anoxic reactor.

The final clarifier used consisted of the exact same setup as the clarifier described above for the secondary effluent system, with a volume of approximately 10.5 gallons.

Heated system. A third system was set up to determine the quality of high temperature performance. The system was initially set up to determine the feasibility of high temperature COD/BOD removal of the pulp mill wastewater, but it was soon determined that such an option was not practical because the wide fluctuations in the COD/BOD strength of the pulp mill wastewater led to unstable sludge characteristics and effluent quality. It had been observed that nitrification was occurring at high temperatures, and thus, the study was soon devoted to determining the

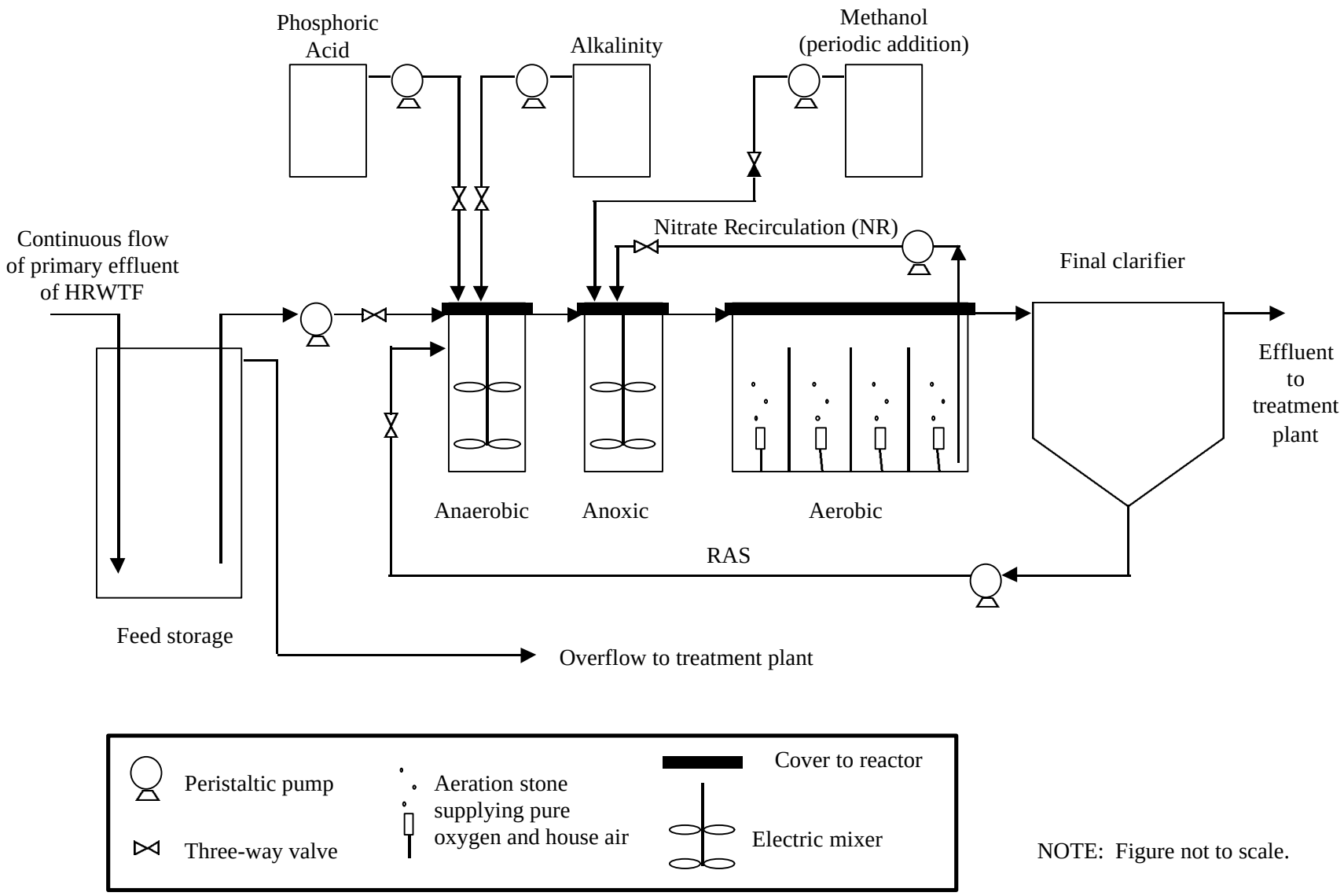

Figure 2. Schematic diagram of the A2/O treatment of the primary effluent of HRWTF.

feasibility of nitrifying the primary effluent of HRWTF at high temperatures. A simple fully aerobic process was used (see Figure 3), which was later modified by adding an anoxic reactor in front of the aerobic reactor, creating a Modified Lutzack-Ettinger (MLE) process for nitrification and denitrification of the primary effluent of the HRWTF at existing high temperatures (see Figure 4). For both high temperature primary effluent studies, the influent screen was placed in the same drum as the system treating the primary effluent.

A covered rectangular plastic reactor was set up with two baffles to create three CSTRs in series in an overflow, underflow, overflow fashion, with a mixed liquor volume of 13.2 liters. The inlet port was located approximately one inch from the bottom of the reactor at the front end in order to avoid short circuiting. The reactor was placed in a hot water bath with thermostat control (Fisher Scientific Isotemp Model 210), such that the bottom half of the reactor was immersed in the water, but the upper half was not. Foam insulation was used on the outside of the reactor to minimize heat loss and temperature variations with depth. A 5 inch aeration stone was located in each reactor and hooked up to both air and high purity oxygen to simulate the high purity oxygen environment in the plant's UNOX process and to provide adequate mixing to keep the culture in suspension. Stainless steel tubing was used in a port at the front end of the reactor to allow for continuous RAS addition. The phosphoric acid solution was pumped directly into the aerobic reactor from the same carboy as was used for the primary and secondary effluent systems, but no alkalinity was added.

Later in the study, a 4 liter plexiglass anoxic reactor was placed up front of the process, constructed in the same fashion as the anoxic reactor of the other systems, to determine if denitrification was feasible as well. The reactor was covered with fiberglass insulation to keep temperatures elevated, and a port in the cap was made for the recirculation of the nitrate rich mixed liquor from the aerobic reactor (which was pumped out of the far end of the aerobic reactor). No methanol was added to the anoxic reactor in this system.

The same clarifier setup as described above was used, with a 10.5 gallon volume. The only modification to the design was covering the clarifier with a foam insulation lid and covering the clarifier with fiberglass insulation to minimize heat loss.

Acclimation to the high temperatures was achieved by starting the system up at ambient temperatures and increasing the temperature within the aerobic reactor by 2 or 3 degrees Celcius after having operated the system for one or two weeks at the preceding temperature. Temperatures were around 30^{o}C at startup, and temperatures were increased to approximately 45^{o}C after approximately two months of operation.

High ammonia side stream treatment train. The third system was set up to detoxify and nitrify/denitrify the side stream which was believed to contain MEKO in addition to high ammonia concentrations. A modified University of Cape Town (MUCT) process was selected, based upon findings of previous research with the waste stream (Smith, 1984; Lubkowitz, 1996) in order to degrade/detoxify the MEKO prior to the nitrifying reactor. Although it was later learned that the side stream did not contain any MEKO, the treatment train has been included in this study to demonstrate the feasibility and quality of nitrification and denitrification of a high ammonia (average ammonia concentration approximately 130 mg-N/L) wastewater, and to demonstrate that treatment of the side stream may be a viable option for achieving the nitrogen removal goals at HRWTF. Figure 5 provides a schematic diagram of the MUCT treatment train. The system was later switched to an A2/O configuration, just as the one depicted in Figure 2, by removing one of the anoxic reactors and switching the recirculation lines accordingly.

A 55 gallon plastic drum was used to store the solution used to feed the system. Instead of continuous feed as was used for the primary and secondary effluent systems, however, 35 gallon batches were grabbed from the industry of interest every one to two days and pumped into the drum. Because there were not enough pumps and pump heads to supply continuous alkalinity and phosphorus addition to the system, the phosphoric acid and bicarbonate solutions were added directly to the feed drum. When fresh batches of feed were grabbed, the phosphoric acid solution was added and mixed into the feed, and alkalinity titrations were then performed in order to determine the appropriate amount of alkalinity to add which would allow for 100% nitrification and denitrification with approximately 50 mg/L as $CaCO_3$ residual alkalinity in the final effluent.

The first three reactors were constructed in the same fashion as the anaerobic reactor from the primary effluent treatment system, with 5 inch plexiglass cylinders, each with a mixer and ports for the required stainless steel tubing for addition or drawing off of appropriate streams.

The first (anaerobic) reactor mixed liquor volume was set at 6 liters. One port with the 1/4 inch stainless steel tubing inserted approximately three fourths of the way into the depth of the mixed liquor was provided for the addition of the anoxic sludge from the second reactor.

The second (anoxic #1) reactor was constructed of the same 5 inch plexiglass with a mixed liquor volume of 4 liters. One port in the top allowed for the addition of RAS, while the second port in the top allowed for pumping out the anoxic sludge into the first (anaerobic) reactor of the sequence.

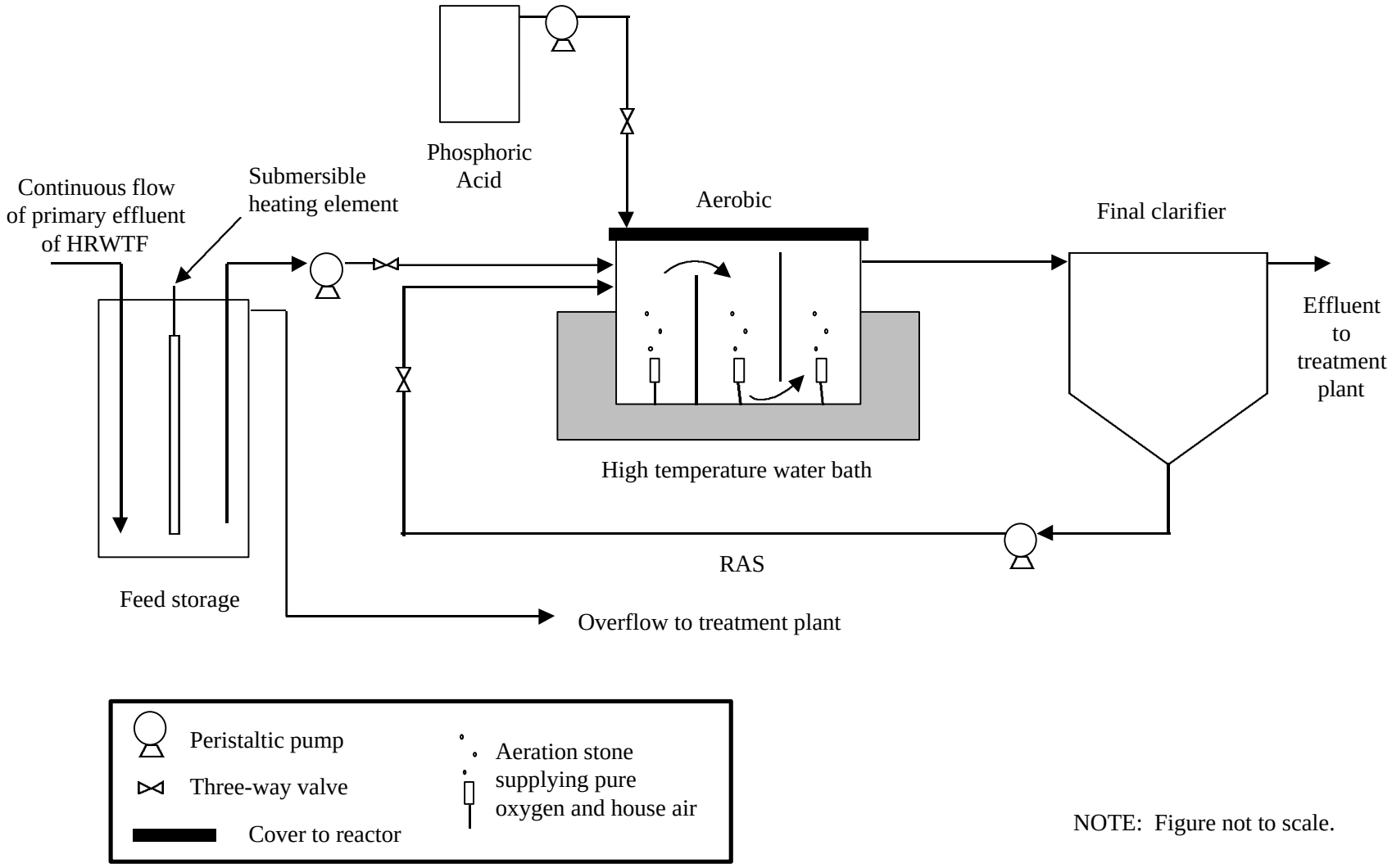

Figure 3. Schematic diagram of the fully aerobic treatment of the primary effluent of HRWTF at high temperatures.

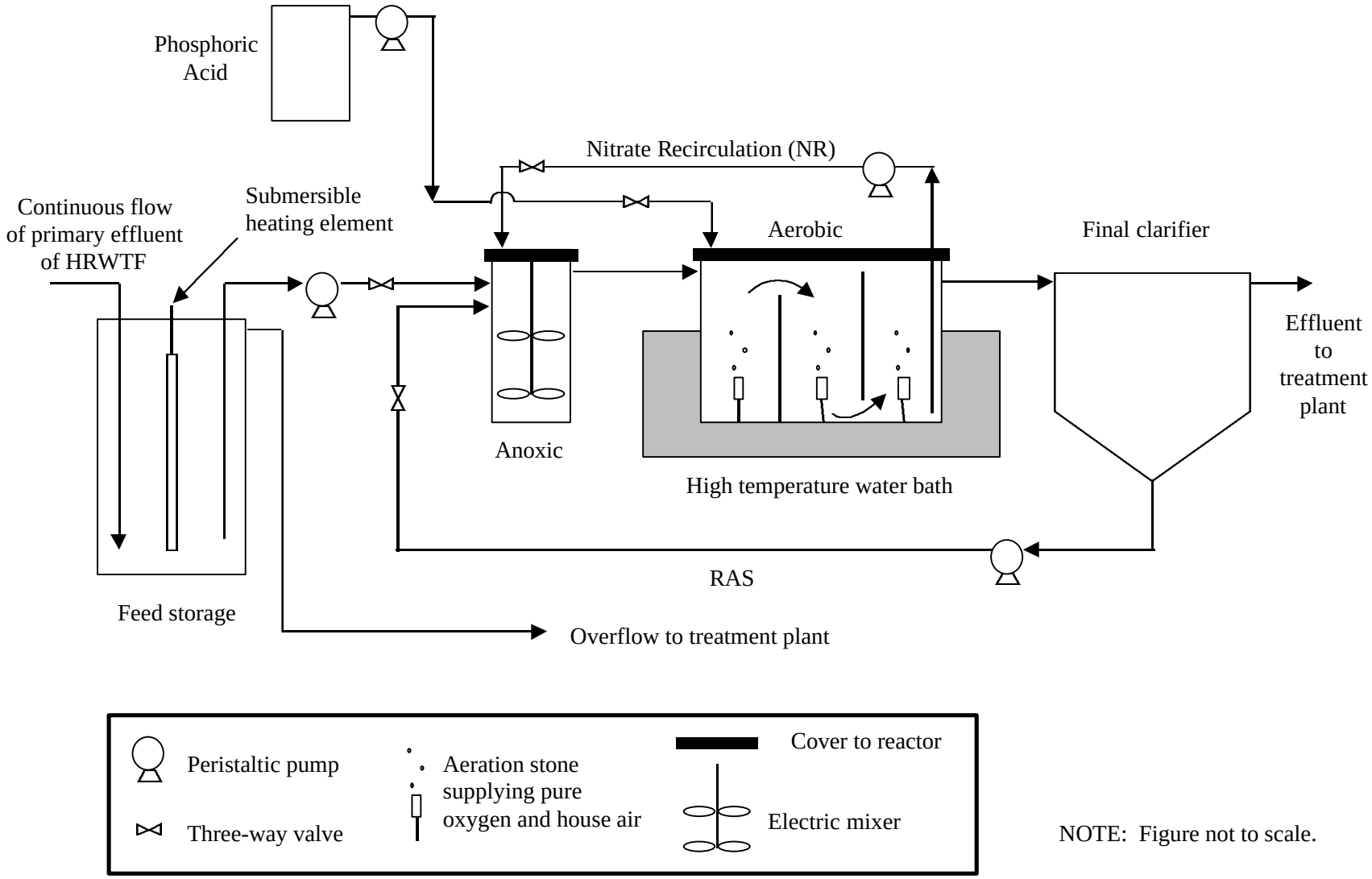

Figure 4. Schematic diagram of the MLE treatment of the primary effluent of HRWTF at high temperatures.

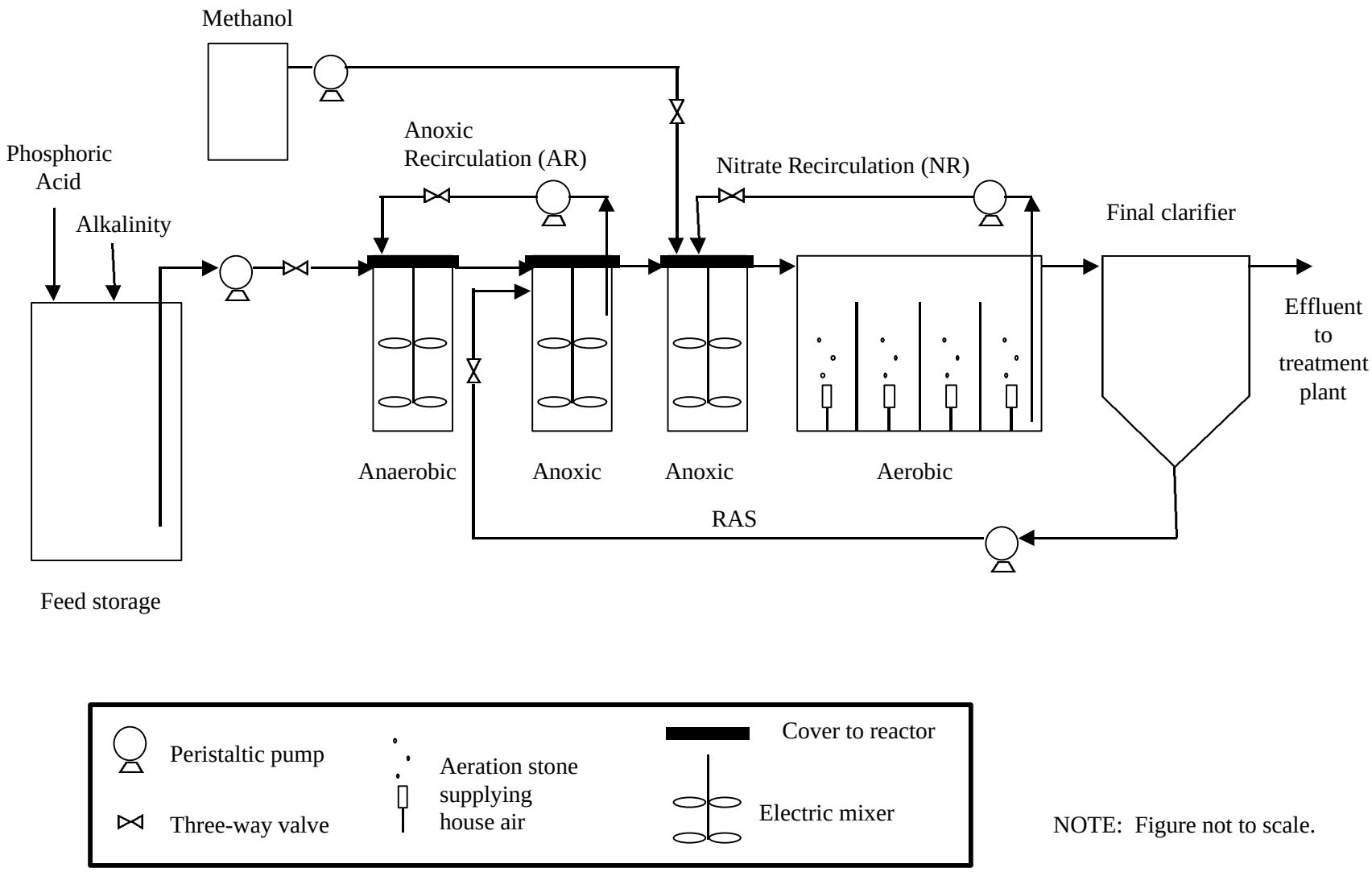

Figure 5. Schematic diagram of the MUCT treatment of the high ammonia side stream prior to mixing with other flows.

The third (anoxic #2) reactor was made the same as the second reactor, with a volume of 4 liters. One port was made for the nitrate recirculation, which was achieved by pumping the nitrate-rich sludge from the fourth (aerobic) reactor into the third reactor. A second port allowed for the continuous addition of methanol.

The fourth (aerobic) reactor was built in the same fashion as the aerobic reactor used to treat the secondary effluent, with a mixed liquor volume of 18 liters, 6 inch aeration stones, plastic baffles, plant air for oxygen, and stainless steel tubing at the end of the reactor for the nitrate recirculation to be pumped from.

The exact same clarifier setup as was used for the primary and secondary effluent treatment systems was used for this system, with the exception that the effluent port was set lower such that the clarifier volume was approximately 7.5 gallons.

Later in the study, one of the anoxic reactors was removed, and the system was reconfigured to an A2/O process (see Figure 2 for a schematic diagram).

Operating Conditions

Below are the target operating conditions which were maintained for the systems. The reasons for the changes made in operating conditions will be discussed in the "Results and Discussion" chapter of this thesis. Because of problems associated with maintaining so many pumps and reactors, occasional system upsets occurred, such as a spill, a seized pump, a disconnected tube, or low concentration RAS due to channeling in the sludge blanket. Thus, the conditions are referred to as *target* operating conditions which were maintained for the majority of the study. Data points which were outliers but could be correlated with an upset were discarded.

Secondary effluent treatment train.

Table 1. Target operating conditions for the aerobic/anoxic secondary effluent treatment train.

Parameter	Dates	Value
Seed:	5/19/97	8.6 L of Chesterfield RAS, 13.5 L of HRWTF MLSS, and 47.9 L of tap water.
Target SRT:	5/19/97-9/8/97	6
Target Total HRT:	5/19/97-7/14/97	7.3
	7/15/97-8/5/97	5
	8/6/97-9/8/97	3.8
RAS (% of Feed Flow Rate)	5/19/97-8/5/97	65
	8/6/97-9/8/97	55

Primary effluent treatment train.

Table 2. Target operating conditions for the A2/O primary effluent treatment train.

Parameter	Dates	Value
Seed:	5/19/97	21.5 L of Chesterfield RAS, 37.7 L of HRWTF MLSS, and 20.4 L of tap water.
Target SRT:	5/19/97-9/8/97	12 days
Target Total HRT:	5/19/97-7/2/97	10 hours
	7/3/97-7/28/97	7
	7/29/97-8/18/97	5.5
	8/19/97-9/8/97	7
RAS (% of Feed Flow Rate)	5/19/97-9/8/97	65%
MLR (% of Feed Flow Rate)	5/19/97-6/3/97	150%
	6/4/97-9/8/97	250%

Heated primary effluent treatment train.

Table 3. Target operating conditions for the fully aerobic and MLE high temperature primary effluent treatment train.

Parameter	Dates	Value
Seed:	5/20/97	40.1 L of HRWTF, and 13.0 L of tap water
Target SRT	5/20/97-6/17/97	6 days
	6/18/97-7/9/97	4 days
	7/10/97-8/20/97	8 days
	8/21/97-9/12/97	12 days
Target Total HRT	5/29/97-6/27/97	5.5 hours
	6/28/97-8/26/97	8 hours
	8/27/97-9/12/97	10 hours (added anoxic reactor)
RAS (% of Feed Flow Rate)	5/20/97-9/12	~200% (RAS was continually adjusted to keep the solids concentrations at desirable levels. No specific RAS was desired).
NR (% of Feed Flow Rate)	8/27/97-9/12/97	300% (added anoxic reactor)

High ammonia side stream treatment train.

Table 4. Target operating conditions for the MUCT and A2/O high ammonia side stream treatment train.

Parameter	Dates	Value
Seed:	5/19/97	13.5 L of Chesterfield RAS, 21.1 L of HRWTF MLSS, and 25.8 L of tap water.
	6/3/97	Removed 12 L of activated sludge and replaced with mixture of 50% RAS from Chesterfield and 50% tap water
Target SRT	5/20/97-7/2/97	18 days
	7/3/97-9/9/97	24 days
Target Total HRT	5/20/97-6/5/97	17 hours
	6/6/97-6/16/97	20
	6/17/97-8/15/97	26 hours
	8/16/97-8/30/97	21 hours
	8/31/97-9/4/97	18.5 hours
	9/5/97-9/14/97	13.5
RAS (% of Feed Flow Rate)	5/20/97-6/3/97	65%
	6/4/97-9/9/97	100%
NR (% of Feed Flow Rate)	5/20/97-6/5/97	150%
	6/6/97-6/12/97	200%
	6/13/97-6/27/97	250%
	6/28/97-9/9/97	300%
AR (% of Feed Flow Rate)	5/19/97-6/5/97	150%
	6/6/97-6/12/97	200%
	6/13/97-8/22/97	250%
	8/22/97-9/9/97	0* (switched to A2/O the night of 8/22/97)

Monitoring Measurements and Procedures

The pH, DO, and temperature of all reactors were measured and recorded daily in order to quickly assess the state of the reactors. The temperature and pH of the feed were also measured and recorded daily. All measurements were performed in the morning, generally between the hours of 08:00 and 11:00.

Because the goals of this project were simply to determine the feasibility of nitrification and denitrification and come up with preliminary designs for the best options to be tested at the pilot scale, optimizing the pH and DO

was not performed. Rather, the DO was simply monitored to ensure a DO of at least 2 mg/L was achieved in the aerobic reactors and a DO of less than 0.5 mg/L was maintained in the anoxic and anaerobic reactors. The pH was monitored to ensure that it was staying between approximately 6 and 8.5. All pH measurements were made using the Fisher Scientific Accumet 1002 which was calibrated at least once each morning using a three point standard curve. All DO measurements were made using a YSI Model 55 DO meter which was calibrated every time the meter was turned on. Temperature measurements were also made with the YSI Model 55 DO meter. DO measurements were often not made in the heated aerobic reactor because the meter could not determine dissolved oxygen concentrations if the temperature was above 46^{o}C. The vigorous aeration provided by the combined air and pure oxygen in addition to the completeness of nitrification suggested that dissolved oxygen levels were sufficient and were not limiting to the nitrification process. Because the parameters were only used for monitoring, the pH, DO, and temperature data is not supplied in this thesis but is available upon request. Temperature data for the high temperature studies is supplied, however, in Appendices 2 and 14. All pH, temperature, and DO readings were taken in the mornings, generally between 08:00 and 11:00.

Influent flow rates were measured, and adjusted if necessary, daily. All other flow rates were measured two to five times per week. Flow rates were determined simply by opening the appropriate three-way valve installed in each line and allowing the stream to flow into a graduated cylinder of an appropriate size for one to three minutes, yielding a flow rate in mL/min. After determining the flow rate, the mixed liquor or stock solution was poured from the graduated cylinder into the appropriate reactor. Flows were measured and adjusted in the mornings, generally between 08:00 and 11:00, and all flow rate data is provided in Appendices 5, 11, 17, and 24.

Every day the MLSS/MLVSS and ETSS were determined by collecting 500 mL of the effluent and approximately 300 mL from each aerobic reactor into plastic sample bottles. Samples for solids analysis were generally collected between the hours of 11:00 and 14:00. All suspended solids tests were performed in accordance with Standard Methods 2540 D and E, filtering an appropriate volume of effluent or mixed liquor. In addition to the daily aerobic reactor MLSS/MLVSS and ETSS tests, twice per week (every Tuesday and Thursday), the MLSS/MLVSS was measured in all reactors (anoxic and anaerobic reactors in addition to aerobic reactors). One would expect relatively constant solids concentrations throughout the entire treatment train of any given system, but this was generally not the case. Since the aerobic reactors contained the majority of the activated sludges, results from the aerobic MLSS and ETSS were used to determine the volume of activated sludge to be wasted to maintain the desired SRT. Because wastage was based upon aerobic mixed liquor concentrations, their MLSS/MLVSS values were based upon duplicates. Only single samples were used for all other reactor solids analyses. Because the effluent solids were generally low, based upon a fairly large sample volume, and based upon only a grab sample out of a continuously flowing treatment train, only single samples were used for ETSS analyses. Samples for all reactors were collected by opening up the port located at mid depth into a plastic sample bottle. Effluent samples were taken by placing the tube from which the effluent flowed into a 500 mL plastic sample bottle. Wastage was performed manually at the end of each day (generally between the 16:00 and 17:00) by opening up the port at the tail end of the aerobic reactor which was at mid depth of the mixed liquor. Calculations for wastage were based upon the below equation:

$$Q_W = \frac{\left(MLSS \times V_T\right) - \left(Q_{in} \times ETSS \times SRT\right)}{\left(MLSS - ETSS\right) \times SRT} \qquad \text{Eq. 1}$$

for

Q_W = Amount of MLSS wasted each day (L/day)
MLSS = Measured MLSS of the main aerobic reactor (mg/L)
V_T = Total volume of all reactors in each system, not including the final clarifier (L)
Q_{in} = Influent flow rate (L/day)
ETSS = Measured ETSS (mg/L)
SRT = The desired/target solids retention time (days)

The volume of mixed liquor removed to perform the MLSS and MLVSS analyses were always incorporated into the total volume of sludge to be wasted (Q_W).

Sampling and Analysis

Samples for COD, BOD_5, TKN, and nutrient (TN, NO_2^-, $NO_3^- + NO_2^-$, NH_x, and TP) analyses were collected periodically from each reactor and the effluent. On days of sampling and analysis, effluent samples were first collected by placing the effluent tubes in respective two-liter plastic pitchers, and the effluent was then poured into three 500 mL plastic bottles, capped, and placed in a refrigerator until analysis could be performed that day (generally within three hours of collection). The reactor samples were then collected by first turning off the mixers or aeration and all pumps in the treatment train an allowing the reactors to settle (typically ten to twenty minutes). 1.5 liters of supernatant was then drawn off into a two-liter plastic pitcher by siphoning with a 1/4-inch plastic tube, being careful not to draw up the sludge if possible. Poor settlability occasionally did not allow for collecting 1.5 liters of supernatant, in which case as much as could be collected was used for the above analyses. The supernatant was then transferred from the pitcher into three 500 mL plastic bottles, capped, and stored in a refrigerator until analyses could be performed that day. All samples were collected in the morning from approximately 08:00 to 09:00. Upon completion of collecting all samples, all mixers, aerators, and pumps were turned back on, and the supernatant volume removed from each reactor was replaced with tap water of the same volume. Twenty-four hour composite samples taken at the plant for the primary and secondary effluents were used for analysis of the parameters for the feed of the primary effluent, secondary effluent, and heated primary effluent treatment systems. 1500 mL of the high ammonia side stream feed were taken directly from the containing drum at the time of sampling by dipping a two-liter plastic pitcher into the drum and pouring it into three sample bottles. It is important to note that, in aerobic reactors, samples were taken from all sections reactor, not the last baffled section (due to the volume required for analysis and the problem of pulling sludge into samples if trying to draw the supernatant out of the last baffled section only). Thus, values reported for aerobic reactors are *averages* over entire reactor, and not the effluent coming from the reactor. For all data reported, soluble samples were used for all reactor and effluent analyses, and total values were used for influent analyses.

COD analyses were based upon the open reflux method (Standard Method 5220 B), but without the refluxing step. Thus, the values are not true COD values and can only be used for relative comparison. It is not known how much volatile or semi-volatile COD was lost in the procedure, and the wastewaters obviously varied from day to day. Thus, the COD concentrations determined were all biased low by an unknown amount. Because of a communication error associated with BOD_5 determinations, the COD analyses were believed to be the most accurate means of estimating organic concentrations and are thus assumed to be correct for the sake of discussion.

BOD_5 analyses were performed, but a communication problem led to analyses involving total BODs, instead of soluble BOD tests, being performed for the reactors. Because this information was not learned until the termination of the project and there was thus no way to determine how much biomass was present in any given sample, the BOD determinations were ignored and the BOD data is not supplied in this thesis.

Nutrient analyses (TN, NO_2, $NO_3^- + NO_2^-$, NH_x, and TP) were all performed with the Lachat QuikChem 8000 nutrient analyzer, which is EPA approved for analysis of ammonia, nitrite, and nitrates ($NO_3 + NO_2$). TKN and SKN analyses were performed using Standard Method 4500 C. An alternative way of determining TKN and SKN was to subtract out the NO_x concentration from the TN concentration determined by the nutrient analyzer. This is not an approved method for determining TKN, but it was believed to be more accurate, since the Standard Method analyses of TKN and SKN concentrations frequently yielded values which were actually lower than the ammonia concentrations or inexplicably higher than the ammonia concentrations. Therefore, the TKN and SKN determinations based upon the nutrient analyses (instead of the Standard Method analyses) were used for all nitrogen balances, and the Standard Method TKN and SKN data is thus not supplied in this thesis but available upon request.

All COD and nutrient data are listed in the Appendices corresponding to each of the overall systems and each of the individual reactors within each system and are referred to specifically in the text of the following chapters where appropriate. Because the TKN determinations were made simply by subtracting the $NO_3^- + NO_2^-$ concentrations from the TN concentrations, separate columns for TKN estimates are not provided in the Appendices listing raw data, but are provided in the Appendices which provide mass balance calculations.

Mass Balances

Where appropriate, nitrogen and COD balances on the overall system or individual reactors within the system were performed. To characterize nitrification, a nitrogen balance on each overall system was performed. The specific nitrification rate of each sludge was estimated to be the observed overall specific nitrification rate in the following manner:

$$\mu_N = (\text{Total mgTKN-N oxidized/day})/\text{Aerobic biomass} \quad \text{Eq. 2}$$

where the amount of TKN oxidized per day was estimated to be:

$$(\text{mg TKN-N/day})_{oxidized} = (\text{mg TKN-N/day})_{in} - (\text{mg SKN-N/day})_{out} - (\text{mg } N_{growth}/\text{day}). \quad \text{Eq. 3}$$

The aerobic biomass was estimated to be the product of the MLVSS measured in the main aerobic reactor and the volume of the main aerobic reactor, yielding the mg of VSS present in the aerobic environment.

The mg/day of either TKN-N entering the system or SKN-N exiting the system is simply the product of the measured influent TKN or effluent SKN (in terms of mg-N/L) and the influent flow rate in L/day.

The mg/day of nitrogen used for growth (N_{growth}) was estimated to be 12% of the amount of VSS grown per day. The amount of VSS grown per day was based upon the amount of VSS wasted from the aerobic reactor each day to maintain the desired sludge age or solids concentration plus the amount of biomass lost in the final effluent in the following manner:

$$VSS_{growth} = (MLVSS \times Q_W) + (0.8 \times ETSS \times Q_{in}) \quad \text{Eq. 4}$$

for:

VSS_{growth} = Amount of biomass grown (mgVSS/day)
MLVSS = Measured MLVSS of the main aerobic reactor (mg/L)
Q_W = Volume of biomass manually wasted from the aerobic reactor (L/day)
ETSS = Measured ETSS (mg/L). The factor of 0.8 is to convert the ETSS to a volatile suspended solids fraction, which was the observed ratio of the measured MLVSS/MLSS for the systems.
Q_{in} = The influent (and effluent) flow rate (L/day)

Similar nitrogen balances were also performed on just the aerobic reactor of the system. Because there was no way to determine where growth was occurring in the systems from the data collected, all growth as estimated above was assumed to take place in the main aerobic reactor. Using such an approach guarantees a conservative approach to estimating the minimum specific nitrification rate, because the maximum amount of TKN consumption for assimilation is assumed. The specific nitrification rates estimated from the individual aerobic reactors were biased low additionally, because the aerobic reactor was generally ammonia limited, and the nitrification rates are merely averages over the entire aerobic reactor.

The amount of TKN entering the aerobic reactor (in terms of mg-N/day) was estimated to be the sum of the products of the TKN and flow rate of each stream entering the aerobic reactor. The amount of SKN exiting the aerobic reactor (in terms of mg-N/day) was estimated to be the product of the SKN measured in the aerobic reactor and the sum of the flow rates entering the aerobic reactor. Because only soluble data was collected from the reactors and final effluents of each system, the TKN entering the aerobic reactor was estimated using the TKN of the influent where appropriate, and the SKN of all other streams entering the aerobic reactor. As an example, a nitrogen balance on the aerobic reactor of the A2/O configuration was performed as follows (see Figure 2 for a diagram depicting flows):

$$TKN_{oxidized} = TKN_{in} - SKN_{out} - N_{growth} \quad \text{Eq. 5}$$

$TKN_{in} = [SKN_{anox}] \times Q_{anox}$
$Q_{anox} = Q_{inf} + Q_{RAS} + Q_{NR}$
$SKN_{out} = [SKN_{aer}] \times (Q_{inf} + Q_{RAS} + Q_{NR})$

for:

$TKN_{oxidized}$ = Total minimum amount of TKN oxidized in the aerobic reactor (mg-N/day)
TKN_{in} = Total amount of TKN entering the aerobic reactor (mg-N/day)
SKN_{out} = Total amount of SKN exiting the aerobic reactor (mg-N/day)
N_{growth} = Total amount of nitrogen required for growth for the day as estimated above (mg-N/day)
$[SKN_{anox}]$ = Measured SKN concentration of the anoxic reactor (mg-N/L)
Q_{anox} = Flow rate exiting the anoxic reactor and entering the aerobic reactor (L/day)
Q_{inf} = Influent flow rate (L/day)
Q_{RAS} = RAS flow rate (L/day)
Q_{NR} = Nitrate recirculation rate (L/day)d
$[SKN_{aer}]$ = Measured SKN concentration of the aerobic reactor (mg-N/L)

Minor flow rates such as those due to alkalinity, phosphorus, or methanol addition were ignored to simplify calculations because they had a minimal effect.

Nitrogen balances were also performed on the overall system, and on just the anoxic reactor of interest, to estimate specific denitrification rates. The overall observed specific denitrification rate was estimated to be the total amount of nitrates removed via dissimilatory activity normalized to the biomass present in the anoxic reactor of the treatment train of interest. The anoxic biomass was estimated to be the product of the MLVSS and the volume of the anoxic reactor for all systems but the MUCT. For the MUCT process, the anoxic biomass was estimated to be the product of the MLVSS and the sum of both anoxic reactor volumes. The amount of nitrates removed via dissimilatory activity was estimated to be:

$$(\text{mg } NO_x\text{-N/day})_{denitrified} = (\text{mg } NO_x\text{-N/day})_{influent} + (\text{mg TKN-N/day})_{oxidized} - (\text{mg } NO_x\text{-N/day})_{effluent} \quad \text{Eq. 6}$$

where the mg TKN-N/day oxidized was determined from the nitrification mass balance described above in Equation 3, and the mg/day of nitrates entering or exiting is simply the product of the mg-N/L of nitrates in the influent or effluent and the influent flow rate in L/day. The nitrate balance performed on the anoxic reactors simply used the nitrate concentrations and flow rates entering and exiting the reactors, with no oxidation term (because it is assumed that zero TKN is oxidized in the anoxic reactor). Specific denitrification rates determined for the anoxic reactors were normalized to the biomass present in the anoxic reactor of interest.

Because COD balances were performed with only the concern of how much COD was removed, without concern for whether the COD was oxidized or assimilated, the COD balances were much simpler. For the overall systems, COD balances were performed as follows:

$$COD_{removed} = \text{Total } COD_{in} - COD_{out} = [COD_{inf}] \times Q_{inf} + [COD_{MeOH}] \times Q_{MeOH} - [COD_{eff}] \times Q_{eff} \quad \text{Eq. 7}$$

for:

$COD_{removed}$ = Amount of COD removed by the system (mg/day)
Total COD_{in} = Total amount of COD entering the system (mg/day)
COD_{out} = Total soluble COD exiting the system (mg/day)
$[COD_{inf}]$ = Influent COD concentration (mg/L)
$[COD_{MeOH}]$ = Theoretical COD concentration of the methanol stock solution (mg/L)
$[COD_{eff}]$ = Effluent soluble COD concentration (mg/L)
Q_{inf} = Influent flow rate (L/day)
Q_{MeOH} = Methanol addition flow rate (L/day)
Q_{eff} = Effluent flow rate = Influent flow rate (L/day).

COD balances on individual reactors were performed in the same fashion, but with the reactor of interest being used for the control volume, and with the appropriate streams. Obviously, the methanol term was used only in the anoxic reactors which received direct methanol addition.

IV. RESULTS AND DISCUSSION

This project focused on quickly estimating the performance of different treatment trains and strategies for a highly complex and variable industrial wastewater in order to come up with the best candidates for more extensive pilot testing. As a result, operating conditions were changed rapidly and minimal amounts of data were collected, which typically did not reflect steady state estimates. Thus, limited conclusions could be drawn from the data with any significant confidence. Conclusions appropriate to this study are with regard to the feasibility of nitrification/denitrification of the wastewater in question; i.e., could the wastewater successfully be nitrified/denitrified, and if so, to what extent and with what limitations (especially with regard to temperature limitations). The most specific conclusions which could be drawn from the data are explained below, with the focus being on selecting the best option given the data collected. Choosing a best treatment option will be based upon both the effluent quality and the size (and therefore cost) of the final concept.

In order to determine likely performance, it is important to understand when or if breakthrough occurred during operation with respect to constituents such as ammonia, nitrates, or COD. Thus, each system is analyzed below with the focus being on understanding when and why breakthrough appeared to occur. Such understanding of the data is important in order to characterized limitations such as contact time in basins, COD requirements for denitrification, and the likelihood of outcompetition by heterotrophs in the nitrifying basins. Once such limitations are fully described, a full scale design can be determined given expected specific activities and biosolids concentrations. Maximum observed specific nitrification and denitrification rates were determined and used as the basis for estimating what the full scale configurations should be.

The study revealed that it appears to be impractical to attempt high temperature nitrification, especially given basin size constraints due to limitations of available land space and the annual temperature change from moderate to high temperatures which occurs every spring. However, discussion will be devoted to the observed effects of a fairly sudden increase in temperature (as is observed every spring at HRWTF) from ambient conditions to high temperature conditions, i.e., an increase from 28^{o}C to 40^{o}C and above, because of its scientific value. Additional discussion will be devoted to the observed trends of a nitrifying/denitrifying system acclimated to higher temperatures.

Before discussing the results, it is important to point out the non-standard techniques involved with the data analyses (which were mentioned previously in the "Materials and Methods" chapter of this thesis). The first non-standard analysis involves the TKN data. The TKN was determined two ways. The first technique was Standard Method 4500 C, and the second technique was to simply subtract the nitrates (NO_2^- + NO_3^-) from the total nitrogen (TN), with both of these parameters being determined by the Lachat nutrient analyzer available at the HRWTF laboratory. It was decided that the Standard Method determination of the TKN was actually the less reliable of the two techniques, based upon the fact that the Standard Method technique yielded TKN values *lower* than the ammonia values or inexplicably higher than the ammonia values determined by the Lachat nutrient analyzer much more often than using the TKN estimated by subtracting the nitrates from the total nitrogen determinations. Thus, although not a standard method or EPA approved technique, the TKN and SKN data reported are based upon this second technique.

The second non-standard technique involved the COD determinations. The HRWTF laboratory performing the COD analyses uses a "very open" reflux technique, where the refluxing step is skipped, and the vapors are allowed to escape during heating of the sample. This is a time-saving approach which enables them to obtain estimates for BOD_5 dilutions, the parameter which they use for compliance data. This fact was not discovered by the researcher until after the bench-scale testing was completed, and thus all COD values reported are only partial CODs. It is not known if only the volatile or both the volatile and semi-volatile COD fractions are affected by this, and no attempt was made to determine this because of the complex and variable composition of the wastewaters treated in this study. The end result is that all COD values reported are biased low by an unknown amount. Another result of the incomplete COD analysis technique is that the systems might actually have been overloaded, especially with respect to the methanol dosage.

The CODs could not be correlated to the BOD_5 data because of a third technicality. Due to a communication error, the soluble BOD_5 samples were not being filtered, only settled; a fact also not discovered until the termination of the project. As a result, it is not known how much biomass was in any given sample or how this affected any given sample. Thus, the BOD values were ignored, and those data are not supplied in this thesis. Only the COD data are used for analyses pertaining to organics, and, although it is known that the values are biased low, they are assumed, for the sake of discussing trends, to be consistent data and approximately correct. Because the COD values of the methanol stock solutions were never measured, the COD due to methanol addition had to be estimated as the 1.5 mg of oxygen required to react stoichiometrically with 1 mg of methanol (CH_3OH) to form

carbon dioxide and water. All other analyses were performed as described in the "Materials and Methods" portion of this thesis.

Results and Discussion: Secondary Effluent Treatment Train

The secondary effluent was a low carbon wastewater (COD concentrations during testing were typically 100 to 250 mg/L) with a relatively high TKN (TKN concentrations during testing were typically 25 to 50 mg-N/L) and was treated with an aerobic/anoxic process in this study. The wastewater was treated by a large aerobic basin followed by a shorter anoxic denitrification zone, with the effluent from the anoxic basin being followed by a very small aeration basin for sludge conditioning, to improve settlability by releasing nitrogen bubbles from the flocs prior to their entering the final clarifier. Methanol was added to the anoxic reactor directly because the waste stream was carbon deficient. Dosage was optimized such that denitrification was never carbon limited, while the minimum possible (recalcitrant) COD was observed in the final effluent.

The sludge age was held relatively constant for the duration of the experiment at a 6 day SRT, with the exception of system upsets or wastage calculation errors. Thus, with the exception of the high temperature study, data from all secondary effluent operation periods were compared to each other.

Although steady-state conditions were probably never reached, quasi-steady-states were delineated for each operation period based upon the MLVSS, SRT, hydraulic loading, COD and nitrogen loading, and COD and nitrogen removal performance. Unless otherwise noted, only data collected during those quasi-steady-state periods were used when computing average values for each operation period or for any other comparison or analysis; and from this point forward, it is to these quasi-steady-state determinations that the term "steady state" will refer. As the COD and nitrogen removal recovered to steady-state values within a few days after a change in operating conditions, it is the MLVSS that dominated the delineation of steady-state operation, which was based upon visually inspecting the plot of the MLVSS with respect to time and selecting the dates between which the MLVSS appeared to maintain a "horizontal" trend (as opposed to an increasing or decreasing trend) with respect to time (see Figure 6).

As this was a real industrial wastewater, the COD and TKN were subject to fluctuations, and the strength of the wastewater in terms of both COD and TKN gradually declined with time during the duration of the experiment. The loadings to the bench-scale system in terms of mg/day were increased, however, by increasing the hydraulic loading. Three separate hydraulic loadings were used, and either a change in biosolids in the system or the composition of the wastewater changed from one relatively steady state period to another during two of the operation periods. Thus, based upon the hydraulic loading, MLVSS, wastewater composition, and the other factors mentioned above, five separate operation periods were delineated and analyzed, with a sixth separate period being delineated for the high temperature study. The reasons for the separate delineations of each operation period are described below in Table 5, and Table 6 provides the overall characteristics of each operation period during moderate temperature testing. Comparisons of performance with respect to loading were made only with the first five operation periods. The results of the high temperature study are treated separately to illustrate the dying off of a system subjected to a sudden increase in temperature, i.e., from approximately 28^{o}C to 40^{o}C and above. Comparisons of the high temperature study (operation period 6) to the performance during the operation period prior to the increase in temperature (operation period 5) are made where appropriate, as both operation periods had relatively similar COD and TKN loadings. Observed trends with respect to loadings and temperature are discussed below.

It was determined that the secondary effluent of HRWTF could be completely nitrified and denitrified, with zero ammonia and near-zero nitrates in the effluent with the treatment strategy tested. Low SKN concentrations were also observed in the effluent, but this was considered recalcitrant since zero ammonia was leaving the system, and since the SKN values were consistently at approximately 1 mg-N/L regardless of the loading. COD removal was also adequate, and the effluent COD was also assumed to be recalcitrant since it remained at a consistent level regardless of loading rates, as well as the fact that the levels were much lower than the influent (which is presently the final effluent of HRWTF). Because chemical analyses were typically not performed during the first few days of a change in loading, it is not known how much COD or nitrogen in any form escaped the system upon the sudden increase, but effluent concentrations within the steady-state range were generally achieved within four or five days of the change in loading, which is slightly less time than the SRT of the sludge. Below is a discussion of the performance with respect to all pertinent wastewater components.

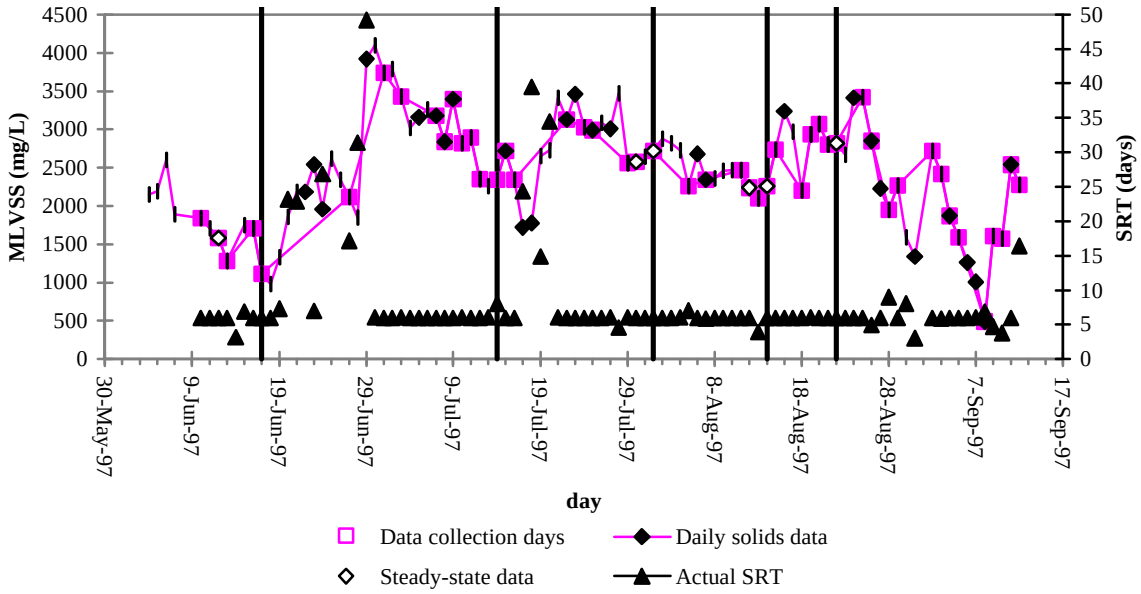

Figure 6. Secondary effluent treatment train: Using the MLVSS and SRT to help delineate steady state operation periods. The bold lines indicate the last day of a steady state operation period.

Table 5. Characterization and delineation of separate operation periods for the secondary effluent treatment train.

Operation period	Steady state dates	Reason for separate delineation
1	6/12-6/17	• Reason for start: Stabilized performance after startup. • Reason for end: Poor removal observed because of infestation of grazers ("sludge worms") from 6/18 to 6/26. • Problems prior to start of next period: Added 2.8 L of nitrifying biomass on 6/28.
2	7/11-7/14	• Reason for start: Performance stabilized by 7/7. • Reason for end: Increased hydraulic loading and methanol dosage after 7/14 sample. • Problems prior to start of next period: Poor performance due to infestation of grazers from 7/17 to 7/18.
3	7/29-8/1	• Reason for start: Performance stabilized by 7/29. • Reason for end: Increased hydraulic loading on 8/5.
4	8/11-8/14	• Reason for start: Performance stabilized by 8/11. • Reason for end: Wastewater COD doubled from 8/14 to 8/15.
5	8/19-8/22	• Reason for start: Performance and wastewater stabilized by 8/19. • Reason for end: Began high temperature study by insulating all reactors and adding heating elements to both aerobic reactors on afternoon of 8/25.
6	8/26-9/12	• Reason for start: High temperature study started 8/26. • Performance never stabilized.

Table 6. Secondary effluent treatment train: Overall performance and loading characteristics.

Steady-state operation period	**1** 6/12-6/17		**2** 7/11-7/14		**3** 7/29-8/1		**4** 8/11-8/14		**5** 8/19-8/22	
SRT (days)	**5.7** 1.3	6	**6.5** 1.0	4	**6.0** 0.0	4	**5.5** 1.0	4	**6.0** 0.0	4
Total HRT (hours)	**7.2**	6	**7.3**	4	**4.8**	4	**3.9**	4	**4.0**	4
Influent flow rate (L/day)	**102** 5	6	**100** 1	4	**152** 3	4	**187** 0	4	**185** 1	4
RAS % of influent flow rate (%)	**64** 4	4	**63** 1	4	**64** 3	4	**52** n/a	2	**53** 1	3
MLVSS (mg/L)	**1430** 286	6	**2463** 294	4	**2625** 72	4	**2268** 153	4	**2908** 125	4
ETSS (mg/L)	**48** 53	6	**17** 26	4	**31** 24	4	**60** 33	4	**9** 8	4
Specific growth rate (1/day)	**0.17** 0.00	4	**0.15** 0.03	4	**0.16** 0.00	4	**0.17** 0.01	3	**0.17** 0.00	4
TKN influent (mg-N/day)	**4117** 1241	3	**3797** 498	2	**6933** 1109	4	**5633** 757	4	**6465** 279	4
SKN effluent (mg-N/day)	**178** 78	3	**81** 48	2	**145** 9	4	**183** 50	4	**188** 33	4
NHx influent (mg-N/day)	**3275** 661	3	**2866** 121	2	**6463** 1047	4	**5308** 338	4	**5639** 569	4
NHx effluent (mg-N/day)	**6** 10	3	**0** 0	2	**0** 0	4	**0** 0	4	**0** 0	4
NOx influent (mg-N/day)	**285** 28	3	**932** 170	2	**1636** 168	4	**939** 532	4	**631** 298	4
NOx effluent (mg-N/day)	**156** 174	3	**0** 83	2	**33** 102	4	**23** 47	4	**31** 57	4
COD influent (mg/day)	**20097** 4686	3	**14818** 10155	2	**21603** 6413	3	**17456** 1830	4	**37221** 5056	4
Methanol addition?	yes		yes		yes		yes		yes	
Total COD in (mg/day)	**59356** 6006	3	**49433** 10149	2	**73171** 6591	3	**69380** 2224	4	**88718** 4842	4
COD effluent (mg/day)	**8464** 2365	3	**6048** 1180	2	**7505** 1137	3	**7628** 619	4	**9574** 438	4
TKN influent (mg-N/L)	**39.5** 10.9	3	**37.9** 5.2	2	**45.4** 7.1	4	**30.1** 4.0	4	**34.9** 1.7	4
SKN effluent (mg-N/L)	**1.7** 0.7	3	**0.8** 0.5	2	**1.0** 0.0	4	**1.0** 0.3	4	**1.0** 0.2	4
NHx influent (mg-N/L)	**31.5** 5.8	3	**28.4** 1.2	2	**42.4** 6.8	4	**28.4** 1.8	4	**30.5** 3.2	4
NHx effluent (mg-N/L)	**0.1** 0.1	3	**0.0** 0.0	2	**0.0** 0.0	4	**0.0** 0.0	4	**0.0** 0.0	4
NOx influent (mg-N/L)	**2.8** 0.3	3	**9.2** 1.7	2	**10.7** 1.2	4	**5.0** 2.8	4	**3.4** 1.6	4
NOx effluent (mg-N/L)	**1.5** 1.7	3	**0.0** 0.8	2	**0.2** 0.7	4	**0.1** 0.2	4	**0.2** 0.3	4
COD influent (mg/L)	**193** 41	3	**147** 102	2	**141** 41	3	**93** 10	4	**201** 28	4
COD effluent (mg/L)	**81** 21	3	**60** 12	2	**49** 8	3	**41** 3	4	**52** 3	4
% bioavailable TKN oxidized (nitrified)	**78** 5	3	**53** 12	2	**76** 4	4	**69** 10	4	**72** 1	4
Overall observed specific nitrification rate* (mg-N/mgVSS/day)	**0.131** 0.046	3	**0.043** 0.021	2	**0.110** 0.021	4	**0.093** 0.023	4	**0.086** 0.003	4

Note: Top value indicates mean value during operation period. The lower value indicates the standard deviation. The value to the right indicates the number of samples in the data set.

* Nitrification was ammonia limited for all operation periods.

Feasibility of nitrification of the Secondary Effluent at moderate temperatures

For all loadings tested at moderate temperatures, the SKN was consistently removed by the system to a range of zero to 2.4 mg-N/L during steady state operation (see Appendix 1). Average steady state effluent SKN concentrations for each operation period were between 0.8 and 1.7 mg-N/L (see Table 6). The treatment train was never loaded high enough to observe what could be regarded as a steady state breakthrough of bioavailable SKN. The effluent ammonia concentrations were also consistently very low. The steady state effluent ammonia concentrations were always non-detectable with the exception of one day on which the ammonia concentration was measured to be 0.2 mg-N/L (see Appendix 1). Even transitional days not associated with system upsets yielded effluent ammonia concentrations in the range of zero to 1.0 mg-N/L (see also Appendix 1). No steady state ammonia breakthrough was observed during any of the loadings tested, indicating that the system was very underloaded with respect to nitrification.

An attempt was made to perform a nitrogen balance on the aerobic reactor in order to determine specific nitrification rates, but with only two to three data points per operation period, little can truly be said with any confidence. As previously discussed in the "Materials and Methods" portion of this thesis, there was no way to determine where nitrogen assimilation was occurring in the system, and it was assumed that all growth took place within the aerobic reactor, which would yield a minimum value for the TKN oxidation estimate. The specific nitrification rate was estimated a second way by using the observed nitrate production. This technique will be biased low because it too is only an average over the entire reactor, but it is biased low additionally because it relies on the assumption that zero denitrification occurred in the aerobic reactor. This second technique for estimating nitrification rates did consistently yield higher rates however, which would be closer to the true nitrification rate. Because the aerobic reactor was generally ammonia limited, the rates are still obviously biased low. The performance of the aerobic reactor is summarized in Table 7. No reason could be found to account for the higher amount of ammonia escaping the aerobic reactor during operation period 3. It was likely due to a minor system upset, because the ammonia concentrations exiting the aerobic reactor during operation period 3 were zero on two days and 5.3 on the third day of data collection (see Appendix 2). The estimated specific nitrification rates correlated well to the F:M value (the total mg-N/day of TKN entering the system normalized to the amount of biomass in the main aerobic reactor; or mgTKN-N/mgVSS/day), but because estimated rates generated from a balance on just the main aerobic reactor were lower than overall observed specific nitrification rate estimates, only the relationship observed in the overall observed specific nitrification rate is provided (see discussion below). Perhaps the most significant information to be gained from the nitrogen balance on the aerobic reactor is the knowledge that nitrification was completed in the aerobic reactor (as indicated by zero ammonia concentrations exiting the aerobic reactor), and very little nitrification was occurring elsewhere in the system.

As described in the "Materials and Methods" chapter of this thesis, a nitrogen balance on the overall system was performed to determine the overall observed specific nitrification rate and compare it to the observed specific nitrification rates determined from the aerobic reactor nitrogen balance. The overall nitrogen balance produced higher estimates of the specific nitrification rate than the nitrogen balance on the main aerobic reactor. While some nitrification would inevitably occur in other zones of the system, the nitrogen balance on just the main aerobic reactor was most likely biased low because of the assumption that all growth occurred within the main aerobic reactor. A plot of the overall observed specific nitrification rate versus the overall nitrification F:M ratio (mgTKN-N/mgVSS/day) is shown in Figure 7. While the influent TKN loading was used for the determination of the F:M ratio, only the biomass present in the main aerobic reactor was used for the F:M or observed specific nitrification rate determinations.

Because the maximum observed specific nitrification rate appeared to follow the same trend as the rest of the data, and did not lie unusually far above or below the apparent trend, the maximum observed rate was assumed to be a valid estimate of the sludge performance. Figure 7 illustrates very well that the system was never stressed with respect to the maximum specific nitrification rate because the curve did not break into a horizontal trend to indicate that the specific nitrification rate was independent of an increased loading. Because it is the only data collected, the maximum observed specific nitrification rate will be assumed to be the true maximum for the sake of discussion of design. Thus, from the existing data, the maximum overall observed specific nitrification rate of the system that treated the secondary effluent of HRWTF was determined to be 0.18 mg-N/mgVSS/day.

Table 7. Secondary effluent treatment train: Aerobic reactor performance and loading.

Steady-state operation period	**1** 6/12-6/17	**2** 7/11-7/14	**3** 7/29-8/1	**4** 8/11-8/14	**5** 8/19-8/22
Aerobic HRT (hours)	2.6	2.7	1.7	1.5	1.5
TKN in (mg-N/day)	3631	4261	6897	6022	6453
SKN out (mg-N/day)	805	452	1051	1138	332
Bioavailable TKN in (mg-N/day)	3418	4191	6650	5804	6302
TKN in - SKN out (mg-N/day)	2826	3809	5846	4884	6121
TKN used for growth by total system (mg-N/day)	826	1730	1563	1414	1743
TKN available for oxidation (mg-N/day)	2593	2461	5087	4389	4558
Estimated minimum SKN nitrified (mg-N/day)	2000	2079	4284	3470	4378
Measured NOx production (mg-N/day)	2629	2983	4172	3670	5097
Estimated minimum specific nitrification rate (mgSKN-N/mgVSS/day)	0.079	0.041	0.091	0.086	0.085
Measured specific NOx production rate (mgNOx-N/mgVSS/day)	0.107	0.059	0.089	0.091	0.099
% bioavailable TKN removed	81	90	90	82	97
Estimated minimum % bioavailable TKN oxidized	57	49	65	57	69
Estimated % oxidizable TKN nitrified	75	83	87	76	96
TKN in (mg-N/L)	21.7	26.3	27.4	21.2	22.7
SKN out (mg-N/L)	4.8	2.8	4.2	4.0	1.2
NHx out (mg-N/L)	2.2	0.0	2.7	0.0	0.0
COD in (mg/day)	21996	33353	22481	21878	41676
COD out (mg/day)	11463	11228	11724	11484	15592
COD in - COD out (mg/day)	10533	22126	10757	10394	26084
COD in (mg/L)	131	205	88	77	147
COD out (mg/L)	69	69	46	41	55

Note: There are two data points for each operation period corresponding to nitrogen data. Standard deviations do not apply.
There are two data points per operation period corresponding to COD data except operation period 3 which has only one data point.

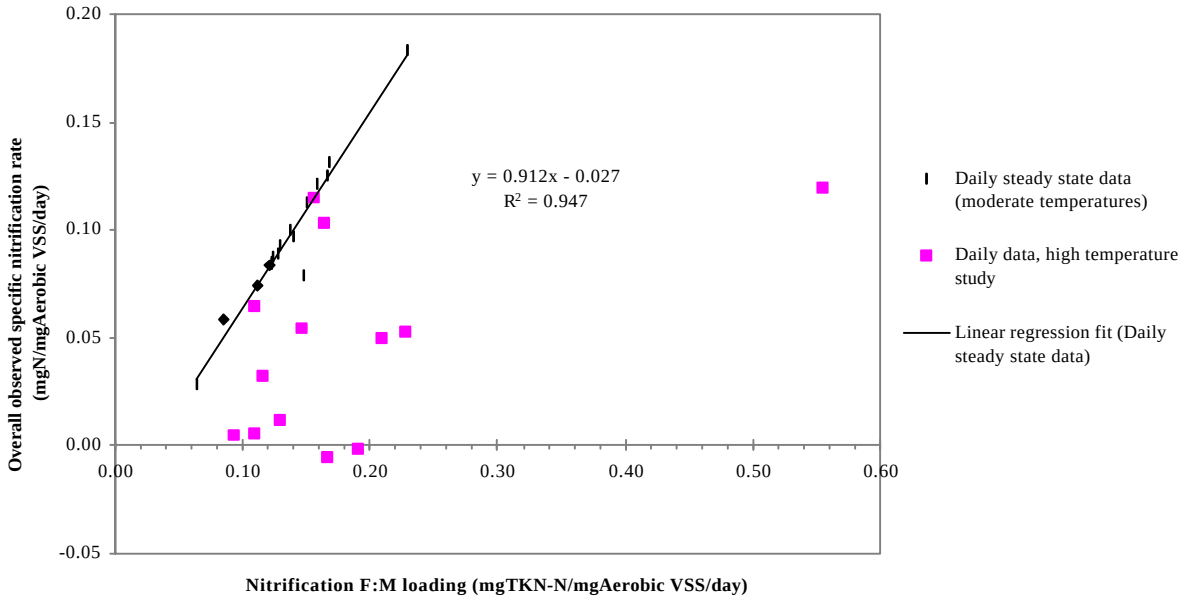

Figure 7. Secondary effluent treatment train: Overall observed specific nitrification rate as a function of the F:M loading.

Feasibility of denitrification of the Secondary Effluent at moderate temperatures

Effluent nitrate concentrations for all loadings tested were excellent, with no obvious nitrate breakthrough occurring. Steady-state average effluent concentrations of NO_x for each respective operation period ranged from 0.1 to 1.5 mg-N/L, with the high values corresponding to the first two operation periods (see Table 6). Earlier steady state effluent NO_x concentrations ranged from zero to 3.5 mg-N/L, which corresponded to the first 45 days of operation. The slightly poorer denitrification performance during the first forty-five days of operation appears to be the result of slow acclimation, a typical response for the startup of methanol denitrifying systems. Steady-state effluent NO_x concentrations after July 10 (i.e., after acclimation) ranged from only zero to 0.8 mg-N/L (see Appendix 1). Although some nitrates were apparently escaping the system on occasion, the levels were not high enough or consistent enough to constitute what should be regarded as nitrate breakthrough (see Appendix 1). As the COD leaving the anoxic reactor was consistently higher than the COD leaving the final clarifier of the treatment train by 64 to 107 mg/L (see Table 8), any nitrates escaping the system were due to insufficient contact time in the anoxic reactor, and not COD limitations.

A mass balance was performed on the anoxic reactor itself, which demonstrated that denitrification of the secondary effluent was, in fact limited by contact time. Operation periods 1 and 2 were associated with methanol acclimation, which is supported by the fact that the anoxic reactor was subjected to the lowest nitrate and hydraulic loadings during these two operation periods, yet these two periods corresponded to the greatest amount of nitrates escaping the anoxic reactor (see Table 8 for all discussion pertaining to performance of the anoxic reactor). Acclimation was complete by operation period 3, as indicated by zero nitrates being detected leaving the anoxic reactor. The COD:NO_x loading to the anoxic reactor remained essentially the same during operation periods 3 through 5, in the range of 11.2 to 11.5, but some nitrates were escaping during operation periods 4 and 5. Both of these operation periods corresponded to the shortest anoxic HRT of 0.7 hours, demonstrating that the maximum specific denitrification rate was being reached. The slightly less than complete denitrification to zero levels of NO_x in the anoxic reactor are of no concern, however, considering the fact that the average concentration of NO_x exiting the anoxic reactor during operation periods 4 and 5 were only 0.6 and 0.5 mg-N/L, respectively. Some denitrification was apparently occurring in the final clarifier, creating an even cleaner final effluent. Average final effluent NO_x concentrations after acclimation, i.e., after operation period 2, were in the range of 0.1 to 0.3 mg-N/L.

As with nitrification, the observed overall maximum specific denitrification rate was compared with the denitrification rate based upon a nitrogen balance performed on just the anoxic reactor. Also as with the nitrification rates, the overall observed rates yielded higher values than the specific rate based upon just the individual reactor. Thus, discussion will be restricted to the overall observed specific denitrification rate. The overall observed specific denitrification rate was normalized to the biomass in just the anoxic reactor, and the F:M ratio is expressed in terms of the influent TKN loading normalized to the biomass in the anoxic reactor, which is shown in Figure 8. No correlation could be made comparing the observed specific denitrification rate to a COD-based F:M ratio.

Table 8. Secondary effluent treatment train: Anoxic reactor performance and loading.

Steady-state operation period	**1** 6/12-6/17	**2** 7/11-7/14	**3** 7/29-8/1	**4** 8/11-8/14	**5** 8/19-8/22
Anoxic HRT (hours)	1.2	1.3	0.8	0.7	0.7
Methanol loading to anoxic reactor (mgCOD/day)	38120	34616	51283	52137	51710
Total COD loading to anoxic reactor (mgCOD/day)	49583	45844	63007	63621	67302
NOx loading to anoxic reactor (mg-N/day)	3054	4067	5919	5617	5874
COD:NOx loading ratio to anoxic reactor	16.2	11.3	10.6	11.3	11.5
Amount of NOx exiting anoxic reactor (mg-N/day)	676	181	0	350	132
Concentration of NOx exiting anoxic reactor (mg-N/L)	4.0	1.1	0.0	0.6	0.5
Amount of COD exiting anoxic reactor (mg/day)	25830	22660	26352	30828	35243
Concentration of COD exiting anoxic reactor (mg/L)	149	135	100	106	121
COD of final effluent (mg/L)	70	51	40	42	54
% NOx denitrified	78	96	100	94	98
Observed specific denitrification rate (mgNOx/mgVSS/day)	0.216	0.168	0.279	0.288	0.245
SKN in (mg-N/L)	4.8	2.8	4.2	4.0	1.2
SKN out (mg-N/L)	2.6	0.9	2.3	0.7	1.0
NHx in (mg-N/L)	2.2	0.0	2.7	0.0	0.0
NHx out (mg-N/L)	1.6	0.0	1.6	0.0	0.0

Note: There are two data points for each operation period corresponding to nitrogen data. Standard deviations do not apply.
There are two data points per operation period corresponding to COD data except operation period 3 which has only one data point.

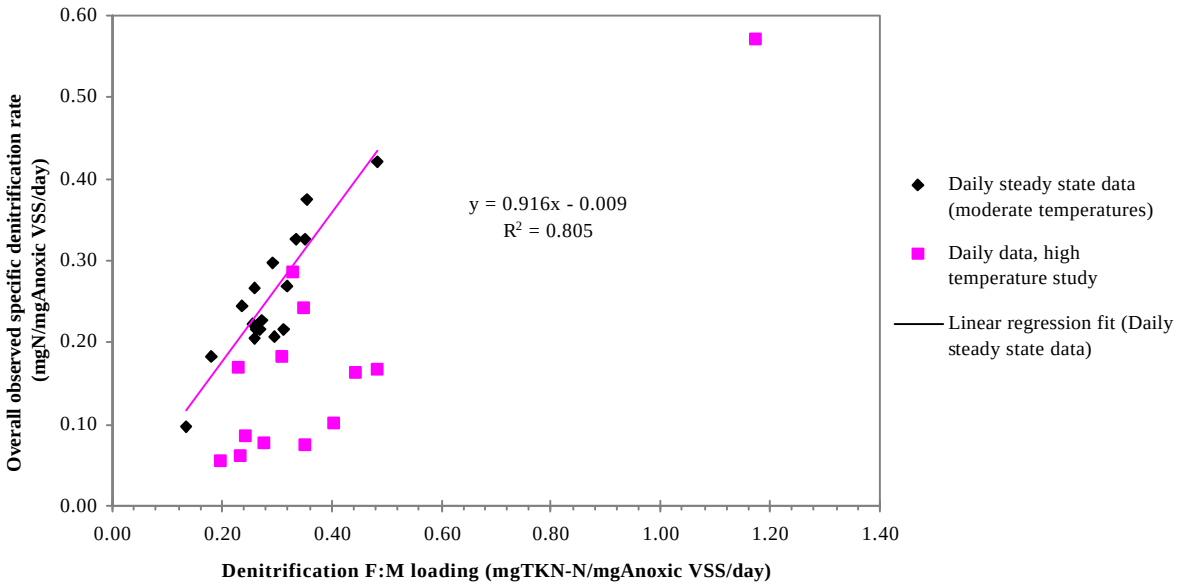

Figure 8. Secondary effluent treatment train: Overall observed specific denitrification rate as a function of the F:M loading.

As with the observed nitrification rates, the overall observed specific denitrification rate did not appear to reach a maximum, as would be indicated by a horizontal trend. For the sake of discussion, the maximum specific denitrification rate observed will be assumed to be the true maximum rate, which was determined to be 0.42 mgN/mgVSS/day.

Other performance characteristics of the Secondary Effluent treatment train at moderate temperatures

As with nitrification and denitrification, removal of organics was essentially complete for all loadings tested. Final effluent steady state soluble COD concentrations during operation periods 3 through 5 were generally in the range of 37 to 58 mg/L, with higher concentrations (ranging from 55 to 105 mg/L) escaping during the first two operation periods (see Appendix 1). As stated above, the poorer response during the first two operation periods was most likely due to slow acclimation to methanol. Because this range of effluent COD concentrations was consistently measured during all loadings tested, it was decided that COD breakthrough did not occur and that the COD in the effluent was recalcitrant to biodegradation. Given that denitrification was generally complete and not COD limited, the recalcitrant effluent COD indicates that the methanol dosage was optimized well.

With respect to determining methanol requirements, excellent nitrate removal was observed for all loadings tested, and the minimum COD:NO_x ratio observed in the anoxic reactor was 10.6, which is thus the maximum required COD loading to the anoxic zone for this treatment strategy. It was further determined that for every 1 lb of TKN-N entering the system in the influent, approximately 0.96 lb of nitrate-N entered the anoxic reactor (see Figure 9). It was also determined that the COD loading to the anoxic reactor from the main aerobic reactor of the treatment train was approximately 50% of the COD loading to the influent (see Figure 10). Thus, given the TKN and COD of the final effluent of HRWTF, methanol requirements for complete denitrification at moderate temperatures can be determined if desired.

The ETSS of the system measured on steady-state data collection days was generally in the range of 2 to 25 mg/L with most outliers corresponding to the system startup, the high temperature study (to be addressed below in the high temperature discussion), and the first week at the highest hydraulic loading (see Appendix 4). The early high ETSS was probably due to the system not being fully acclimated, while the higher ETSS at the higher hydraulic loading was probably due to a washing out of poorer settling biomass followed by a final shift to a better settling

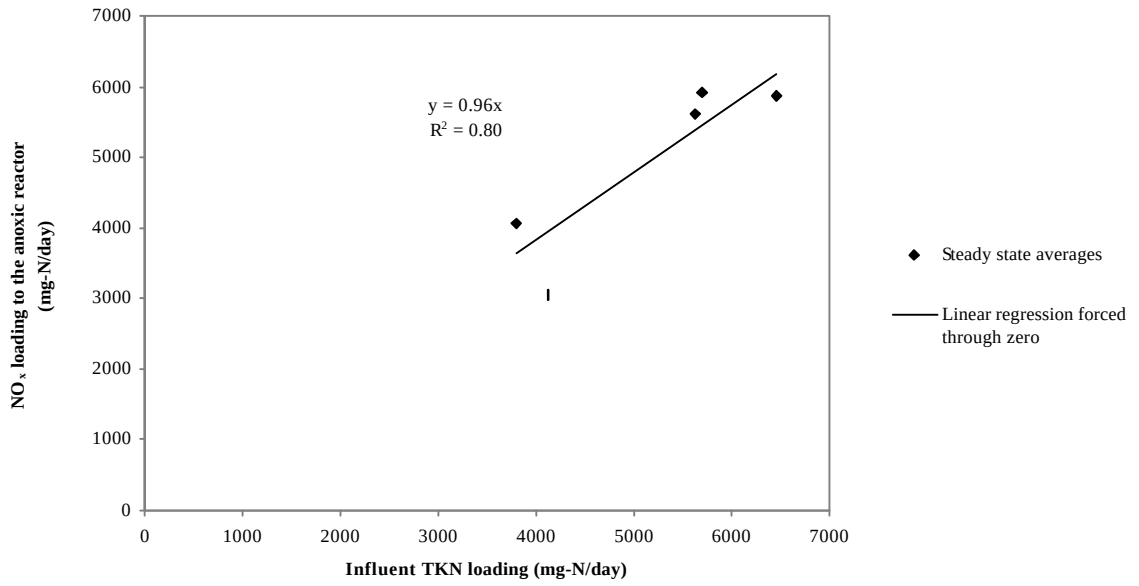

Figure 9. Secondary effluent treatment train: Estimating the relationship between the influent TKN loading and the NO_x loading to the anoxic reactor. The slope of the linear fit is the fraction of influent TKN that reaches the anoxic reactor as nitrates.

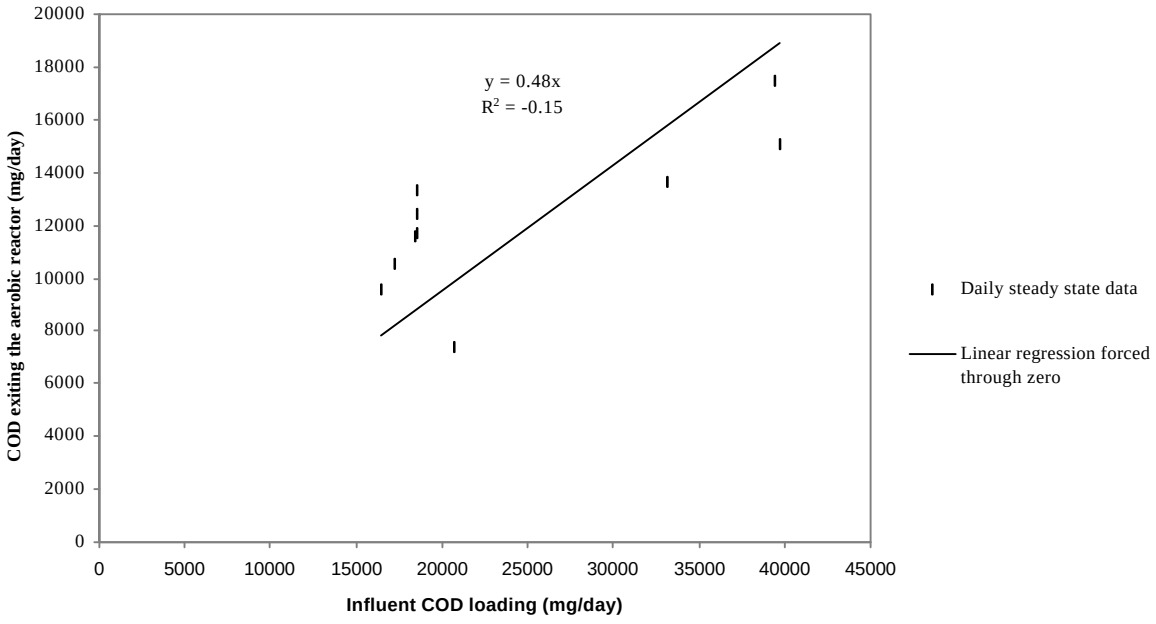

Figure 10. Secondary effluent treatment train: Estimating the amount of influent COD which is consumed prior to the wastewater reaching the anoxic basin. The slope of the linear fit is the approximate percent of COD consumed before anoxic treatment.

sludge, as indicated by the better ETSS during the fifth operation period which was at the same hydraulic loading as the fourth operation period (see Figure 11 and Table 6). Although data corresponding to steady state data-collection days yield reasonably good ETSS values, the daily ETSS values, even when ignoring severe upsets such as infestation by grazers, indicate the effluent solids quality was quite sporadic (see Appendix 4 and Figure 11). This was either due to the method of data collection (one grab sample per day), or perhaps an indication that the aerobic/anoxic treatment of the secondary effluent for nitrification and denitrification yielded a sludge with unstable settlability characteristics. The ETSS might also have been affected by the C:N ratio, however.

The C:N ratio was calculated to be the ratio of bioavailable COD to bioavailable TKN. The bioavailable COD was estimated as the total COD entering the system through the influent and methanol addition minus the soluble COD of the final effluent (which was assumed to be biologically recalcitrant). The bioavailable TKN was estimated to be the TKN entering the system in the influent less the soluble organic nitrogen in the final effluent of the system, with the effluent soluble organic nitrogen being estimated as the effluent SKN minus the effluent soluble ammonia concentration. Because hydraulic loading varied and has a significant affect on the various parameters, each hydraulic loading was analyzed separately with respect to trends resulting from varied C:N ratios. Thus, operation periods 1 and 2 were analyzed together, operation period 3 was analyzed by itself, operation periods 4 and 5 were analyzed together, and the high temperature study was analyzed by itself. With the limited amount of data collected, no truly significant trends with respect to the C:N ratio could be determined. However, the C:N ratio may, in fact, have had an impact on the effluent solids quality. For instance, it was observed that all steady-state points beyond a C:N ratio of approximately 14 corresponded to a lower and more consistent ETSS than the highly scattered data for C:N ratios below this value (see Figure 12). Because of the scatter, and the fact that such C:N ratios were achieved only during the lowest hydraulic loading and the high temperature study, it is not clear whether there is no significant trend associated with the C:N ratio, or whether the addition of a carbon source to keep the C:N ratio above 14 would truly help maintain a consistently low ETSS for this system. Further testing would certainly be required, but discovering ways to maintain a stable effluent quality is ample justification if treatment of the secondary effluent were to be considered the best option.

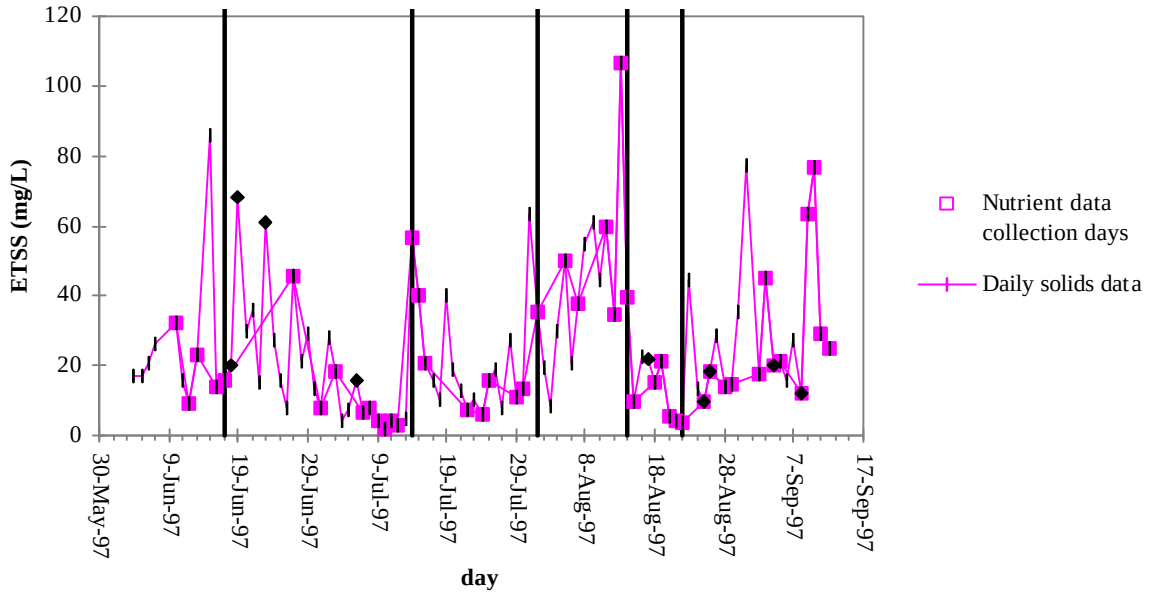

Figure 11. Secondary effluent treatment train: Daily effluent suspended solids quality The bold lines indicate the last day of a steady state operation period.

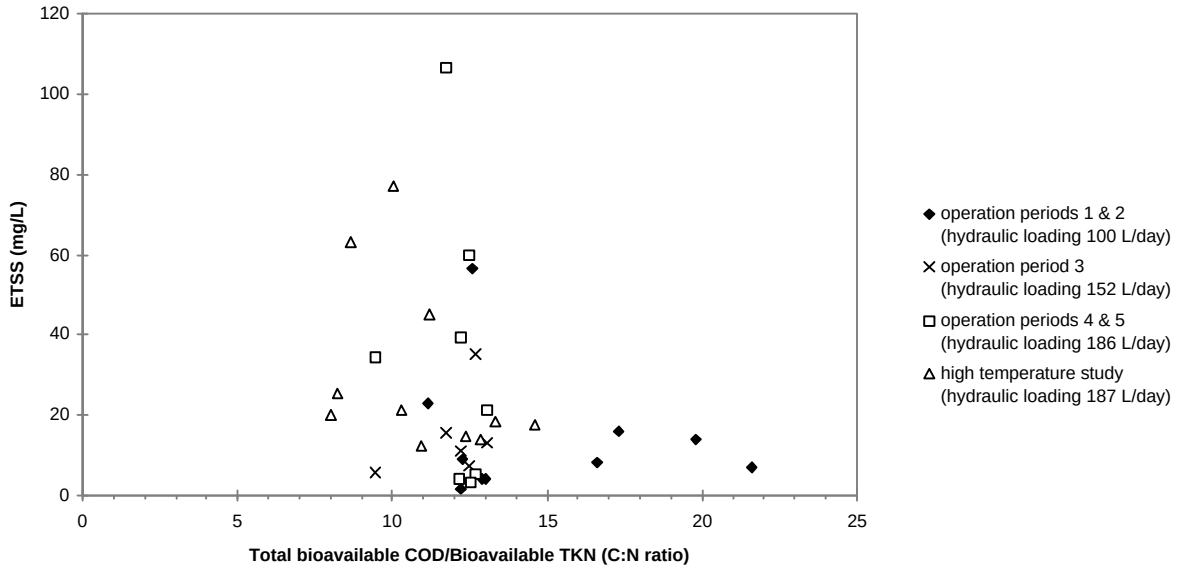

Figure 12. Secondary effluent treatment train: The effect of C:N loading on effluent suspended solids concentrations.

Full scale design of the secondary effluent treatment train

Regardless of the trace amounts of nitrates escaping the system during steady-state operation, the effluent quality of each operation period, with average SKN values ranging from 0.8 to 1.7, average ammonia concentrations ranging from zero to 0.1, average NO_x concentrations ranging from 0.1 to 1.5, and average COD concentrations ranging from 41 to 81 was excellent for all loadings tested (see Table 6). Because no breakthrough was observed for any of the wastewater constituents monitored, and because a reasonable ETSS was achieved after acclimation (although it is important to note that effluent solids quality was sporadic), the maximum observed specific removal rates and hydraulic loadings will be used as the basis for scaling up the treatment train to full size. Bear in mind that this is only a rough approximation and that pilot testing is to be done before a true full-scale design is to be selected.

The system tested tended to operate at an MLVSS of approximately 2500 mg/L, which will thus be assumed for the full scale design. Assuming nitrification and denitrification will be operated at 80% of the observed maximum specific activities to provide a safety factor against spikes in loading, a specific nitrification rate of 0.15 mgN/mgVSS/day and specific denitrification rate of 0.34 mgN/mgVSS/day will be assumed. The average TKN concentration of the final effluent of HRWTF during this study was 35.2 mg-N/L. Scaling the hydraulic loading up to 50 MGD (189.25 million liters per day), an average TKN loading of 6.66×10^9 mg-N/day (14,700 lb-N/day) is expected. Complete nitrification at the assumed specific nitrification rate would thus require 4.44×10^{10} mg of VSS. Assuming a 2500 mg/L MLVSS concentration, an aerobic basin volume of 1.78×10^7 L, or 4.70 million gallons would be required. Assuming complete nitrification and complete denitrification, in addition to the assumed denitrification rate of 0.34 mgN/mgVSS/day, an anoxic basin of 2.07 million gallons (7.84×10^6 L) would be required to denitrify the 6.66×10^9 mg-N/day with a 2500 mg/L MLVSS concentration.

Given the 50% RAS (approximately 100 L/day) and the reasonable ETSS with a 20 minute retention time in the reaeration basin, a full size reaeration basin of 1.08 million gallons should suffice. If the basin is scaled up to a more commonly accepted 30 minute retention time (Sedlak, 1991), the basin would be 1.62 million gallons. Although the data suggest the 20 minute retention time would be sufficient, a 30 minute reaeration basin would be used in the full size design estimate to remain conservative and allow for any possible increases in hydraulic loading. Thus, the total volume of all three basins (using the larger reaeration basin) would be 8.39 million gallons. At 50 MGD, the overall HRT in the basins would be 4.0 hours.

In terms of solids handling, with the exception of the first operation period, the amount of biomass produced appeared to be linearly dependent upon the COD loading to the system (see Figure 13). Assuming a linear trend and using a linear regression fit, it is estimated that 470 lbs of biomass (VSS) will be grown/wasted for every 10,000 lb of COD entering the system. Note that roughly 50 to 80% of this COD loading is methanol required for denitrification of a low-carbon effluent (see Table 6). Assuming the average COD loading of operation period 5 (88,718 mg/day) is scaled up to the 50 MGD design (by the factor 9.96 x 10^5), 88,274 kg/day (or 195,100 lb/day) of COD is expected to be removed by the system. This should result in 9,170 lbs of biomass production per day to be wasted.

The 6 day SRT was not varied at all during the experiment, but proved to be sufficient, as complete nitrification and denitrification was consistently achieved, and acclimation always occurred within 4 to 5 days of a change in operating conditions.

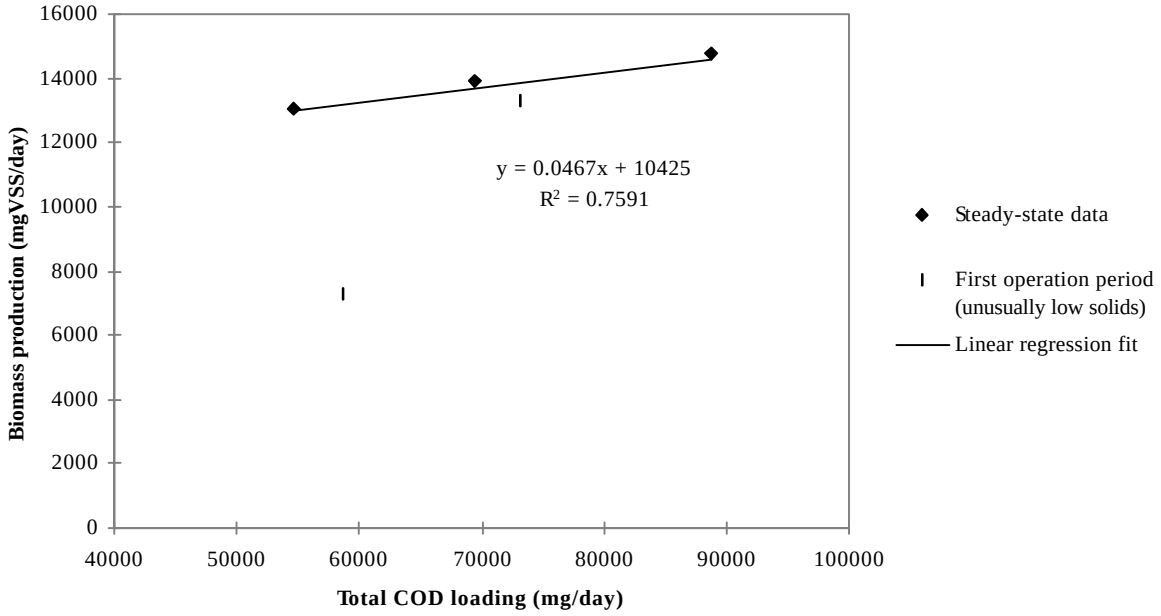

Figure 13. Secondary effluent treatment train: Estimating biomass production as a function of the total COD loading (including both the influent and methanol addition).

Feasibility of high temperature nitrification of the secondary effluent

During the last phase of operation, the operating temperature of the main aerobic reactor was raised to the range of 40 to 47°C. The temperature effects are compared to the operation period preceding the temperature increase (operation period 5), as similar nitrogen, carbon, and hydraulic loadings were maintained. It is important to note that only the main aerobic reactor was kept in the critical temperature range of 45°C and above, and that the anoxic reactor and small aerobic reactor were maintained, on average, at only 37.9 and 36.7°C, respectively. Despite this fact, nitrification was lost from the system, although it was readily recovered when the temperature was decreased. However, the cooler zones may have been what allowed the nitrifying population to stay alive and resume nitrification once the temperature decreased again to ambient conditions. Denitrification remained complete, but very small amounts of nitrates were entering the anoxic reactor (due to the greatly reduced nitrification activity), and the anoxic reactor was only kept in the temperature range of 33.3 to 44.5°C.

Despite the cooler temperatures in the other zones of the system, and the fact that the nitrifying biomass was added to the system twice during the eighteen-day high temperature study, nitrification could not be sustained by the system at the loading rate tested, and biosolids were rapidly declining toward zero (see Figure 14). Effluent SKN concentrations rose from a range of 0.8 to 1.1 mg-N/L during steady state operation in period 5 to concentrations as high as 25.7 during the high temperature study (see Table 9). Note that the effluent concentration climbed from 1.0 to 12.6 mg-N/L during the first four days, indicating that nitrification activity was definitely occurring for the first few days before it was apparently eliminated (see discussion of nitrification estimates below). The high temperatures were apparently severely inhibitory to nitrification, as the addition of nitrifying biomass on August 31 and September 8 did not result in a decrease in effluent SKN concentrations measured on September 2 and September 9. Although the temperature did not rise to 45^{o}C and above until September 7, the apparent dying off of nitrification within the first five days indicates that the lower temperatures (ranging from 37.9 to 44.8^{o}C) were sufficiently hot enough to inhibit nitrification. Similar trends were observed regarding the effluent ammonia concentrations, with a steady state effluent ammonia concentration of zero during operation period 5 for all days, rising to values as high as 25.5 mg-N/L during the high temperature study (see Table 9). During the first four days of the high temperature study, the effluent ammonia concentration steadily rose from zero to 12.0 mg-N/L. Effluent nitrate concentrations remained near or at zero during the high temperature study, indicating that denitrification remained, although it is important to note that the anoxic reactor was not kept as hot (with an average temperature of only 37.9^{o}C during the high temperature study), and the nitrate loading was drastically reduced due to the decreased nitrification. It was determined that the rising ammonia and SKN concentrations in the effluent were definitely the result of reduced nitrification by performing a nitrogen balance on the overall system.

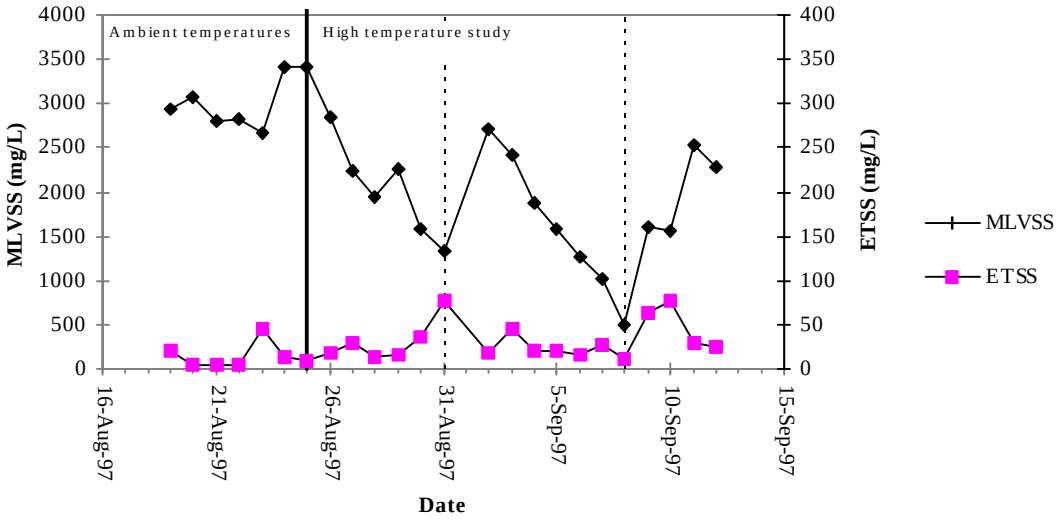

Figure 14. Secondary effluent treatment train: The effect of a sudden increase in temperature from approximately 27^{o}C to 40^{o}C and above on biosolids. The bold line corresponds to the last day of operating at ambient temperatures. The dashed lines indicate that biomass was added to the system at the end of that day.

Table 9. Secondary effluent treatment train: Comparison of overall performance at high temperatures to moderate temperature overall performance at similar loadings.

Date	Aerobic Temperature (^{o}C)	Anoxic Temperature (^{o}C)	Influent TKN (mg-N/L)	Effluent SKN (mg-N/L)	Influent NHx (mg-N/L)	Effluent NHx (mg-N/L)	Influent NOx (mg-N/L)	Effluent NOx (mg-N/L)	Total COD in (mg/L)	Effluent COD (mg/L)	% of TKN oxidized (%)
Operation period 5											
(moderate temperatures)											
19-Aug-97	29.0	28.2	34.6	1.1	34.6	0.0	5.8	0.6	492	54	71
20-Aug-97	28.0	26.8	37.2	0.8	30.7	0.0	3.2	0.0	513	50	72
21-Aug-97	27.2	26.7	34.8	1.1	29.9	0.0	2.3	0.0	463	54	72
22-Aug-97	27.0	26.5	33.2	1.1	26.8	0.0	2.4	0.0	450	49	71
Operation period 6									499	106	
(high temperatures)											
26-Aug-97	39.0	33.5	29.5	1.0	25.9	0.0	3.8	0.3	464	84	61
27-Aug-97	37.9	33.3	32.7	1.5	27.2	0.0	3.1	0.0	no data	50	78
28-Aug-97	41.0	35.6	30.8	7.3	26.1	6.1	1.3	0.0	462	82	65
29-Aug-97	39.2	35.8	31.8	12.6	26.3	12.0	6.2	0.0	446	60	38
2-Sep-97	42.1	36.1	24.6	14.5	24.1	14.5	5.5	0.0	419	61	5
3-Sep-97	41.0	36.3	30.2	19.7	28.3	19.7	5.3	0.0	393	56	9
4-Sep-97	43.8	35.3	41.7	25.7	39.1	25.5	4.1	0.0	393	61	24
5-Sep-97	44.0	37.9	32.0	19.3	29.0	18.4	3.7	0.0	399	78	25
8-Sep-97	45.4	39.0	26.3	19.3	25.4	19.3	6.8	0.2	396	108	22
9-Sep-97	47.1	44.5	25.8	20.1	23.4	20.1	6.2	0.0	394	171	-3
10-Sep-97	46.8	43.2	29.2	21.5	25.2	21.5	7.3	0.0	376	84	-1
11-Sep-97	46.0	39.8	bad data	17.2	bad data	16.5	bad data	0.0	388	89	5
12-Sep-97	23.3	24.2	25.5	15.7	22.9	15.7	1.4	0.0	425	215	28

Note: The temperature was reduced after data collection on 9/11/97.
Note: Hydraulic loadings were practically identical for both operation periods 5 & 6.

A balance on bioavailable nitrogen was performed by using the influent TKN, effluent SKN, and estimating the nitrogen used for growth, as described in the "Materials and Methods" portion of this thesis. From such a nitrogen balance, it was determined that approximately 72% of the bioavailable TKN entering the system was oxidized during the fifth operation period, while the percent of bioavailable TKN oxidized by the system during the high temperature study fell from a range of 61 to 78% during the first three days to values as low as zero during the remainder of the experiment (see Figure 15). It is important to note that the day after the temperature was decreased on September 12 (to 23.3^{o}C), the amount of bioavailable TKN oxidized rose from 5% the day before (aerobic temperature 46^{o}C) to 28%. Thus, while nitrification was obviously inhibited at the high temperatures, microbes able to perform nitrification had not been completely killed or washed out. This temperature decrease was performed only five days after the addition of two nitrifying sludges, one of which was acclimated to similarly high temperatures. Thus, it can only be said that nitrification could resume after five days of the high aerobic temperatures. This is significant, however, because this indicates that exposure to high temperatures in the aerobic reactor did not kill all nitrifying organisms. No data was collected to determine if the apparent cessation of nitrification was due to inactivation of nitrifying enzymes, denaturing of nitrifying enzymes, or the cessation of the production of nitrifying enzymes, but the organisms responsible were obviously not all killed off by the high temperature exposure, or nitrification could not have resumed immediately upon decreasing the temperature. Because nitrification did appear to occur at least partially for a few days after the increase in temperature, it seems most logical to conclude that the nitrifying enzymes remained intact and functioning at the high temperatures, and that it is the production of nitrifying enzymes which ceases at high temperatures (Randall, 1999). It is possible, of course, that the enzymes are actually denaturing faster than they are being produced at the higher temperatures, or that the enzymes are functioning exocellularly from lysed cells, which also might result in a tapering off of nitrification.

Not enough data was collected to determine if what little nitrification there was occurred in the main aerobic reactor or in other, cooler, zones. A nitrogen balance could not be performed over just the main aerobic reactor because there was no way to determine where growth, i.e., nitrogen assimilation, was occurring in the system. Additionally, there was definitely some denitrification occurring in the main aerobic reactor during the high temperature study, as indicated by the nitrate concentration leaving the aerobic reactor actually being lower than the nitrate concentration entering the reactor on occasion (see Table 10). Nitrification within the main aerobic reactor was obviously inhibited, however. This is obviously true because the ammonia concentrations exiting the aerobic reactor increased from zero during the fifth operation period to the range of 8.6 to 29.7 mg-N/L during the high temperature study (see Table 10). Additionally, the nitrates exiting the aerobic reactor decreased from an average of 20.7 mg/L during operation period 5 to the range of 0.5 to 10.2 mg/L during the high temperature study. Note that, from the nitrate and ammonia concentrations exiting the main aerobic reactor, nitrification appeared to be inhibited

by day 3 of the high temperature study (August 28), but the effluent ammonia and SKN concentrations, as well as the percent bioavailable TKN oxidized estimated on the same day for the overall system, suggest the system was not yet nitrification inhibited (see Tables 9 and 10). This is strong evidence that at least some nitrification was being carried out in the other, cooler zones, and could very well be the reason the nitrifying population was able to stay alive and resume nitrification upon reducing the temperature.

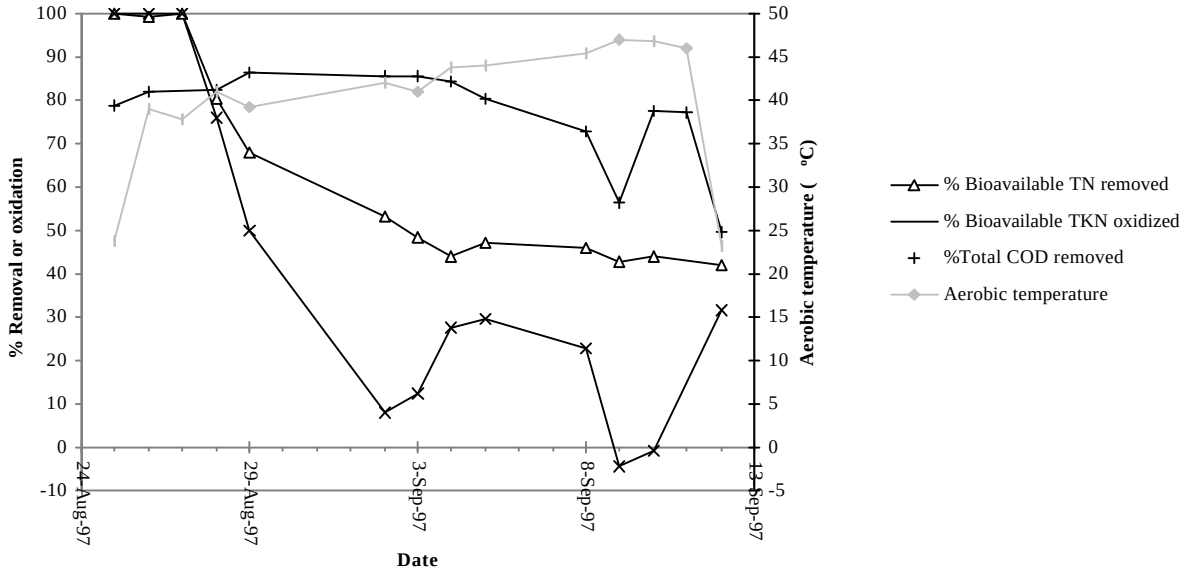

Figure 15. Secondary effluent treatment train: The effect of large changes in temperature on percent removals by a nitrifying/denitrifying sludge acclimated to moderate temperatures.

Table 10. Secondary effluent treatment train: Comparison of aerobic performance at high temperatures to moderate temperature aerobic performance at similar loadings.

Date	Aerobic Temperature (^{o}C)	TKN in (mg-N/L)	SKN out (mg-N/L)	NHx in (mg-N/L)	NHx out (mg-N/L)	NOx in (mg-N/L)	NOx out (mg-N/L)	COD consumed (mg/day)
Operation period 5 (moderate temperatures)								
19-Aug-97	29.0	22.6	2.4	22.6	0.0	4.0	21.5	27233
21-Aug-97	27.2	22.9	0.0	19.3	0.0	1.5	20.0	24935
Operation period 6 (high temperatures)								
28-Aug-97	41.0	22.9	9.4	19.3	8.6	0.8	10.2	21810
4-Sep-97	43.8	36.2	29.7	34.5	29.7	2.7	3.3	7897
9-Sep-97	47.1	23.2	21.9	22.3	21.9	4.1	1.2	14566
10-Sep-97	46.8	26.2	22.9	23.9	22.9	4.8	0.6	-5001
11-Sep-97	46.0	23.5	20.1	20.7	20.1	3.6	0.6	-18130
12-Sep-97	23.3	21.7	18.8	20.2	18.8	0.8	0.5	20281

Note: The temperature was reduced after data collection on 9/11/97.

Note: Hydraulic loadings were practically identical for both operation periods 5 & 6.

Heterotrophic activity in the aerobic reactor was also apparently inhibited by the high temperatures. Because COD consumption can be thought of strictly in terms of the quantity of COD removed, there was no concern whether the COD consumption was for energy production or assimilation (growth), and a COD balance could thus be performed on the main aerobic reactor. The COD balance on the aerobic reactor indicated that, on average, approximately 26,000 mg/day of COD were consumed in the main aerobic reactor during the fifth operation period. During the high temperature study, the amount of COD consumed in the main aerobic reactor fell from 21,528 mg/day on the third day of the high temperature study (August 28) to much lower values, and even negative values (implying COD was released in the main aerobic reactor, possibly due to cell lysis) (see Table 10). The overall consumption of COD was not affected as badly however, indicating that heterotrophic activity was able to continue in the cooler zones, i.e., anoxic reactor, reaeration basin, and clarifier. The overall percent of COD consumed only dropped from an average of 89% during operation period 5 (with the remaining COD being recalcitrant), to a range of 49 to 86% during the high temperature study (see Figure 15). The effluent soluble COD rose from a steady state average of 52 mg/L to a range of 50 to 215 mg/L during the high temperature study, which was generally lower than the influent COD to the system (not including the methanol addition which nearly tripled the total amount of COD entering the system).

Although there is only one data point, it is interesting to note that the effluent COD actually increased after the temperature decreased (see Table 9). There is not enough data to draw any significant conclusion, but this may be indicative of heterotrophs acclimating to the higher temperatures and being inhibited by the sudden decrease in temperature, while the autotrophic population was being suppressed by the high temperatures and quickly able to resume energy production upon relief of the high temperature stress.

Results and Discussion: Primary Effluent Treatment Train

During testing, the primary effluent of HRWTF generally had a COD in the range of 480 to 690 mg/L and a TKN in the range of 29 to 51 mg-N/L. Prior to testing of the bench scale design, it had been decided that an A2/O process was likely to yield successful nitrification and denitrification of the wastewater. Thus, the effluent from the primary clarifiers of HRWTF served as the influent to a single sludge process comprised of an anaerobic basin followed by an anoxic basin, and then an aerobic basin, with nitrate recirculation from the aerobic reactor to the anoxic reactor, and the RAS being ducted from the final clarifier to the anaerobic reactor. The system was operated both with and without methanol addition, with methanol being added directly to the anoxic reactor. Given the apparent slow acclimation to methanol as a carbon source, it was decided later that methanol addition periods were not long enough to determine if methanol addition truly would have enhanced denitrification, although data collected suggest it did not.

Similar to the secondary effluent treatment train, the sludge age was held relatively constant, but at 12 days instead of 6 days, for the entire experiment. Thus, unless otherwise indicated, all steady state data from all operation periods could be compared. As with the secondary effluent treatment train, the carbon and nitrogen concentrations of the wastewater tended to fluctuate, and the carbon and nitrogen loading was increased by increasing the hydraulic loading. In this manner, the loading, for the most part, was increased in time throughout the experiment. Also similar to the secondary effluent treatment train was the fact that operating conditions were changed relatively quickly, and true steady states were never reached. Quasi-steady states were delineated based upon relatively constant trends observed with respect to MLVSS, the SRT, hydraulic loading, COD and nitrogen loading, and COD and nitrogen removal performance. Because recirculation rates were also varied for this treatment train, this was one additional factor to consider when delineating steady state operation periods. Also as observed for the secondary effluent treatment train, effluent quality tended to reach steady state concentrations before the MLVSS, meaning the MLVSS played a large role in defining steady state. Selection based upon similar MLVSS concentrations and stable SRTs during similar operating conditions is depicted in Figure 16. From consideration of all pertinent factors, eight (8) steady state operation periods were delineated. The reasons for the separate delineations of each operation period are described below in Table 11, and Table 12 provides overall characteristics for each operation period.

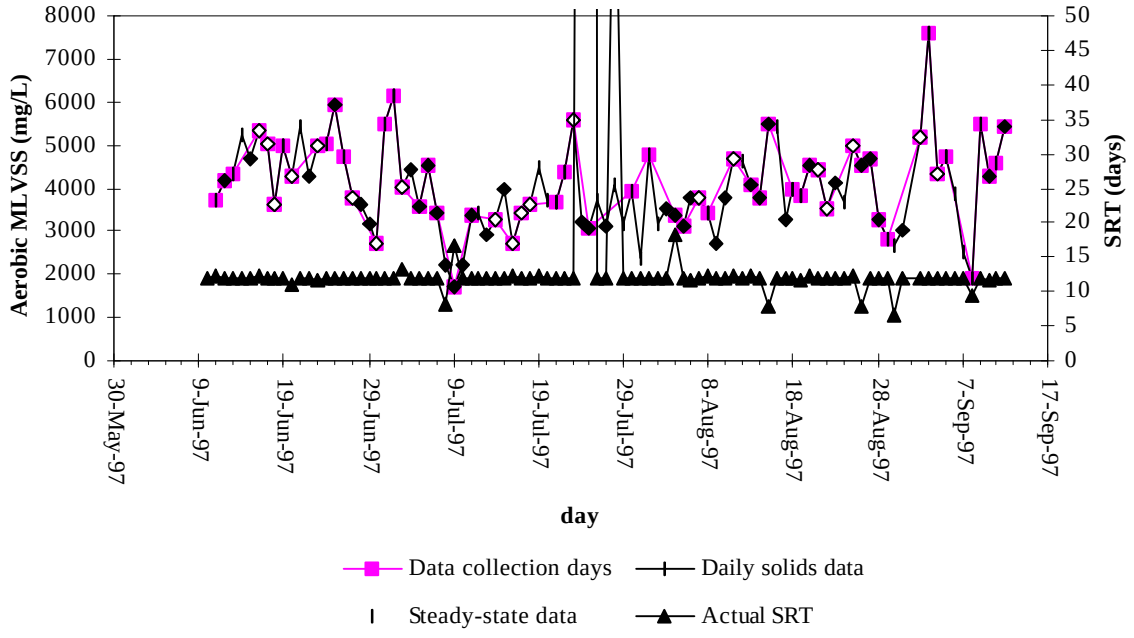

Figure 16. Primary effluent treatment train: Using the MLVSS and SRT to help delineate steady state operation periods. The bold lines indicate the last day of a steady state operation period.

Table 11. Characterization and delineation of separate operation periods for the primary effluent treatment train.

Operation period	Steady state dates	Reason for separate delineation
1	6/16-6/23	• Reason for start: Stabilized performance after startup on 5/19. • Reason for end: Began methanol addition 6/24.
2	6/27-6/30	• Reason for start: Performance stabilized by 6/27. • Reason for end: Increased RAS and NR % of flows at 16:00 on 6/30 to 90% and 300%, respectively.
3	7/2-7/3	• Reason for start: Performance stabilized by 7/2. • Reason for end: Increased influent flow rate after 7/3 samples. Reduced RAS and NR back to original 65% and 250%, respectively.

4	7/14-7/23	• Conditions delaying acclimation:
		  • Stopped methanol addition after 7/3.
		  • Problems with final clarifier and pumps 7/6, 7/9, and 7/11.
		• Reason for start: Performance stabilized by 7/14.
		• Reason for end: Pump failure evening of 7/23.

5	8/5-8/11	• Conditions delaying acclimation:
		  • Increased flow rates on morning of 7/29.
		  • Problem with final clarifier 8/3.
		• Reason for start: Performance stabilized by 8/5.
		• Reason for end: COD of wastewater doubled 8/13.

6	8/18-8/19	• Reason for start: Wastewater and performance stabilized by 8/18. • Reason for end: Decreased all flows proportionally after 8/19 sample.
7	8/21-8/25	• Reason for start: Performance stabilized by 8/21. • Reason for end: Began methanol addition 8/26.
8	9/2-9/5	• Reason for start: Performance stabilized by 9/2. • Reason for end: Nitrification/denitrification under normal conditions study terminated.

It was determined that the primary effluent of HRWTF could be successfully nitrified completely at low enough loadings with the A2/O treatment train tested, but denitrification was never completed down to zero nitrates, with steady state effluent NO_x concentrations ranging from 1.9 to 7.6 mg-N/L after the first 35 days of operation (see Appendix 6). Denitrification down to 8 mg-N/L of effluent nitrates or less was regarded as complete denitrification for this system for three reasons: this was a post-nitrification process (which makes some effluent nitrates inevitable), the nitrates exiting the anoxic reactor were generally zero, and after acclimation, effluent nitrates were always below 8 mg-N/L despite increases in loading. Complete nitrification was defined as occurring when no ammonia was detected in the effluent. Unlike the secondary effluent treatment train, occasional low concentrations of ammonia (1.1 to 5.4 mg-N/L; see Appendix 6) appeared in the effluent even during the quasi-steady state periods when zero ammonia was generally detected. This was possibly an indication that the performance was slightly less stable, but may have been due merely to insufficient time being allowed for acclimation, as operating conditions were generally changed within 12 days. Effluent COD concentrations remained relatively constant throughout the study, despite the increased loading, indicating that the COD in the effluent was, for all intents and purposes, recalcitrant, and that COD removal could be regarded as complete for all loadings tested. As with the secondary effluent treatment train, nitrogen and COD balances were performed on the overall system and individual reactors, and performance with respect to the above mentioned parameters is discussed below.

Feasibility of nitrification of the Primary Effluent at moderate temperatures

Complete nitrification was possible for all operation periods except operation period 6, where the average steady state effluent ammonia concentration was 10.0 mg-N/L (see Table 12 for this value and all other values referred to in this discussion). Both operation periods 5 and 6 were at the highest hydraulic loading rate used, and methanol addition was not used for either operation period, but complete nitrification was only observed during operation period 5. The difference between the two operation periods was a sudden increase in the influent COD from a concentration of 500 mg/L on August 11 (the last day of operation period 5; see Appendix 6) to a

Table 12. Primary effluent treatment train: Overall performance and loading characteristics.

Steady-state operation period	1 6/16-6/23		2 6/27-6/30		3 7/2/97-7/3/97		4 7/14-7/23		5 8/5-8/11		6 8/18-8/19		7 8/21-8/25		8 9/2-9/5	
SRT	**11.8**	8	**12.0**	4	**12.6**	2	**12.0**	10	**12.0**	7	**11.8**	2	**12.0**	5	**12.0**	4
(days)	0.3		0.0		n/a		0.1		0.1		n/a		0.1		0.0	
Total HRT	**9.5**	8	**9.3**	4	**8.7**	2	**6.8**	10	**5.4**	7	**5.5**	2	**6.9**	5	**7.0**	4
(hours)	0.3		0.2		n/a		0.1		0.1		n/a		0.1		0.1	
Influent flow rate	**102.6**	8	**102.6**	4	**110.2**	2	**136.8**	10	**178.4**	7	**175.7**	2	**137.4**	5	**136.8**	4
(L/day)	5		2		n/a		6		3		n/a		2		1	
RAS % of influent flow rate	**65**	8	**66**	4	**88**	2	**66**	10	**64**	7	**52**	2	**67**	5	**68**	4
(%)	2		1		n/a		2		1		n/a		3		2	
NR % of influent flow rate	**245**	8	**264**	4	**338**	2	**252**	10	**241**	7	**199**	2	**257**	5	**263**	4
(%)	5		26		n/a		4		5		n/a		3		3	
MLVSS	**4736**	8	**3320**	4	**5095**	2	**3887**	10	**3601**	7	**3910**	2	**4146**	5	**5460**	4
(mg/L)	632		492		n/a		785		613		n/a		592		1467	
ETSS	**23**	8	**17**	4	**27**	2	**10**	10	**10**	7	**11**	2	**3**	5	**9**	4
(mg/L)	12		6		n/a		7		6		n/a		2		1	
Specific growth rate	**0.085**	6	**0.083**	2	**0.080**	2	**0.083**	7	**0.083**	3	**0.085**	3	**0.083**	3	**0.084**	4
(1/day)	0.003		n/a		n/a		0.001		0.001		0.001		0.001		0.000	
TKN influent	**3940**	6	**4626**	2	**5282**	2	**5251**	7	**6499**	3	**6679**	2	**4827**	2	**4576**	3
(mg-N/day)	835		n/a		n/a		221		1898		1823		n/a		656	
SKN effluent	**244**	6	**431**	2	**498**	2	**192**	7	**351**	3	**1858**	2	**268**	2	**371**	3
(mg-N/day)	69		n/a		n/a		61		282		2043		n/a		98	
NHx influent	**3259**	6	**4340**	2	**4958**	2	**4998**	7	**6362**	3	**5959**	2	**3974**	2	**4288**	3
(mg-N/day)	335		n/a		n/a		328		1982		805		n/a		671	
NHx effluent	**32**	6	**216**	2	**314**	2	**21**	7	**177**	3	**1763**	2	**0**	2	**236**	3
(mg-N/day)	79		n/a		n/a		55		307		2178		n/a		82	
NOx influent	**431**	6	**1335**	2	**1111**	2	**677**	7	**1808**	3	**1473**	2	**240**	2	**471**	3
(mg-N/day)	347		n/a		n/a		402		297		n/a		n/a		273	
NOx effluent	**1000**	6	**1135**	2	**632**	2	**856**	7	**1009**	3	**634**	2	**612**	2	**492**	3
(mg-N/day)	451		n/a		n/a		138		317		n/a		n/a		211	
COD influent	**63319**	6	**58740**	2	**64087**	2	**69997**	7	**90926**	3	**111908**	2	**80478**	2	**71498**	3
(mg/day)	9309		n/a		n/a		5877		7225		n/a		n/a		3768	
Methanol addition?	no		yes		yes		no		no		no		no		yes	
Total COD in	**63319**	6	**62330**	2	**67677**	2	**69997**	7	**90926**	3	**111908**	3	**80478**	3	**100345**	4
(mg/day)	9309		n/a		n/a		5877		7225		6460		4215		4930	
COD effluent	**6202**	6	**5085**	2	**5929**	2	**6633**	7	**7250**	3	**10892**	3	**7402**	3	**6883**	4
(mg/day)	1051		n/a		n/a		1131		2194		0		1210		1586	
TKN influent	**38.0**	6	**44.6**	2	**47.3**	2	**37.8**	7	**36.8**	3	**38.0**	2	**35.3**	2	**33.6**	3
(mg-N/L)	6.4		n/a		n/a		1.3		10.0		n/a		n/a		5.0	
SKN effluent	**2.4**	6	**4.2**	2	**4.4**	2	**1.4**	7	**2.0**	3	**10.6**	2	**2.0**	2	**2.7**	3
(mg-N/L)	0.6		n/a		n/a		0.4		1.5		n/a		n/a		0.7	
NHx influent	**31.6**	6	**41.9**	2	**44.3**	2	**35.9**	7	**36.0**	3	**33.9**	2	**29.1**	2	**31.5**	3
(mg-N/L)	1.7		n/a		n/a		2.3		10.4		n/a		n/a		5.1	
NHx effluent	**0.3**	6	**2.1**	2	**2.7**	2	**0.2**	7	**1.0**	3	**10.0**	2	**0.0**	2	**1.7**	3
(mg-N/L)	0.8		n/a		n/a		0.4		1.7		n/a		n/a		0.6	
NOx influent	**4.1**	6	**12.8**	2	**9.9**	2	**4.9**	7	**10.3**	3	**8.4**	2	**1.8**	2	**3.4**	3
(mg-N/L)	3.3		n/a		n/a		2.9		1.9		n/a		n/a		2.0	
NOx effluent	**9.7**	6	**11.0**	2	**5.7**	2	**6.2**	7	**5.7**	3	**3.6**	2	**4.5**	2	**3.6**	3
(mg-N/L)	4.3		n/a		n/a		1.0		1.7		n/a		n/a		1.5	
COD influent	**614**	6	**571**	2	**579**	2	**503**	7	**516**	3	**637**	3	**584**	3	**523**	4
(mg/L)	75		n/a		n/a		43		43		37		23		23	
COD effluent	**60**	6	**50**	2	**54**	2	**48**	7	**41**	3	**62**	3	**54**	3	**50**	4
(mg/L)	9		n/a		n/a		8		12		0		8		11	
% of bioavailable TKN	**45**	6	**58**	2	**56**	2	**69**	7	**72**	3	**43**	3	**65**	3	**42**	4
oxidized (nitrified)	16		n/a		n/a		9		9		48		4		18	
Overall observed specific	**0.015**	6	**0.037**	2	**0.020**	2	**0.036**	7	**0.049**	3	**0.031**	3	**0.028**	3	**0.014**	4
nitrification rate	0.010		n/a		n/a		0.012		0.028		0.037		0.005		0.011	
(mg-N/mgVSS/day)	(NHx limited)		(NHx limited)		(NHx limited)		(NHx limited)		(NHx limited)				(NHx limited)		(NHx limited)	

Note: Top value indicates mean value during operation period. The lower value indicates the standard deviation. The value to the right indicates the number of samples in the data set.

concentration of 1090 mg/L on August 13, with an average influent COD of 637 mg/L during operation period 6. The strong COD loading resulted in effluent ammonia concentrations over 9.9 mg-N/L until August 19 (see Appendix 6). It was observed that the percent of bioavailable TKN oxidized dropped from an average of 72% to 43% between operation period 5 and 6. While the effluent COD was higher during operation period 6 than operation period 5, the amount of COD removed was actually higher during operation period 6 (taking the difference of the total COD entering the system and the COD of the effluent), indicating that heterotrophic activity was increasing while autotrophic activity was decreasing. Thus, it appears that the increase in the COD loading resulted in outcompetition for oxygen by the heterotrophs.

Because the COD spike observed during operation period 6 was the only COD spike observed during the experiment, it is not known whether a reduction in the hydraulic loading was, in fact, helpful. The loading was reduced at the time of operation because ammonia breakthrough was witnessed, but it was not realized that the breakthrough was due to high COD loading until after termination of the project when all the data were analyzed together. Because the COD spike lasted at least six days, a flow diversion or equalization approach is probably not feasible. Thus, operating at lower loadings substantially below the sludge's maximum capability is probably the only solution which would allow at least partial dampening of such spikes.

Therefore, in terms of achieving consistent and complete nitrification, a lower loading such as that of the second-highest hydraulic loading tested, appears to be the maximum acceptable loading rate, although no COD spikes were observed during the lower loadings to verify that better recovery was likely. This loading rate corresponded to operation periods 4, 7, and 8. Effluent ammonia concentrations during these operation periods ranged from 0.0 to 1.4 mg-N/L (see Appendix 6). Steady state average effluent ammonia concentrations were 0.2, 0.0, and 1.7 mg-N/L, respectively (see Table 12). The higher effluent ammonia concentrations observed during operation period 8 were also associated with higher total COD loadings to the system (in terms of mgCOD/day). Although operation period 8 was associated with the highest total COD loading, the influent COD, i.e. the COD of the HRWTF primary effluent, was similar to the other two operation periods. The difference in the COD was due to methanol addition. Thus, it is not clear whether the slightly higher effluent ammonia concentration was associated with simple outcompetition for oxygen by heterotrophs, or acclimation to methanol. There is evidence suggesting the incomplete nitrification was due to methanol toxicity, however.

Operation period 5 had a similar COD loading to operation period 8, with average steady state total COD loadings (including the addition of methanol) of 90,926 and 100,345 mg/day, respectively; but no methanol was added during operation period 5. Additionally, operation period 5 actually had a higher ammonia loading than operation period 8, with ammonia loadings of 6362 and 4288 mg-N/day, respectively (see Table 12). Yet, no ammonia was detected in the final effluent on two of the three steady state days during operation period 5, while the effluent ammonia concentrations during operation period 8 were within the range of 1.4 and 2.4 mg-N/L (see Appendix 6). Although methanol concentrations were not measured in the aerobic reactor, there was bioavailable COD entering the aerobic reactor during operation period 8, as indicated by the difference in the COD entering and exiting the reactor, which could have contained methanol (see Table 13). In addition, the recorded COD values are known to be biased low, indicating that even more COD, possibly in the form of methanol, was present than was measured. Since it has been shown that short-chain alcohols such as methanol are toxic to *Nitrosomonas* (Azevedo *et al.*, 1995 citing Hooper and Terry, 1973), it is quite plausible that the increase in the effluent ammonia during operation period 8 was due not simply to outcompetition for oxygen by the heterotrophs, but to acclimation to the methanol addition. Thus, nitrification at the second highest hydraulic loading (approximately 140 L/day) appears to be sufficiently complete and stable if no methanol is added directly to the system. Adequate acclimation time should be allowed to determine if methanol addition is a viable option for denitrification of the primary effluent, but there appeared to be enough COD present to denitrify the wastewater without methanol addition (see the denitrification discussion below).

As with the secondary effluent treatment train, nitrogen balances on both the overall system and the aerobic reactor were performed in order to estimate specific nitrification rates. The overall balance yielded higher estimates and a better correlation to the F:M loading. From the balance on the total system, the maximum observed overall specific nitrification rate was determined to be 0.078 mgN/mgVSS/day, which did correspond to the highest F:M loading. From the plot of observed overall specific nitrification rates depicted in Figure 17, it appears that the true maximum rate was not reached, because the trend still appeared to be first order and increasing with respect to F:M loading. For the sake of discussion, however, the maximum rate witnessed will be assumed to be the maximum specific nitrification rate of the sludge under the implemented operating conditions.

Table 13. Primary effluent treatment train: Aerobic reactor performance and loading.

Steady-state operation period	**1** 6/16-6/23	**2** 6/27-6/30	**3** 7/2/97-7/3/97	**4** 7/14-7/23	**5** 8/5-8/11	**6** 8/18-8/19	**7** 8/21-8/25*	**8** 9/2-9/5
Aerobic HRT (hours)	1.6	1.4	1.3	1.1	0.9	0.9	1.1	1.1
SKN in (mg-N/day)	6110	6468	3477	5148	5861	12926	11641	4522
SKN out (mg-N/day)	818	1754	1559	538	1240	8436	755	903
Bioavailable SKN in (mg-N/day)	5853	6288	3306	5023	5433	12926	11568	4451
SKN in - SKN out (mg-N/day)	5292	4715	1918	4610	4621	4490	10886	3619
SKN used for growth by total system (mg-N/day)	1747	1074	2477	1661	1200	1616	1795	3036
SKN available for oxidation (mg-N/day)	4106	5214	829	3363	4232	11310	9772	1414
Estimated minimum SKN nitrified (mg-N/day)	3545	3641	-559	2949	3421	2874	9091	582
Measured NOx production (mg-N/day)	4970	5062	3428	3946	5208	3797	2635	3015
Estimated minimum specific nitrification rate (mgSKN-N/mgVSS/day)	0.035	0.049	-0.003	0.029	0.036	0.026	0.073	0.003
Measured specific NOx production rate (mgNOx-N/mgVSS/day)	0.046	0.068	0.020	0.036	0.055	0.035	0.021	0.014
% bioavailable SKN removed	88	75	58	92	83	35	94	81
Estimated minimum % bioavailable SKN oxidized	44	58	-17	57	55	22	79	13
Estimated % oxidizable SKN nitrified	67	70	-67	86	73	25	93	41
SKN in (mg-N/L)	14.3	15.1	7.0	8.9	8.1	17.3	20.1	7.6
SKN out (mg-N/L)	1.9	4.1	3.1	0.9	1.7	11.3	1.3	1.5
NHx out (mg-N/L)	0.0	2.2	0.0	0.0	0.0	11.3	0.0	0.0
COD in (mg/day)	51749	30200	24491	49044	57521	70862	43995	41165
COD out (mg/day)	25863	19846	23992	28980	43322	35058	24892	27443
COD in - COD out (mg/day)	25886	10354	500	20064	14199	35804	19103	13722
COD in (mg/L)	122	70	49	85	79	95	76	69
COD out (mg/L)	61	46	48	50	60	47	43	46

Note: There is only one data point per operation period for balances on the aerobic reactor, with the exception of operation periods 1 and 4 which had 2 data points. Thus, standard deviations are not applicable, and the numbers of data points are not listed in the table for the balance on the aerobic reactor.

* No data samples were collected directly from the reactors during operation period 7. Thus, data collected on August 20 is used, as this was during the same operating conditions but regarded as in a transitory state.

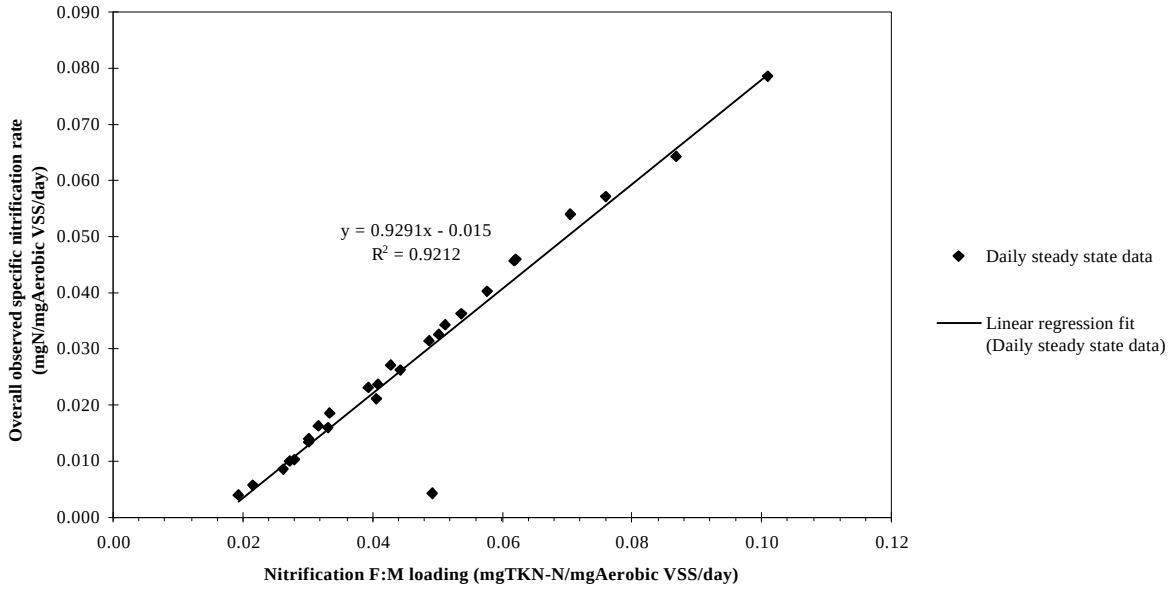

Figure 17. Primary effluent treatment train: Overall observed specific nitrification rate as a function of the F:M loading.

Feasibility of denitrification of the Primary Effluent at moderate temperatures

With the exception of the first two operation periods, denitrification was carried out consistently to the same degree at all other loadings tested. Steady state effluent NO_x concentrations ranged from 1.9 to 7.6 mg-N/L throughout operation periods 3 through 8, with steady state average effluent concentrations ranging from 3.6 to 6.2 mg-N/L (see Table 12 and Appendix 6). Because some effluent nitrates are inevitable for a post-nitrification system, and because steady state soluble effluent nitrate concentrations were in the range of 1.9 to 7.6 (see Appendix 6) for all loadings tested after operation period 2, i.e., after acclimation, removal of nitrates in the final effluent below 8 mg-N/L was regarded as complete denitrification for this system.

Although there is little data, with only one or two data points per operation period, mass balances on the anoxic reactor were performed to characterize denitrification performance. Complete denitrification with respect to the anoxic reactor was defined as zero nitrates being detected exiting the reactor. While complete denitrification appeared to occur for the overall system for all operation periods but operation periods 1 and 2 (see Table 12), complete denitrification according to the mass balance performed on the anoxic reactor occurred for all operation periods except operation periods 2, 3, and 5 (see Table 14). Although only 0.7 mg-N/L of nitrates were measured exiting the anoxic reactor during operation period 5, this was considered to be a sign that the maximum capacity of the sludge to denitrify the wastewater was, in fact, being reached (to be discussed below).

The percent of denitrification occurring in the anoxic reactor correlated well with the COD:NO_x loading to the anoxic reactor, as depicted in Figure 18. With the exception of operation period 3, the COD exiting the anoxic reactor was always greater than the COD of the final effluent. Therefore, it would seem that only operation period 3 was COD limited. However, as shown in Figure 18, complete denitrification appeared to require a COD:NO_x loading ratio of 15; and thus, both operation periods 2 and 3 were apparently COD limited. The data point that falls below the curve corresponds to operation period 5, which had the shortest HRT and highest nitrate loading of all the operation periods. It is peculiar that the lowest percent denitrification occurred in the anoxic reactor during operation period 3, yet the final effluent indicated that complete denitrification was occurring during that operation period (see Tables 12 and 14). With a COD:NO_x ratio of 9.6 during operation period 3, it appears that denitrification in the anoxic reactor was COD limited, and the COD exiting the anoxic reactor was actually measured to be lower than the COD of the final effluent. It was determined that the difference was that a great deal of nitrates were actually being removed in the anaerobic reactor on the day data was collected during operation period 3 (July 2; see Appendix 7). Significant nitrate removal in the anaerobic reactor was actually a common occurrence (see Appendix 7).

Table 14. Primary effluent treatment train: Anoxic reactor performance and loading.

Steady-state operation period	**1** 6/16-6/23	**2** 6/27-6/30	**3** 7/2/97-7/3/97	**4** 7/14-7/23	**5** 8/5-8/11	**6** 8/18-8/19	**7** 8/21-8/25*	**8** 9/2-9/5
Anoxic HRT (hours)	0.49	0.42	0.41	0.35	0.28	0.27	0.35	0.34
Methanol addition?	no	yes	yes	no	no	no	no	yes
Methanol loading to anoxic reactor (mgCOD/day)	0	3590	3590	0	0	0	0	28205
Total COD loading to anoxic reactor (mgCOD/day)	43599	42741	49565	68707	81161	78906	58091	154658
NOx loading to anoxic reactor (mg-N/day)	2955	4446	5178	2390	3456	2340	1606	1834
COD:NOx loading ratio to anoxic reactor	14.8	9.6	9.6	28.8	23.5	33.7	36.2	84.3
Amount of NOx exiting anoxic reactor (mg-N/day)	0	2369	4596	0	482	23	14	0
Concentration of NOx exiting anoxic reactor (mg-N/L)	0.0	5.5	9.3	0.0	0.7	0.0	0.0	0.0
Amount of COD exiting anoxic reactor (mg/day)	51749	30200	24491	49044	57521	70862	43995	41165
Concentration of COD exiting anoxic reactor (mg/L)	122	70	49	85	79	95	76	69
COD of final effluent (mg/L)	60	50	54	48	41	62	54	50
% NOx denitrified	100	47	11	100	91	99	99	100
Observed specific denitrification rate (mgNOx-N/mgVSS/day)	0.088	0.090	0.011	0.071	0.101	0.068	0.041	0.028

Note: There is only one data point per operation period for balances on the anoxic reactor, with the exception of operation periods 1 and 4 which had 2 data points. Thus, standard deviations are not applicable, and the numbers of data points are not listed in the table for the balance on the anoxic reactor.

* No data samples were collected directly from the reactors during operation period 7. Thus, data collected on August 20 is used, as this was during the same operating conditions but regarded as in a transitory state.

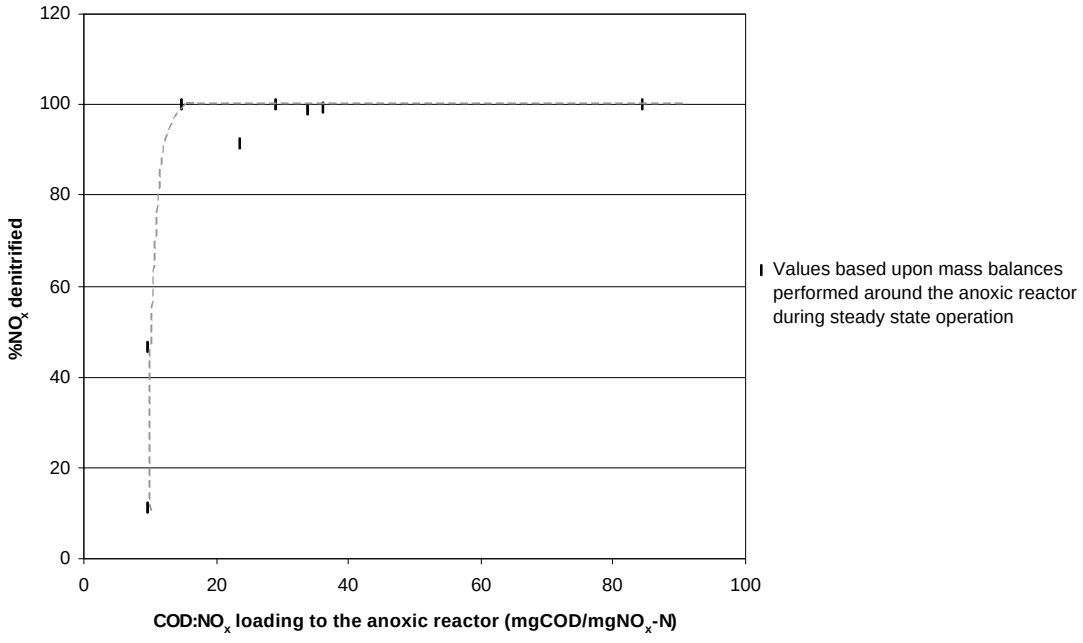

Figure 18. Primary effluent treatment train: Using the percent denitrification in the anoxic reactor as a function of the COD:NO_x loading ratio to the anoxic reactor to determine the critical COD:NO_x loading. The dashed curve represents the anticipated performance for this activated sludge and wastewater.

Methanol was added directly to the anoxic reactor, but only for operation periods 2, 3, and 8. Not enough methanol was added during operation periods 2 and 3 to prevent COD limitations in the anoxic reactor, but reasonable denitrification was observed in the overall system regardless, with steady state average effluent NO_x concentrations of 11.0 and 5.7 mg-N/L, respectively. No methanol was added during operation periods 1, 4, 5, 6, and 7, with average effluent NO_x concentrations being 9.7, 6.2, 5.7, 3.6, and 4.5 mg-N/L, respectively (see Table 12). Thus, methanol addition appeared to have no effect on the extent of denitrification. Although probably not enough time was ever allowed for acclimation to methanol as a carbon source for denitrification, it is doubtful that methanol addition was even necessary because the wastewater appeared to have enough COD of its own for denitrification. For instance, during operation periods 4, 5, 6, and 7, i.e. operation periods in which no methanol was added and the sludge appeared to be fully acclimated, no nitrates were ever detected exiting the anoxic reactor, and the COD exiting the reactor was in the range of 22 to 65 mg/L higher than the soluble COD of the final effluent (see Table 14 and Appendix 8). The only day on which denitrification was definitely COD limited, as indicated by the fact that the COD exiting the anoxic reactor was measured to be 10 mg/L lower than the final effluent and the fact that 9.3 mg-N/L of nitrates were escaping the anoxic reactor, was on July 2 (during operation period 3) when methanol was actually being added (see Appendices 6 and 8). Because there was this one occurrence of COD limitations, it should be accepted that occasional days of COD limitations for complete denitrification will occur, but the occurrence appears to be rare for this wastewater. This is of little concern with the A2/O configuration used to treat the primary effluent, however, because the anaerobic reactor often served as at least a partially anoxic reactor and was not COD limited. Thus, much of the nitrates that would exit the system are denitrified in the anaerobic reactor via the RAS.

As with the secondary effluent treatment train, the observed overall maximum specific denitrification rate was compared with the denitrification rate based upon a nitrogen balance performed on just the anoxic reactor. Also as with the secondary effluent system, the overall observed rates yielded higher values than the specific rate based upon just the individual reactor. Thus, discussion will be restricted to the overall observed specific denitrification rate. The overall observed specific denitrification rate was normalized to the biomass in just the anoxic reactor, and the F:M ratio was expressed in terms of the influent TKN loading normalized to the biomass in the anoxic reactor, which is shown in Figure 19. No correlation was apparent when comparing the observed specific denitrification rate to a COD-based F:M ratio.

As with the observed nitrification rates, the overall observed specific denitrification rate did not appear to reach a maximum, as indicated by a horizontal trend. For the sake of discussion, the maximum specific denitrification rate observed will be assumed to be the true maximum rate, which was determined to be 0.26 mgN/mgVSS/day.

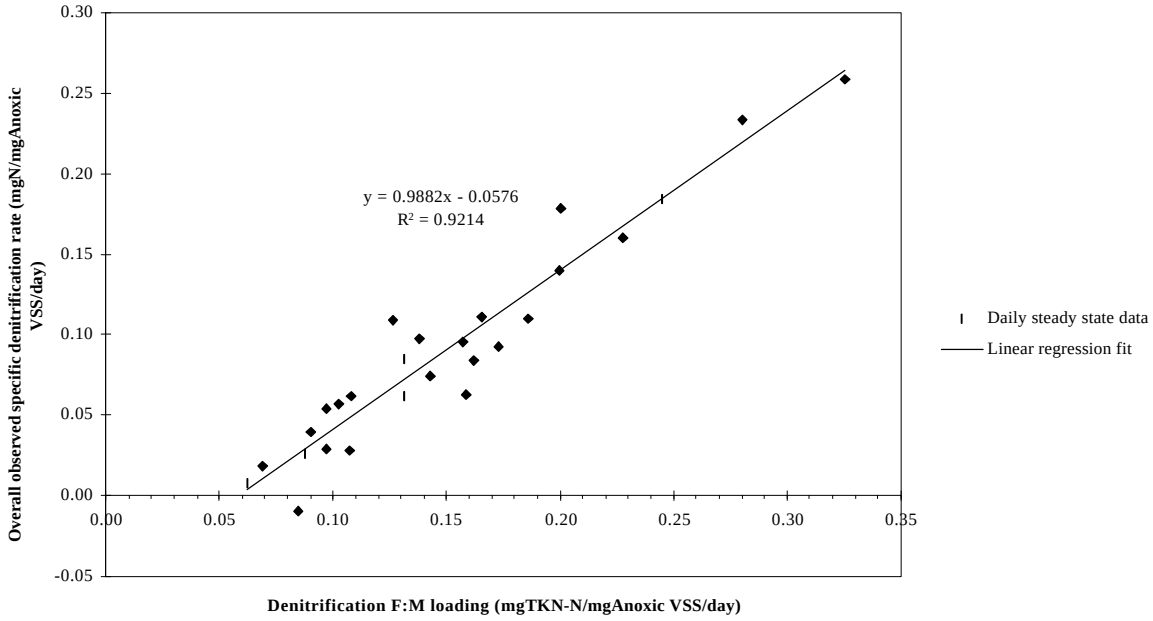

Figure 19. Primary effluent treatment train: Overall observed specific denitrification rate as a function of the F:M loading.

Other performance characteristics of the Primary Effluent treatment train at moderate temperatures

COD removal by the treatment train tested was essentially complete for all loadings tested. Steady state effluent COD concentrations were roughly the same for all operation periods (including the first two), with steady state soluble CODs in the effluent ranging from 32 to 78 mg/L (see Appendix 6). Even transitional days yielded effluent soluble CODs in a range similar to steady state days. Because the effluent COD was approximately the same for all total COD loadings (both with and without methanol addition), the COD of the effluent was regarded as recalcitrant. The fact that these values were similar to the final effluent of the secondary effluent treatment train lends further support to the belief that this last portion of soluble COD truly is very resistant to biodegradation (see Appendices 1 and 6). Because effluent COD concentrations were similar for all loadings tested, it is evident that the system was underloaded with respect to COD removal, and thus nitrification will be the dominant factor in designing the full scale system.

The ETSS of the system was consistently in the range of 1 to 25 mg/L with few outliers in any operation period (see Figure 20 and Appendix 10). Given the consistency of the ETSS for all operation periods, it can be concluded that the A2/O treatment of the primary effluent yields a very stable sludge in terms of effluent solids quality regardless of methanol addition, hydraulic loading, or the C:N ratio. Thus, determining an appropriate design for the treatment of this system will not be governed by the effluent solids quality.

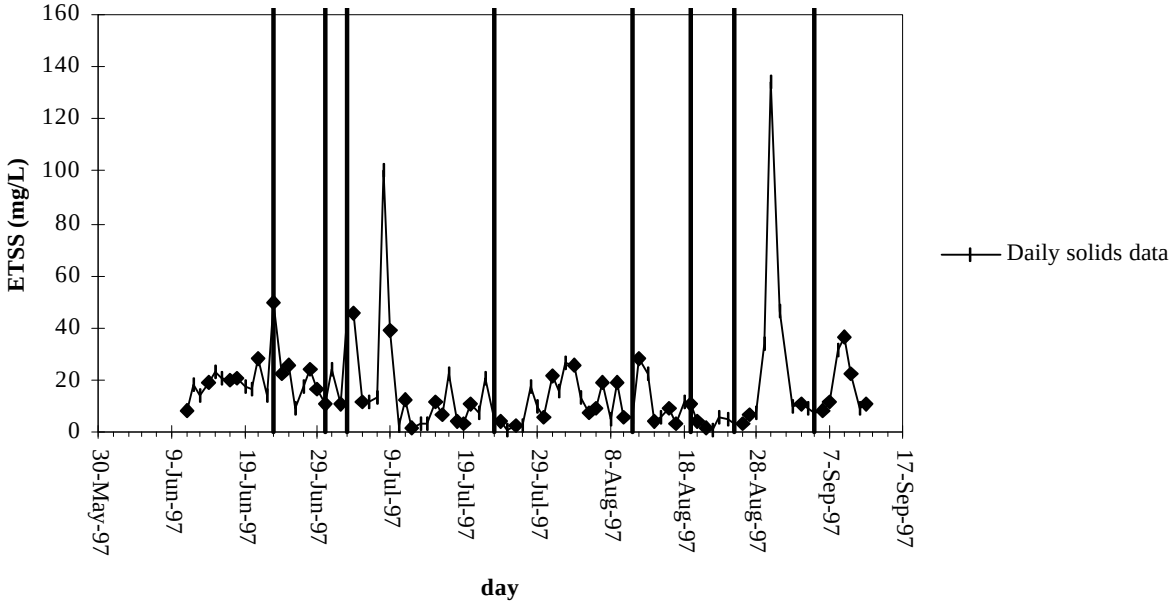

Figure 20. Primary effluent treatment train: Daily effluent suspended solids quality The bold lines indicate the last day of a steady state operation period.

Full scale design of the primary effluent treatment train

Because both denitrification and COD removal was complete (in terms of final effluent quality) for all loadings tested once acclimation was achieved, nitrification is the major limitation and consideration for design of a full sized treatment train. In addition to substantial ammonia breakthrough at the highest loading rate, nitrification was apparently slower to acclimate than denitrification and COD removal. Soluble effluent COD concentrations and soluble effluent nitrate concentrations generally appeared to be within the range of steady state concentrations within one to two days of a change in operating conditions, while effluent ammonia concentrations generally took an additional one to three days to reach steady state levels (see Appendix 6). Using a higher SRT might have produced

a sludge which acclimated to complete nitrification quicker, but experimentation with varying SRT was not performed. Estimating the full scale design will be based upon the conditions tested.

The system tested tended to operate at an MLVSS of approximately 3800 mg/L, which will thus be assumed for the full scale design. Assuming nitrification will be operated at 70% of the observed maximum specific activity as a safeguard against COD spikes, and denitrification will be operated at 80% of the observed maximum to provide a safety factor against occasional spikes in loading, a specific nitrification rate of 0.055 mgN/mgVSS/day and specific denitrification rate of 0.21 mgN/mgVSS/day will be assumed. The average TKN concentration of the primary effluent of HRWTF during this study was 37.9 mg-N/L. Scaling the hydraulic loading up to 50 MGD (189.25 million liters per day), an average TKN loading of 7.17×10^9 mg-N/day (15,800 lb-N/day) is expected. Complete nitrification at the assumed specific nitrification rate would thus require 1.30×10^{11} mg of VSS. Assuming a 3800 mg/L MLVSS, an aerobic basin volume of 3.43×10^7 L, or 9.07 million gallons would be required. Assuming complete nitrification and complete denitrification, in addition to the assumed denitrification rate of 0.21 mgN/mgVSS/day, an anoxic basin of 2.41 million gallons (8.88×10^6 L) would be required to denitrify the 7.17×10^9 mg-N/day with a 3800 mg/L MLVSS concentration.

The A2/O process was selected as the option to treat the primary effluent as opposed to merely an anoxic/aerobic process because the anaerobic reactor allows for the likelihood of detoxification of compounds which might be present in the primary effluent of the complex industrial wastewater of HRWTF, as well as the possible breakdown of complex organics which might be recalcitrant to anoxic and aerobic biodegradation. Because toxicity tests were not performed on an anoxic/aerobic treatment of the primary effluent, it is not be known if the anaerobic zone is necessary for such a function. The fact that soluble effluent COD concentrations for the primary effluent treatment train were approximately the same as those of the secondary effluent treatment train indicates that the COD exiting both systems was recalcitrant to anaerobic activity at the loading rates tested. Thus, although toxicity testing would be required to demonstrate if the anaerobic basin was unnecessary, the full scale design could possibly realize substantial savings by removing the anaerobic zone. For the sake of discussion of the best treatment options, it will be assumed that the anaerobic zone should be present in the final design for safety against toxic spikes. Thus, assuming the anaerobic reactor would be scaled up based on the proportion of the anaerobic and anoxic reactors tested (a 4 L anaerobic reactor was used with an 8.5 L anoxic reactor), an anaerobic basin of 1.13 million gallons would be incorporated into the full scale design.

Including the anaerobic basin, a total basin volume for A2/O treatment of the primary effluent of 12.61 million gallons should be sufficient (not including final clarifier design), with an overall HRT of 6.1 hours. A substantial savings would be realized, however, when one considers the fact that this is treatment of the primary effluent, and that existing structures involved in secondary treatment would be incorporated into the design. The existing secondary treatment process at HRWTF involves 5.76 million gallons of UNOX aeration basins, which would be used as part of the A2/O treatment of the primary effluent. Thus only 6.85 million gallons (12.61 million gallons - 5.76 million gallons) would be added to the existing process, with a corresponding additional HRT of 3.3 hours. This does not include the final clarifier volume, but the existing 9.58 million gallons of final clarifiers would be used as well.

With respect to solids handling, a plot was made of the amount of biomass grown and wasted at steady state COD loadings (see Figure 21). Although the data are scattered for both the daily steady state data and steady state averages for each operation period, such an approach was used to estimate biomass production to maintain consistency with the way in which the different systems were analyzed. A linear trend was assumed, and a least squares regression line was fit to the steady state average data. Assuming a linear trend is correct, it is estimated that 212 lbs of biomass (VSS) will be grown/wasted for every 10,000 lbs of COD entering the system. Assuming an average COD concentration of 590 mg/L (as was observed during this experiment), at a full scale 50 MGD, approximately 32,000 lbs of COD would enter the system per day, resulting in 680 lbs of biomass grown and wasted per day.

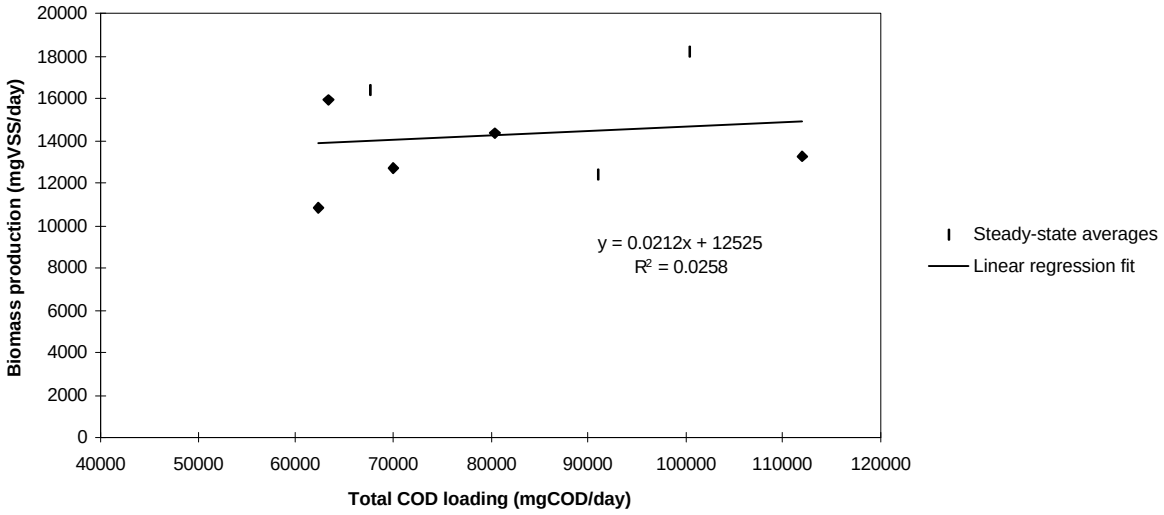

Figure 21. Primary effluent treatment train: Estimating biomass production as a function of the total COD loading (including both the influent and methanol addition).

Results and Discussion: Heated Primary Effluent Treatment Train

During initial testing, an attempt was made to determine if COD/BOD removal could be achieved for the pulp mill contributing the high temperature wastewater using a fully aerobic process. It was decided that the combination of the high temperature environment (approximately 40 to 45°C) and the wide variations in the daily COD of the influent made consistent treatment of the high temperature waste stream prior to its mixing with the other waste streams virtually impossible. It was observed, however, that nitrification appeared to be occurring, because nitrates were showing up in the effluent when the only form of nitrogen in the influent was Kjeldahl nitrogen. Because this was initially an investigation of the feasibility of high temperature COD removal, an attempt was made to kill off nitrification during the study by reducing the sludge age to 4 days. It proved difficult to maintain enough biomass in the system at such a low SRT, however, so the sludge age was raised again to eight (8) days. Because nitrification continued to occur, it was decided that the feasibility of nitrification of the primary effluent of HRWTF at its high summer temperatures should be tested. Thus, the same fully aerobic system was used with the primary effluent of HRWTF as its influent.

It was determined that nitrification of the primary effluent was, in fact, possible at temperatures in the range of 40 to 47°C. Maintaining a stable MLVSS proved to be quite difficult without often underwasting, however, and the ETSS tended to be higher and more sporadic than the sludge resulting from A2/O treatment of the primary effluent at moderate temperatures (aerobic temperatures generally within 24 to 30°C).

Similar to the other treatment trains tested, quasi-steady state operation periods were delineated by consideration of stable performance with respect to MLVSS, the SRT, hydraulic loading, COD and nitrogen loading, COD and nitrogen removal, and the treatment train configuration. As with the other treatment trains, the MLVSS tended to be the last parameter to reach a steady trend and strongly influenced the delineation of steady state operation. From all considerations, seven (7) steady state periods were delineated (see Figure 22) and are characterized in Table 16. The reasons for the separate delineations of each operation period are described below in Table 15.

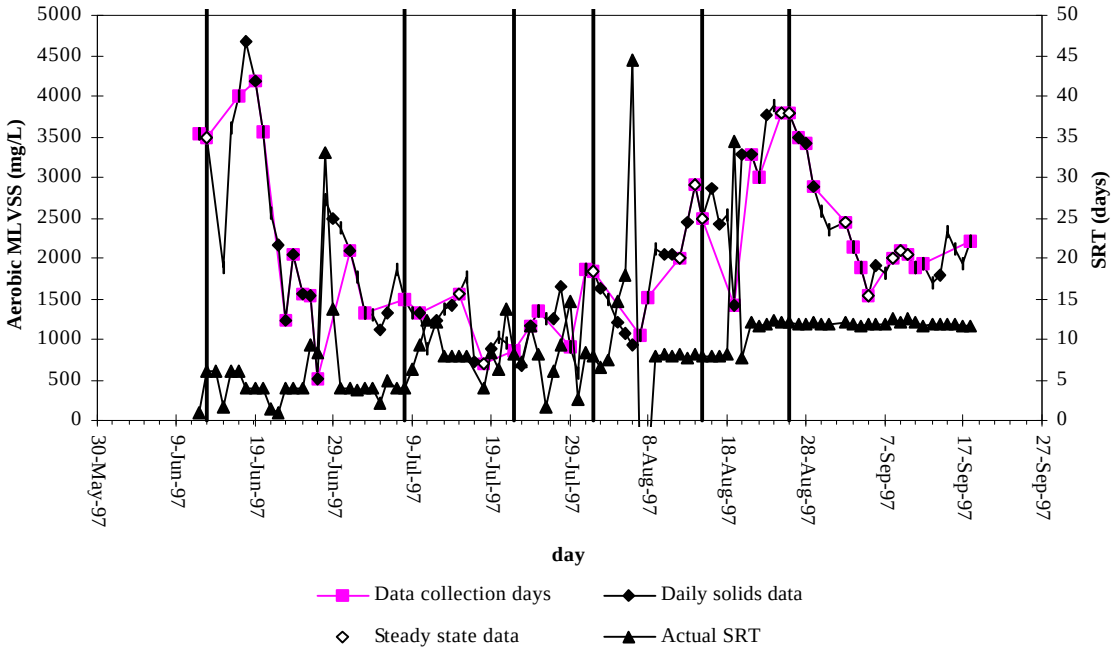

Figure 22. Heated primary effluent treatment train: Using the MLVSS and SRT to help delineate steady state operation periods. The bold lines indicate the last day of a steady state operation period.

Table 15. Characterization and delineation of separate operation periods for the heated primary effluent treatment train.

Operation period	Steady state dates	Reason for separate delineation
1	6/12-6/13	• Stabilized performance after startup on 5/19.
2	7/3-7/8	• Large COD spike on 6/17. • Switched from target SRT of 6 days to 4 days on 6/18. • Added 3 L of biomass and decreased flows on 6/27. • Performance and wastewater stabilized by 7/3.
3	7/15-7/22	• Changed to Target SRT of 8 days on 7/10. • Performance stabilized by 7/15.
4	7/25-8/1	• Doubled ammonia supplement starting 7/23. • Performance stabilized by 7/25..
5	8/12-8/15	• Switched from pulp mill wastewater to HRWTF primary effluent on 8/4. • Added 2.6 L and 2.1 L of nitrifying MLSS on 8/7 and 8/8, respectively because of low solids. • Performance and MLVSS stabilized by 8/12.
6	8/25-8/26	• Changed to target SRT of 12 days starting 8/21. • Performance stabilized by 8/25.
7	9/2-9/12	• Added 4 L anoxic reactor to front end of process at 15:30 on 8/26. • Performance stabilized by 9/2.

The pulp mill waste with ammonium chloride (NH_4Cl) addition (due to the low nitrogen content of the wastewater) was fed to the system from operation periods 1 through 4. It was attempted to maintain a 4 day SRT during operation periods 1 and 2, but maintaining a stable MLVSS was difficult, and many days of zero wastage were required to avoid losing the activated sludge (see Appendix 16). The hydraulic and nutrient loading was reduced from operation period 1 to operation period 2 in an attempt to achieve better performance and maintain a more stable MLVSS. From operation period 2 onward, the hydraulic loading was kept the same. Operation periods 3 and 4 were run at an 8 day SRT with the same wastewater and hydraulic loading as operation period 2, but the ammonia loading was doubled (by doubling the amount of ammonium chloride being added) from operation period 3 to operation period 4 to determine if nitrification was, in fact, occurring, as would be witnessed by an increase in effluent nitrates (assuming little denitrification was occurring). Having observed significant effluent nitrates, the feed was switched from the low-nitrogen, high COD/BOD pulp mill wastewater to the primary effluent of HRWTF on August 4. Thus, operation periods 5 through 7 contain the only data pertaining to the feasibility of high temperature nitrification of the primary effluent of HRWTF. All data will be used in discussion of the feasibility of high temperature nitrification in general, however. The SRT was switched from 8 days during operation period 5 to 12 days during operation period 6 in an attempt to favor nitrification more and see if the effluent ammonia concentration could be brought completely to zero which was, in fact, the case. The difference between operation period 6 and 7 was the addition of an anoxic reactor at the front of the treatment train during operation period 7 to determine if the nitrified wastewater could also be denitrified at the high temperatures.

Feasibility of high temperature nitrification, especially of the primary effluent:

There is evidence that nitrification was occurring during all seven operation periods. The bulk of the argument hinges upon the fact that, with the exception of operation period 7 when an anoxic reactor was added, the effluent nitrate concentrations were always higher than the influent nitrate concentrations during steady state operation and even transitional days (see Table 16 and Appendix 12). A nitrogen balance was performed on the overall system in the same fashion described previously, using the influent TKN concentration, effluent SKN concentration, influent flow rate, and 12% of the estimated VSS growth. The approach to a nitrogen balance was apparently flawed, however, as demonstrated by the fact that negative values were calculated for the percent of bioavailable TKN oxidized during operation periods 1 and 2 (see Table 16 and Appendix 12). Part of the problem was apparently estimating the amount of nitrogen required for growth, because the amount of nitrogen calculated to be required for growth was almost always much higher than the TKN loading to the system during these two operation periods (see Appendix 12). The sporadic and high daily ETSS concentrations were most likely the cause of the poor nitrogen balance (see Appendix 16 and Figure 23), as opposed to the possibility that negative growth

Table 16. Heated primary effluent treatment train: Overall performance and loading characteristics.

Steady-state operation period	**1** 6/12-6/13		**2** 7/3-7/8		**3** 7/15-7/22		**4** 7/25-8/1		**5** 8/12-8/15		**6** 8/25-8/26		**7** 9/2-9/12	
Configuration	Fully aerobic		Fully aerobic		Fully aerobic		Fully aerobic		Fully aerobic		Fully aerobic		MLE	
SRT	**3.4**	2	**4.2**	5	**8.0**	7	**7.3**	8	**8.0**	4	**12.2**	2	**12.0**	11
(days)	3.6		0.4		3.0		4.1		0.2		0.0		0.3	
Total HRT	**5.2**	2	**7.9**	5	**7.7**	7	**7.9**	8	**7.8**	4	**8.0**	2	**10.4**	11
(hours)	0.1		0.1		0.3		0.0		0.1		0.2		0.4	
Influent flow rate	**61.2**	2	**40.1**	5	**41.0**	7	**40.3**	8	**40.9**	4	**39.6**	2	**39.8**	11
(L/day)	1.0		0.4		1.4		0.0		0.7		1.0		1.4	
RAS % of influent flow rate*	**90**	2	**218**	5	**297**	7	**247**	8	**205**	4	**215**	2	**207**	11
(%)	1		72		16		61		4		n/a		8	
NR % of influent flow rate	n/a		n/a		n/a		n/a		n/a		n/a		**298**	11
(%)														
MLVSS	**3520**	2	**1410**	5	**1037**	7	**1488**	8	**2470**	4	**3785**	2	**1994**	11
(mg/L)	35		127		461		450		446		7		241	
ETSS	**526**	2	**107**	5	**37**	7	**91**	8	**19**	4	**10**	2	**9**	11
(mg/L)	730		70		20		81		9		1		8	
Specific growth rate	**0.643**	2	**0.242**	5	**0.136**	7	**0.207**	8	**0.129**	4	**0.082**	2	**0.083**	11
(1/day)	0.672		0.018		0.054		0.177		0.006		0.000		0.002	
TKN influent	**922**	2	**451**	2	**462**	3	**1104**	4	**1454**	2	**1237**	2	**1233**	7
(mg-N/day)	154		46		49		76		80		21		157	
SKN effluent	**171**	2	**43**	2	**48**	3	**88**	4	**269**	2	**151**	2	**49**	7
(mg-N/day)	118		1		30		93		39		63		11	
NHx influent	**638**	2	**395**	2	**412**	3	**1104**	4	**1300**	3	**1152**	2	**1098**	7
(mg-N/day)	82		69		74		76		67		57		186	
NHx effluent	**0**	2	**0**	2	**16**	3	**62**	4	**183**	3	**0**	2	**0**	7
(mg-N/day)	0		0		28		72		22		0		0	
NOx influent	**0**	2	**0**	2	**0**	3	**0**	4	**123**	2	**162**	2	**217**	8
(mg-N/day)	0		0		0		1		34		91		150	
NOx effluent	**540**	3	**178**	3	**189**	3	**795**	4	**503**	2	**905**	2	**193**	8
(mg-N/day)	57		98		94		73		179		1		68	
COD influent	**20788**	2	**18184**	2	**13671**	3	**11007**	3	**27066**	3	**25090**	2	**24177**	9
(mg/day)	2077		5531		4861		3146		3651		643		3744	
COD effluent	**3555**	2	**5080**	2	**2497**	3	**1774**	3	**1826**	3	**2161**	2	**2171**	9
(mg/day)	665		2338		530		140		220		308		385	
TKN influent	**15.1**	2	**11.2**	2	**11.2**	3	**27.4**	4	**36.1**	2	**31.3**	2	**30.8**	7
(mg-N/L)	2.3		1.2		1.5		1.9		2.0		1.3		4.0	
SKN effluent	**2.8**	2	**1.1**	2	**1.1**	3	**2.2**	4	**6.7**	2	**3.8**	2	**1.2**	7
(mg-N/L)	1.9		0.0		0.7		2.3		1.0		1.5		0.3	
NHx influent	**10.4**	2	**9.8**	2	**10.0**	3	**27.4**	4	**32.1**	3	**29.1**	2	**27.5**	7
(mg-N/L)	1.5		1.7		2.0		1.9		1.9		2.2		4.8	
NHx effluent	**0.0**	2	**0.0**	2	**0.4**	3	**1.5**	4	**4.5**	3	**0.0**	2	**0.0**	7
(mg-N/L)	0.0		0.0		0.7		1.8		0.6		0.0		0.0	
NOx influent	**0.0**	2	**0.0**	2	**0.0**	3	**0.0**	4	**3.1**	2	**4.1**	2	**5.5**	8
(mg-N/L)	0.0		0.0		0.0		0.0		0.8		2.4		3.8	
NOx effluent	**8.8**	3	**4.4**	3	**4.6**	3	**19.7**	4	**12.5**	2	**22.9**	2	**4.8**	8
(mg-N/L)	0.8		2.4		2.4		1.8		4.4		0.6		1.7	
COD influent	**340**	2	**451**	2	**329**	3	**273**	3	**667**	3	**634**	2	**610**	9
(mg/L)	40		137		107		78		91		33		88	
COD effluent	**58**	2	**126**	2	**60**	3	**44**	3	**45**	3	**55**	2	**55**	9
(mg/L)	10		58		12		3		5		6		9	
Estimated amount of TKN oxidized by the overall system	**-2850**	2	**-156**	2	**166**	3	**745**	4	**695**	2	**592**	2	**831**	7
(mg-N/day)	3820		2		148		220		143		81		162	
Estimated % bioavailable TKN oxidized using a nitrogen balance	**-392**	2	**-39**	2	**36**	3	**69**	4	**50**	2	**54**	2	**70**	7
	527		4		27		18		9		3		5	
% bioavailable TKN showing up as NOx in the final effluent	**72**	2	**45**	2	**43**	3	**74**	4	**27**	2	**69**	2	**-4**	7
	4		29		18		10		10		14		16	
Overall observed specific nitrification rate	**-0.061**	2	**-0.008**	2	**0.014**	3	**0.044**	4	**0.023**	2	**0.012**	2	**0.031**	7
(mg-N/mgVSS/day)	0.082		0.001		0.014		0.028		0.010		0.002		0.008	

Note: Top value indicates mean value during operation period. The lower value indicates the standard deviation. The value to the right indicates the number of samples in the data set.

* RAS was not kept very constant, but this was to keep biosolids in system, NOT to help denitrification. Therefore, separate operation periods were not delineated for changes in the RAS flow rate.

might have occurred (meaning cell lysis occurred faster than cell growth). Lower and less variable ETSS concentrations were observed in the other operation periods (see Figure 23), and the nitrogen balance for those operational periods did indicate that TKN was being oxidized (see Table 16). Estimates of the amount of TKN oxidized for operation periods 3 through 7 indicated that the system, on average, oxidized 36 to 70 percent of the bioavailable TKN entering the system.

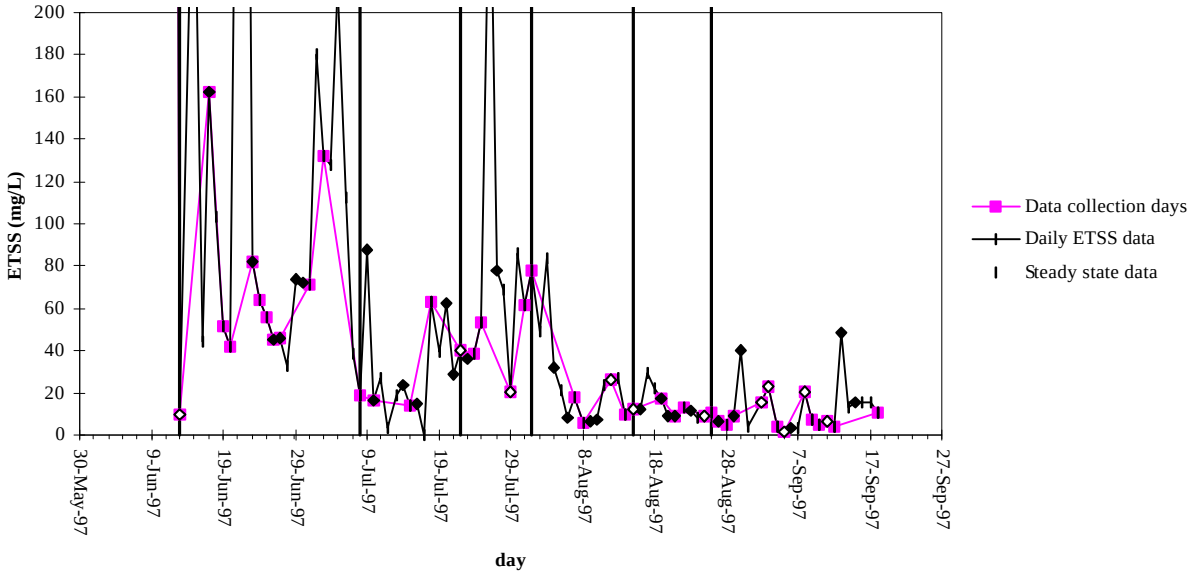

Figure 23. Heated primary effluent treatment train: Daily effluent suspended solids quality The bold lines indicate the last day of a steady state operation period.

An alternative method was used to attempt estimating the amount of TKN oxidized. An estimate was made of the apparent NO_x production (i.e., the mg-N/day of nitrates measured exiting in the final effluent less the nitrates in the influent) and compared to the mg-N/day of bioavailable TKN entering the system (i.e., the difference between the influent TKN and effluent soluble organic nitrogen). Because the system was fully aerobic for operation periods 1 through 6, it was assumed that essentially zero denitrification was occurring in the system during those times. Such an approach is obviously not valid for operation period 7, when denitrification was expected. With such an assumption, the ratio of the observed mg of nitrate-N per day apparently produced by the system divided by the mg per day of bioavailable TKN-N entering the system should represent the percent of TKN which was oxidized. This assumption was obviously incorrect, as nitrates exiting the clarifier were frequently lower than nitrates entering the clarifier. Surprisingly, nitrates exiting the clarifier were occasionally actually higher than the nitrates entering the clarifier (see Appendix 15). Because there was not as much data collected from the aerobic reactor directly, and because there was no trend with respect to how much nitrate consumption or production occurred in the clarifier, the assumption of zero activity in the clarifier is maintained for the sake of discussion, and it is understood that such estimates of percent nitrification would tend to be biased low. The anomaly is, however, this estimate tended to provide a higher estimate of the percent TKN oxidation than the nitrogen balance (see Table 16). From both analyses, ignoring negative values which are obviously incorrect, it is estimated that the high temperature system was able to nitrify, on average, 27 to 74% of the bioavailable TKN (see Table 16). Given the peculiar results from both ways of estimating the percent of TKN oxidation, all that can truly be concluded from the study is that TKN oxidation was certainly occurring. Regardless of how much TKN was oxidized (as opposed to assimilated), average steady state effluent ammonia concentrations were in the range of zero to 4.5 mg-N/L at the loadings tested (see Table 16), indicating that complete ammonia removal is possible at high temperatures, and that much of the removal can be attributed to TKN oxidation (i.e., nitrification).

Complete nitrification, as indicated by zero ammonia being detected in the final effluent in conjunction with effluent nitrates being detected, was actually achieved at least on occasion during all operation periods except operation period 5 (see Appendix 12). The feed was switched from the pulp mill wastewater to the primary effluent on August 4, and the TKN loading increased slightly, so it is possible that incomplete nitrification during operation period 5 was due to acclimation to the different wastewater. However, the last steady state day of operation period 5 corresponded to the thirteenth day of operation at the new conditions (approximately 1.5 SRTs), and acclimation should have been relatively complete; thus, it was assumed that the average 4.5 mg-N/L of ammonia in the final effluent was as complete as the nitrification would get at that loading and SRT (see Table 16).

Increasing the SRT to 12 days on August 21 resulted in complete nitrification by August 25 (during operation period 6), but this also coincided with a decrease in the TKN loading from an average of 1454 mg-N/day during operation period 5 to 1237 mg-N/day during operation period 6, while the COD loadings remained similar. Thus, it is not clear whether increasing the sludge age benefited nitrification and would necessarily result in complete nitrification on a regular basis for the primary effluent at extant high temperatures, but it is commonly accepted that higher sludge ages tend to increase the consistency and extent of nitrification. Both techniques for estimating the relative percent of nitrification suggested the percent of bioavailable TKN which was oxidized increased from operation period 5 to operation period 6, lending further evidence to the claim that the increase in SRT did, in fact, enhance nitrification performance (see Table 16).

As with the high temperature study for the secondary effluent treatment train, there were cooler zones in which nitrification might have been occurring, or at least which might have been allowing for growth to catch up with possible cell lysis in the stressful high temperature environment. Additionally, the DO in the final clarifier was not necessarily zero. From the data collected from July 27 to August 24, the final clarifier had an average temperature of 38.3^{o}C and an average DO of 0.6 mg/L (see Appendix 15). After the anoxic reactor was added, the conditions became even more favorable for nitrification in the final clarifier, with an average temperature of 35.5^{o}C and an average DO of 0.9 mg/L measured from August 27 to September 7. Because it was observed that the final clarifier was substantially cooler than the aerobic reactor (as was the anoxic reactor once it was added during operation period 7) and that the DO in the clarifier was not necessarily zero, especially considering the fact that microenvironments of trapped oxygen within flocs are possible, an attempt was made to determine if nitrification was definitely occurring in the aerobic reactor and to what extent.

As with the high temperature study of the secondary effluent, a good nitrogen balance could not be performed on the aerobic reactor itself. For the sake of discussion, a nitrogen balance was performed on the aerobic reactor with the assumption that all TKN assimilation (i.e., growth) occurred in the aerobic reactor of the heated primary effluent treatment train. Thus, discussion of oxidation occurring in the aerobic reactor pertains to the minimum possible amount of TKN oxidized within the aerobic reactor. Even with the assumption that all growth occurred within the aerobic reactor, the two forms of TKN oxidation estimation indicated that TKN was being oxidized in the aerobic reactor. Similar problems to the nitrogen balance of the overall system arose with the nitrogen balance on the aerobic reactor, with negative percent nitrification estimates arising. The estimate of the percent of bioavailable TKN showing up as nitrates exiting the aerobic reactor posed an additional problem, however, which was that more nitrate-nitrogen was apparently formed in the aerobic reactor than the amount of bioavailable TKN-nitrogen entering in one day during operation period 7, which resulted in a calculated average of over 100% of TKN being converted to nitrates (see Table 17). This posed an additional problem because this technique does not even take growth into account. Thus, if 100% of the bioavailable TKN was converted to nitrate-nitrogen, zero net growth, by definition, occurred that day. Regardless of the problems associated with both forms of nitrification estimates, if one takes into consideration the two means of estimating the percent of nitrification and the simple fact that nitrates exiting the aerobic reactor were always greater than the nitrates entering it (see Table 17 and Appendix 14), it is certain that a substantial amount of nitrification was occurring in the aerobic reactor, whether or not nitrification was occurring in the cooler clarifier.

As with the secondary effluent high temperature study, it is unclear whether or not the cooler zones were the primary reason a nitrifying population was able to survive. Given that the average clarifier temperature prior to the addition of the anoxic reactor was approximately 38^{o}C, that nitrification showed signs of inhibition, and the MLVSS began to drop drastically in this temperature range for the secondary effluent high temperature study, it appears that continued nitrification of the primary effluent at high temperatures was due to the sludge actually being acclimated to the high temperatures. It is important to note that this is still not the same as keeping the sludge exposed to temperatures of approximately 45^{o}C twenty-four hours per day, however, as would be expected in the existing process of HRWTF. Additionally, it should be noted that the sludge was very gradually acclimated to the higher temperatures by increasing the temperature from approximately 30^{o}C at startup to approximately 45^{o}C over the course of approximately two months (see Appendix 14). This is obviously not nearly as stressful as the increase

to approximately 45°C over the course of a few days, as would actually be observed in the existing process. When such a stress was applied to a nitrifying sludge, as was the case with the secondary effluent high temperature study, the system died off within a few days. Such a pattern in a full scale plant would be unacceptable, and the gradual acclimation to temperatures each year would obviously be impossible without the addition of a cooling process. Furthermore, the MLVSS concentrations changed sporadically, as did the ETSS (see Table 16 and Figures 22 and 23), and many days of zero wastage were necessary to avoid losing the system (see Appendix 16). Thus, nitrification of the HRWTF wastewater at high temperatures is not a practical solution, although it has been shown that nitrification can be sustained at temperatures near 45°C if an appropriate SRT is used and if acclimation to the temperatures is made gradually. Keeping a cooler zone may or may not be required to sustain nitrifying biomass; no experiments were conducted to answer this question.

Table 17. Heated primary effluent treatment train: Aerobic reactor performance and loading.

Steady-state operation period	**1** 6/12-6/13	**2** 7/3-7/8	**3** 7/15-7/22	**4** 7/25-8/1	**5** 8/12-8/15	**6** 8/25-8/26	**7** 9/2-9/12
Bioavailable TKN loading to the aerobic reactor (mg-N/day)	726	no data	541	895	1764	no data	394
SKN exiting aerobic reactor (mg-N/day)	171	no data	337	158	643	no data	243
NHx loading to the aerobic reactor (mg-N/day)	696	no data	529	932	1694	no data	368
NHx exiting aerobic reactor (mg-N/day)	0	no data	200	0	353	no data	0
NOx loading to the aerobic reactor (mg-N/day)	452	no data	257	1392	880	no data	823
NOx exiting aerobic reactor (mg-N/day)	1065	no data	304	1864	1388	no data	1210
NOx out - NOx in (mg-N/day)	613	no data	47	472	508	no data	386
Estimated minimum amount of TKN oxidized in the aerobic reactor (mg-N/day)	-5556	no data	50	633	691	no data	3
Estimated % bioavailable TKN oxidized using a nitrogen balance on aerobic reactor	-765	no data	9	71	39	no data	-105
% bioavailable TKN showing up as NOx exiting aerobic reactor	84	no data	9	53	29	no data	121

Note: There is only one or zero data points per operation period for balances on the aerobic reactor, with the exception of operation period 7 which had 2 data points. Thus, standard deviations are not applicable, and numbers of data points are not listed in the table.

For the sake of comparing performances of the different sludges and systems, the overall observed specific nitrification rate has been plotted against the F:M ratio in Figure 24. What is perhaps most interesting about the figure is the fact that two different wastewaters and three different sludge ages (4, 8, and 12 days) all appear to fit on the same curve. Because the rates are overall observed rates (and not measured rates), and because complete nitrification occurred on several days, it is not clear whether nitrification rates were more strongly affected by other factors (such as temperature) than sludge age or not. What can be said of the figure is that, as with the other systems, nitrification of the heated primary effluent treatment train did not appear to have reached a maximum rate, as indicated by the rate not appearing to reach a plateau.

With regard to denitrification, conditions and loadings were kept basically the same between operation periods 6 and 7, with the only difference being the addition of a 4 L anoxic reactor to the front of the treatment train. Adding the anoxic reactor did result in a decrease in the final effluent nitrates from an average concentration of 22.9 mg-N/L during operation period 6 to 4.8 mg-N/L during operation period 7. Thus, denitrification could definitely be carried out to some extent in the high temperature system, although it was not quite complete under the conditions used. Despite the fact that data was collected for fifteen days (allowing slightly over 1 SRT for acclimation), 2 mg-N/L of nitrates were detected escaping the anoxic reactor on September 11 (see Appendix 13). The COD exiting the anoxic reactor during operation period 7 was higher than the COD of the final effluent on both days that data was

collected (see Appendix 12 and Appendix 13), thus denitrification was not COD limited. Not enough time was available to determine if complete denitrification could be achieved. Thus, it is not known if denitrification simply required more time for acclimation, or if it was somehow rate limited at the high temperatures. At the end of operation period 7, the project was terminated in order to proceed with selection for pilot-scale testing, and it was thus not determined to what extent the system could have been pushed or how it could have been optimized.

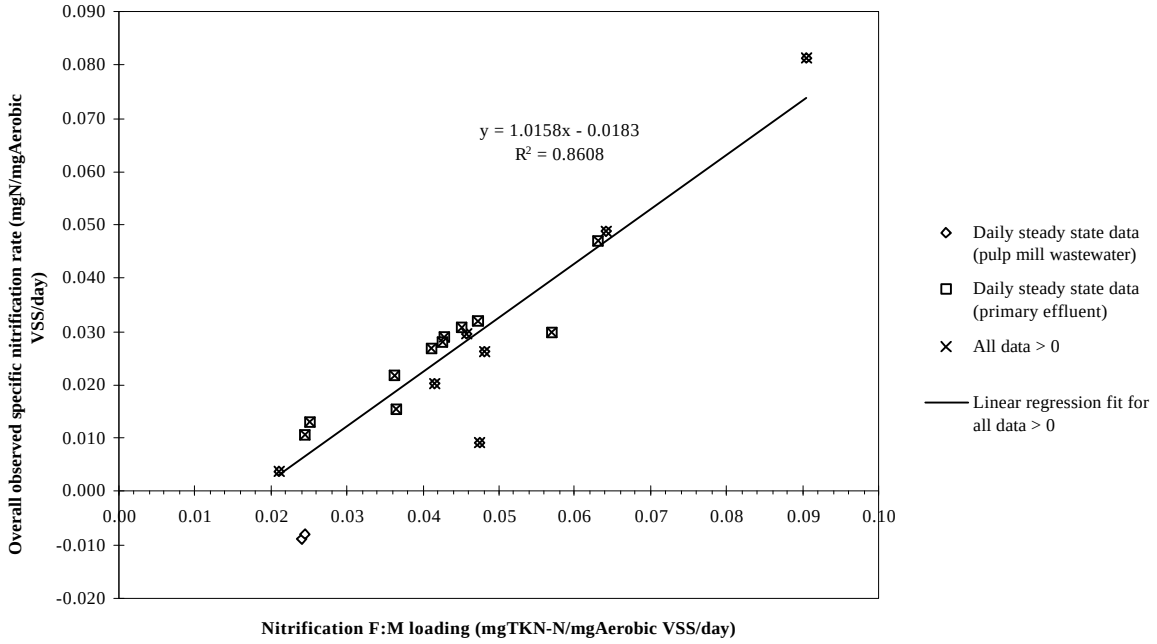

Figure 24. Heated primary effluent treatment train: Overall observed specific nitrification rate as a function of the F:M loading.

Because it was decided that high temperature nitrification of the primary effluent of HRWTF does not appear to be a practical solution due to the difficulty of maintaining nitrification after a fairly sudden increase in temperature, design of a full scale treatment process at high temperatures is not discussed in this thesis.

Results and Discussion: High Ammonia Side Stream Treatment Train

At the time of testing, it was believed that one of the industries contributing a substantial amount of flow to HRWTF contained high concentrations of methylethyl ketoxime (MEKO), a compound known to be inhibitory to nitrification (Smith, 1994; Lubkowitz, 1996). It was discovered after the termination of the investigation that the said industry had changed its processes such that MEKO was no longer present in its final effluent. The study for the side stream has been included in this thesis however, because of its value in demonstrating the feasibility of nitrification and denitrification of a high ammonia (approximately 100 to 150 mg-N/L) wastewater with relatively low COD concentrations (generally in the range of 300 to 700 mg/L during this study), as well as the likelihood of treating the side stream separately to achieved the nitrogen removal goals of HRWTF.

Two strategies were used for the treatment of the side stream. The first strategy used was an MUCT approach, i.e., an anaerobic basin followed by two anoxic zones, and finally an aerobic zone. This first treatment strategy was designed for the removal/detoxification of MEKO (Lubkowitz, 1996), but as stated above, was not necessary because no MEKO was present. Removing one of the anoxic reactors and creating an A2/O process was then tested for treatment of the same waste stream. Similar effluent quality was achieved, which was not expected until it had been discovered that no MEKO was present. Thus, although no detoxification was necessary for the side stream, it was demonstrated that a high ammonia waste stream could be completely nitrified, but, as both systems tested were post-nitrification for a very high ammonia waste stream, denitrification could not be completed to zero levels of nitrates. The most successful periods of operation did remove nitrates down to the range of 6.2 to 15.0 mg-N/L however (to be discussed below).

The system was initially run at an 18-day SRT in the MUCT configuration. After forty (40) days of operation, with nitrification still incomplete and with difficulty keeping the MLVSS above 2000 mg/L, the system was run with a target SRT of 24 days. After three weeks of operation (from July 1 to July 21), including several days of zero wastage or mixed liquor addition, the MLVSS appeared to stabilize, as did performance. The MUCT system was operated at the 24 day SRT for four weeks (until August 22), at which point the system was reconfigured by removing one of the 4 liter anoxic reactors and operating the system as an A2/O process with the same 24 day SRT. The A2/O process was run for the remainder of the experiment which terminated September 12.

As with the other treatment trains, true steady states were never reached, but quasi-steady states were delineated based upon relatively constant trends observed with respect to MLVSS, the SRT, hydraulic loading, recirculation rates, COD and nitrogen loading, and COD and nitrogen removal performance. Just as with the other treatment trains, loading was varied primarily by varying the hydraulic loading. In terms of determining when performance had reached steady state, the MLVSS tended to take longer to adjust than the effluent quality. Thus, delineating when acclimation to new operating conditions had occurred was dominated by the MLVSS, as illustrated in Figure 25. From all considerations, six (6) steady state operation periods were defined, and the reasons for the separate delineations of each operation period are described below in Table 18. The characteristics of each operation period are summarized in Table 19.

It was determined that the high-ammonia side stream could be successfully nitrified completely with an appropriate SRT for both the MUCT process and the A2/O process at low enough loadings. The final effluent ammonia concentration could not be kept as consistently near zero, however, as the other lower concentration waste streams, i.e. the primary and secondary effluents of HRWTF. This could have been due to any of a variety of factors, with likely influences being variations in influent ammonia concentrations as great as 40 mg-N/L from day to day or lack of adequate time for acclimation of the 24 day SRT sludge to frequent changes (generally within one to two weeks) in loading or operating conditions. Final effluent nitrate concentrations did not reach zero levels even when complete denitrification was observed in the anoxic zone and a 300% nitrate recirculation rate was used. Average effluent NO_x concentrations were 9 to 10 mg-N/L under such conditions (to be discussed below). Because the loadings were increased in time, but the system was switched over to an A2/O configuration for the last three weeks of operation, only the A2/O process was pushed to the point of breakthrough. After initial acclimation and optimization, nitrates and COD did not show significant breakthrough until the final (highest) loading rate was tested. Slight ammonia breakthrough appeared to occur at lower loadings, however.

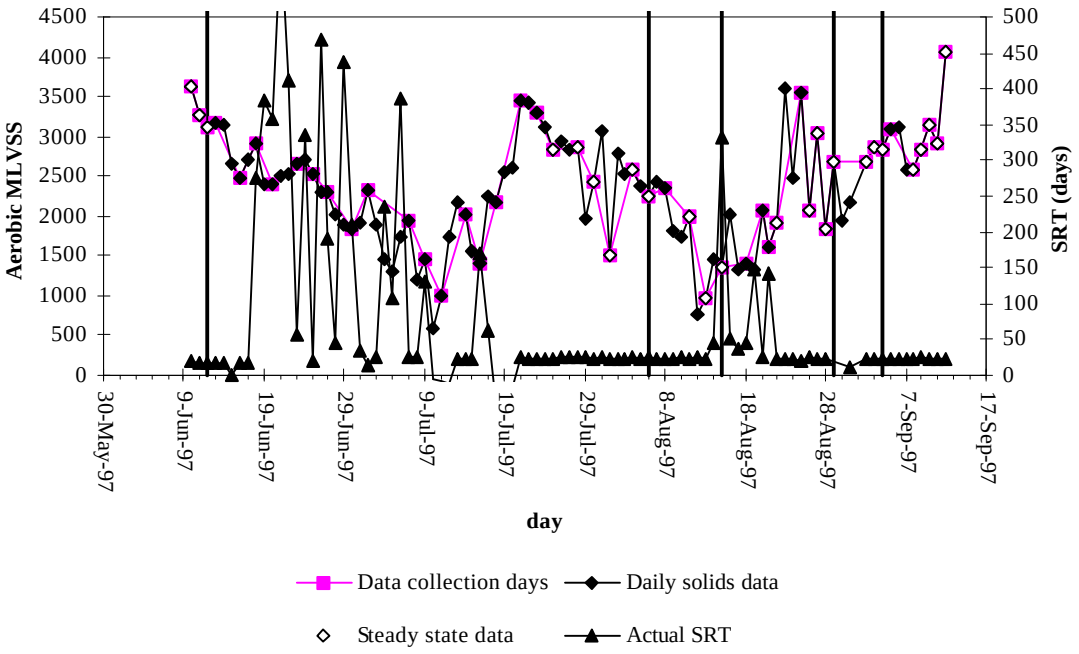

Figure 25. High ammonia side stream treatment train: Using the MLVSS and SRT to help delineate steady state operation periods. The bold lines indicate the last day of a steady state operation period.

Table 18. Characterization and delineation of separate operation periods for the high ammonia side stream treatment train.

Operation period	Steady state dates	Reason for separate delineation
1	6/10-6/12	• Reason for start: Stabilized performance after startup with 18 day SRT. • Reason for end: Decreased hydraulic loading on 6/13.
2	7/25-8/6	• Conditions delaying acclimation: • Changed various operating conditions from 6/17 to 7/9, including zero wastage to improve nitrification and increase MLSS, and varying recirculation rates. • Added 5 L of nitrifying MLSS on 7/10 and 5 L of MLSS on 7/11. • Changed to and maintained a 24 day SRT starting 7/13. • Began methanol addition on 7/17. • Added 1 L, 1.6 L, and 2 L of nitrifying biomass on 7/18, 7/19, and 7/20, respectively. • Reason for start: Performance stabilized by 7/25. • Reason for end: Doubled methanol dosage 8/8.
3	8/11-8/15	• Reason for start: Performance stabilized by 8/11. • Reason for end: Reconfigured to A2/O by removing the first anoxic reactor at 16:15 on 8/21.
4	8/22-8/29	• Reason for start: Performance and MLVSS on 8/22 were within range of all other steady state days. • Reason for end: Increased flow rates on morning of 8/30.
5	9/2-9/4	• Reason for start: Performance stabilized by 9/2. • Reason for end: Increased hydraulic loading morning of 9/5. RAS was increased proportionally, but nitrate recirculation (NR) was decreased from 300% to 280% due to slight calculation error.
6	9/8-9/12	• Reason for start: Performance stabilized by 9/8. • Reason for end: Study terminated after 9/12.

Table 19. High ammonia sided stream treatment train: Overall performance and loading characteristics.

Steady-state operation period	**1** 6/10-6/12		**2** 7/25-8/6		**3** 8/11-8/15		**4** 8/22-8/29		**5** 9/2-9/4		**6** 9/8-9/12	
Configuration	MUCT		MUCT		MUCT		A2/O		A2/O		A2/O	
SRT	**18.3**	3	**24.2**	13	**24.1**	3	**23.5**	7	**23.6**	3	**24.2**	4
(days)	0.3		0.5		0.8		1.2		0.2		0.6	
Total HRT	**20.5**	3	**26.8**	13	**26.2**	3	**18.6**	8	**16.1**	3	**11.7**	4
(hours)	0.8		0.5		0.7		0.3		0.6		0.4	
Influent flow rate	**37.4**	3	**28.6**	13	**29.3**	3	**36.1**	8	**41.8**	3	**57.4**	4
(L/day)	1.4		0.5		0.8		0.6		1.4		1.9	
RAS % of influent flow rate	**89**	3	**103**	11	**100**	1	**104**	5	**104**	2	**102**	3
(%)	3		7		n/a		4		5		0	
Anaerobic recirculation (AR)	**185**	3	**240**	11	**229**	1	**n/a**		**n/a**		**n/a**	
% of influent flow rate	7		8		n/a							
Nitrate recirculation (NR)	**187**	3	**297**	11	**286**	1	**301**	5	**302**	2	**280**	3
% of influent flow rate	9		9		n/a		8		17		6	
Aerobic MLVSS	**3337**	3	**2540**	13	**1245**	3	**2653**	8	**2797**	3	**3243**	4
(mg/L)	254		437		660		698		101		560	
ETSS	**14**	3	**22**	13	**26**	3	**22**	8	**16**	3	**13**	4
(mg/L)	8		20		19		22		16		7	
Specific growth rate	**0.055**	3	**0.040**	5	**0.045**	3	**0.042**	6	**0.041**	3	**0.036**	5
(1/day)	0.001		0.001		0.025		0.003		0.001		0.001	
TKN influent	**5454**	3	**4281**	5	**3397**	2	**3779**	5	**4412**	3	**8701**	5
(mg-N/day)	218		465		94		328		105		2844	
SKN effluent	**1792**	3	**123**	5	**132**	2	**101**	5	**142**	3	**655**	5
(mg-N/day)	271		54		44		53		35		694	
NHx influent	**5004**	3	**4281**	5	**3397**	2	**3581**	5	**4265**	3	**8617**	5
(mg-N/day)	95		465		94		255		32		2944	
NHx effluent	**1774**	3	**58**	5	**0**	2	**29**	5	**74**	3	**570**	5
(mg-N/day)	271		67		0		46		33		663	
NOx influent	**0**	3	**2**	5	**0**	2	**2**	5	**0**	3	**0**	5
(mg-N/day)	0		4		0		2		0		0	
NOx effluent	**1020**	3	**1110**	5	**657**	2	**364**	5	**375**	3	**952**	5
(mg-N/day)	93		214		123		145		84		377	
COD influent	**16832**	3	**10846**	5	**7315**	3	**14748**	6	**12712**	3	**36301**	5
(mg/day)	3162		1811		1131		1665		1279		6461	
Methanol addition?	no		yes		yes		yes		yes		yes	
Total COD in	**16832**	3	**17684**	5	**19794**	3	**32013**	6	**31858**	3	**62626**	5
(mg/day)	3162		1811		2260		1692		1718		6461	
COD effluent	**1464**	3	**1020**	5	**936**	3	**1251**	6	**1386**	3	**2734**	5
(mg/day)	218		195		275		229		413		262	
TKN influent	**145.7**	3	**148.6**	5	**118.0**	2	**104.1**	5	**105.8**	3	**151.8**	5
(mg-N/L)	1.9		16.1		3.3		8.9		5.4		47.5	
SKN effluent	**47.8**	3	**4.3**	5	**4.6**	2	**2.8**	5	**3.4**	3	**11.3**	5
(mg-N/L)	6.6		1.9		1.5		1.5		0.9		11.9	
NHx influent	**133.7**	3	**148.6**	5	**118.0**	2	**98.7**	5	**102.2**	3	**150.3**	5
(mg-N/L)	2.8		16.1		3.3		6.1		2.8		49.3	
NHx effluent	**47.3**	3	**2.0**	5	**0.0**	2	**0.8**	5	**1.8**	3	**9.9**	5
(mg-N/L)	6.3		2.3		0.0		1.3		0.8		11.3	
NOx influent	**0.0**	3	**0.1**	5	**0.0**	2	**0.0**	5	**0.0**	3	**0.0**	5
(mg-N/L)	0.0		0.2		0.0		0.1		0.0		0.0	
NOx effluent	**27.2**	3	**38.5**	5	**22.8**	2	**10.0**	5	**9.0**	3	**16.6**	5
(mg-N/L)	1.4		7.4		4.3		3.8		2.2		6.4	
COD influent	**448**	3	**377**	5	**254**	3	**407**	6	**304**	3	**636**	5
(mg/L)	70		63		40		39		21		115	
COD effluent	**39**	3	**35**	5	**33**	3	**35**	6	**33**	3	**48**	5
(mg/L)	4		7		10		6		9		5	
% of bioavailable TKN oxidized	**55**	3	**90**	5	**92**	2	**91**	4	**89**	3	**89**	5
	5		1		3		2		1		5	
Overall observed specific	**0.050**	3	**0.112**	3	**0.130**	2	**0.102**	2	**0.077**	3	**0.139**	4
*nitrification rate	0.006		0.031		0.066		0.011		0.002		0.035	
(mg-N/mgVSS/day)												

Note: The top value indicates the mean value observed during that operation period. The lower value indicates the standard deviation. The value to the right indicates the number of samples in the data set.

* Overall observed specific nitrification rate data correspond to non-ammonia-limited days, except for operation period 3, which was ammonia limited both days.

Feasibility of nitrification of the High-Ammonia Side Stream

Despite allowing forty days to acclimate, complete nitrification could apparently not be achieved with an 18 day SRT at the loading rate tested during operation period 1, with an average effluent ammonia concentration of 47.3 mg-N/L (see Table 19 for these values and all other values mentioned in this discussion). The loading was decreased after operation period 1, and the SRT was increased to 24 days in order to favor nitrification as much as possible during operation period 2. Changing the SRT obviously enhanced nitrification because the overall observed specific nitrification rate corresponding to non-ammonia-limited days increased from an average of 0.050 mg-N/mgVSS/day during operation period 1 to an average of 0.112 mg-N/mgVSS/day during operation period 2 despite decreased TKN loading. The percent of TKN oxidized also increased from an average of 55% during operation period 1 to an average of 90% during operation period 2. The percent of bioavailable TKN oxidized was definitely associated with the change in the SRT, and it actually proved to be independent of the TKN loading for the range of loadings tested. The average percents of TKN oxidized during operation periods 2 through 6, which were all associated with a 24 day SRT, were between 89 and 92% for average TKN loadings ranging from 3397 to 8701 mg-N/day. Thus, the increase in the SRT did greatly enhance nitrification. The combination of increasing the SRT from 18 days to 24 days and decreasing the TKN loading from an average of 5454 mg-N/L to 4281 mg-N/L resulted in an improvement in effluent quality from an average soluble effluent ammonia concentration of 47.3 mg-N/L to an average of 2.0 mg-N/L. Despite the same hydraulic loading during operation period 2 and 3, the TKN loading fell from an average of 4281 to 3397 mg-N/day from operation period 2 to 3, due to the influent TKN concentration falling from an average of 148.6 to 118.0 mg-N/L. With such a decrease in loading, complete nitrification (zero effluent ammonia) was observed on both steady state days during operation period 3.

Removing one of the anoxic reactors and using an A2/O process (as was done just prior to operation period 4) resulted in continued success for nitrification of the side stream. Complete nitrification (zero mg-N/L of ammonia detected in the final effluent) was achieved with the A2/O process during operation period 4, with occasional ammonia breakthroughs of only 1.1 and 2.9 mg-N/L (see Appendix 18), resulting in an average soluble effluent ammonia concentration of 0.8 mg-N/L (see Table 19). The TKN loading only increased slightly from operation period 3 to operation period 4, with average TKN loadings of 3397 and 3779 mg-N/day, respectively, and only two data points exist for operation period 3. Thus, it is not clear whether the slight increase in the TKN and hydraulic loading actually resulted in slightly less stable nitrification, or if not enough data was collected during operation period 3 to observe an occasional ammonia spike in the final effluent. The system was probably being pushed toward true ammonia breakthrough, however, because increasing the TKN loading from a steady state average of 3779 (operation period 4) to 4412 (operation period 5) mg-N/day resulted in an increase in the effluent ammonia concentration, from an average of 0.8 mg-N/L to 1.8 mg-N/L (see Table 19), with no days of zero effluent ammonia occurring during operation period 5. The slight ammonia breakthrough that was observed during operation periods 4 and 5 was probably due to cell lysis products of the high-SRT biomass in the final clarifier being flushed out before it was oxidized, or possibly the deamination of complex nitrogenous compounds, rather than incomplete nitrification, because complete nitrification was observed for all three operation periods (operation periods 3 through 5) as determined by using the aerobic reactor as a control volume (see the discussion below).

Increasing the TKN loading to a steady state average of 8701 mg-N/day during operation period 6 resulted in an increase in the average effluent ammonia concentration to 9.9 mg-N/L, indicating that ammonia breakthrough had definitely occurred (see Table 19). Although the average data for operation period 6 suggests the system was stressed in terms of nitrification, earlier data during that operation period corresponded to lower loadings and better ammonia removal. For instance, zero ammonia was detected in the final effluent on September 8, when the TKN loading was only 6085 mg-N/day. The following day, the loading decreased slightly to 5643 mg-N/day, and an effluent ammonia concentration of 1.7 mg-N/L was detected (see Appendix 18). Although this is a slight breakthrough of ammonia, it was determined that low ammonia concentrations were detected on at least one day when complete nitrification had occurred within the aerobic reactor (to be discussed in the following paragraph). Much more ammonia escaped the system on the other days during operation period 6 which corresponded to higher TKN loadings in the range of 8885 to 12177 mg-N/day (see Appendix 18). Thus, it appears that the system could still fully nitrify TKN loadings up to approximately 6000 mg-N/day, but could not fully nitrify loadings beyond this. It is important to note, however, that the last day data was collected during operation period 6 corresponded to only seven days after the loading was increased. Given that the sludge had a 24 day SRT, and that the MLVSS was still increasing in response to the increased loading (see Figure 25), it is quite possible that complete nitrification, or at least better nitrification, would have occurred had sufficient time for acclimation been allowed.

As with the other systems tested, a nitrogen balance was performed on the overall system as well as just the aerobic reactor. Little data was collected from the individual reactors, and only one data point exists for operation periods 1, 3, 4, and 5. Thus, there is little that can be said with any confidence. From the data that was collected,

however, complete nitrification appeared to occur in the aerobic reactor during operation periods 3, 4, and 5, as indicated by the zero detectable ammonia exiting the reactor (see Table 20). The final effluent ammonia concentrations corresponding to the days on which data were collected from the aerobic reactor during those operation periods were 0, 0, and 1.1 mg-N/L, respectively (see Appendices 18 and 22). It is not known why there would be apparent ammonia release on only one of the three days, but the discrepancy is an indication that trace amounts of ammonia in the final effluent of this system might still correspond to complete nitrification. As mentioned, the non-zero effluent ammonia detected on a day when no ammonia was detected leaving the aerobic reactor was possibly an indication of a high death and decay rate of the high-SRT sludge, or possibly deamination of complex compounds.

Because, as with the other systems, the overall nitrogen balance yielded higher specific nitrification rate estimates than the nitrogen balance around just the aerobic reactor, only the overall observed specific nitrification rate is discussed. Despite the obvious ammonia breakthrough that occurred during operation period 6, a plateau in the overall observed specific nitrification rate plotted against the F:M ratio was not apparent (see Figure 26). There may be a slight shouldering effect, but there is not enough data to verify such a claim. As mentioned above, it is likely that the sludge might still have been adapting to the new conditions and had not reached steady state. Because the project was terminated before true steady state could be verified, it will be assumed that the maximum observed specific nitrification rate was the maximum specific nitrification rate that the sludge could have achieved. From the existing data, depicted in Figure 26, the apparent maximum overall observed specific nitrification rate was 0.18 mgN/mgVSS/day.

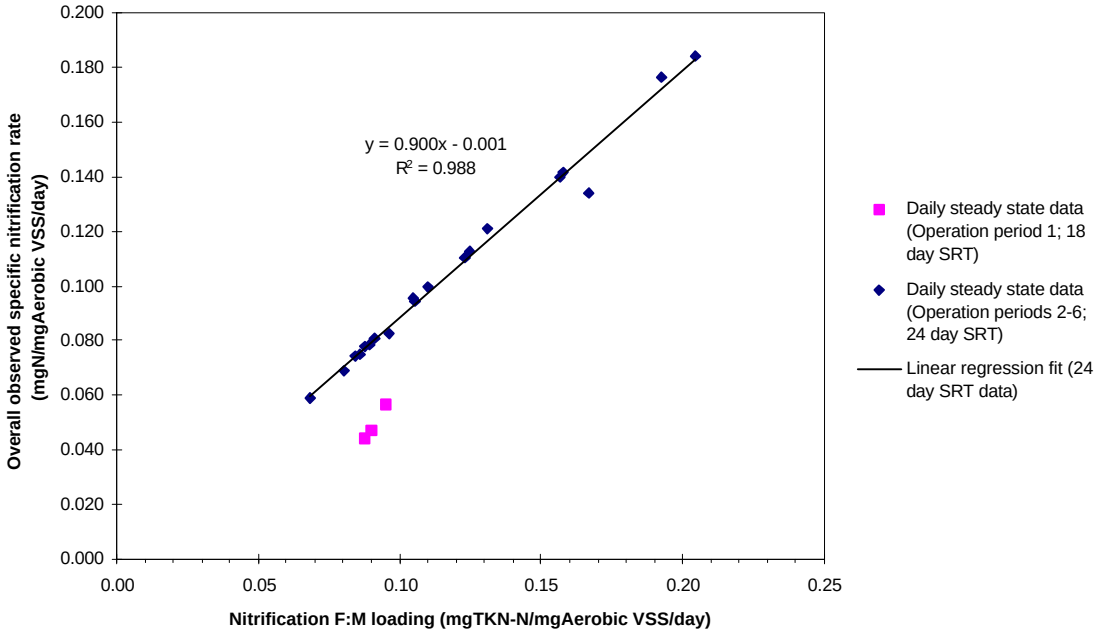

Figure 26. High ammonia side stream treatment train: Overall observed specific nitrification rate as a function of the F:M loading.

Table 20. High ammonia side stream treatment train: Aerobic reactor performance and loading.

Steady-state operation period	**1** 6/10-6/12	**2** 7/25-8/6	**3** 8/11-8/15	**4** 8/22-8/29	**5** 9/2-9/4	**6** 9/8-9/12
Aerobic HRT (hours)	3.1	3.1	3.0	2.4	2.1	1.5 0.0
SKN in (mg-N/day)	8821	5409	3333	3290	1789	13870 3647
SKN out (mg-N/day)	7021	2018	726	287	321	7773 3914
Bioavailable SKN in (mg-N/day)	8715	5364	3127	3157	1670	13606 3545
SKN in - SKN out (mg-N/day)	1800	3391	2607	3003	1468	5427 1639
SKN used for growth by the total system (mg-N/day)	698	358	179	426	400	465 89
SKN available for oxidation (mg-N/day)	8017	5006	2948	2731	1270	13105 3453
Estimated minimum SKN nitrified (mg-N/day)	1102	3033	2428	2576	1068	4926 1731
Measured NOx production (mg-N/day)	2871	3162	2501	2760	1636	5057 1372
Estimated minimum specific nitrification rate (mgSKN-N/mgVSS/day)	0.019	0.072	0.141 (ammonia limited)	0.047 (ammonia limited)	0.021 (ammonia limited)	0.080 0.041
Measured specific NOx production rate (mgNOx-N/mgVSS/day)	0.049	0.075	0.145 (ammonia limited)	0.050 (ammonia limited)	0.032 (ammonia limited)	0.088 0.035
% bioavailable SKN removed	21	67	83	95	88	43 23
Estimated minimum % bioavailable SKN oxidized	13	60	78	82	64	39 23
Estimated % oxidizable SKN nitrified	14	65	82	94	84	41 24
SKN in (mg-N/L)	63.1	38.1	22.9	18.1	8.5	48.7 8.9
SKN out (mg-N/L)	50.3	14.1	5.0	1.6	1.5	27.7 13.7
NHx out (mg-N/L)	48.3	8.5	0.0	0.0	0.0	25.5 14.5
COD in (mg/day)	5308	7582	12978	25466	13762	43206 17573
COD out (mg/day)	5867	5885	3471	2075	5461	15843 2375
COD in - COD out (mg/day)	-559	1697	9507	23391	8301	27362 16228
COD in (mg/L)	38	52	86	135	63	148 60
COD out (mg/L)	42	40	23	11	25	54 8

Note: There is only one data point per operation period for balances on the aerobic reactor, with the exception of operation periods 2 and 6 which had 2 and 3 data points, respectively. Thus, standard deviations are only applicable for operation period 6, and the are displayed as the lower values in that column.

Feasibility of denitrification of the High-Ammonia Side Stream

In terms of evaluating denitrification performance, the second anoxic reactor of the MUCT process and the only anoxic reactor of the A2/O process were analyzed as a control volume for characterization of denitrification. Their performance is described in Table 21. As with the aerobic reactor data, there was only one data point per operation period for operation periods 1, 3, 4, and 5. Considering denitrification was definitely COD limited during operation period 1 (as indicated by the fact that the COD exiting the anoxic reactor was the same as the COD of the final effluent) and definitely nitrate limited during operation periods 4 and 5 (as indicated by zero detectable nitrates exiting the anoxic reactor), it is not surprising that the observed specific denitrification rate in the anoxic reactor could not be correlated to COD loading, COD:NO_x loading, or the HRT of the anoxic reactor. Despite the small amount of data, a good trend for denitrification with respect to the ratio of COD to nitrate loading in the anoxic reactor was observed. No trend was observed when plotting the percent denitrification versus the anoxic HRT however.

Table 21. High ammonia side stream treatment train: Second (or only) anoxic reactor performance and loading.

Steady-state operation period	**1** 6/10-6/12	**2** 7/25-8/6	**3** 8/11-8/15	**4** 8/22-8/29	**5** 9/2-9/4	**6** 9/8-9/12
Configuration	MUCT	MUCT	MUCT	A2/O	A2/O	A2/O
Anoxic HRT (hours)	0.69	0.68	0.66	0.53	0.46	0.34 0.00
Methanol addition?	no	yes	yes	yes	yes	yes
Methanol loading to anoxic reactor (mgCOD/day)	0	6838	12992	17094	19487	26325 0
Total COD loading to anoxic reactor (mgCOD/day)	9530	13537	18521	25920	27122	47269 2090
NOx loading to anoxic reactor (mg-N/day)	2280	3544	2095	1643	975	3799 583
COD:NOx loading ratio to anoxic reactor	4.2	4.1	8.8	15.8	27.8	12.7 2.4
Amount of NOx exiting anoxic reactor (mg-N/day)	1643	2456	1026	0	0	1333 455
Concentration of NOx exiting anoxic reactor (mg-N/L)	11.8	17.2	7.1	0.0	0.0	4.7 1.6
Amount of COD exiting anoxic reactor (mgCOD/day)	5308	7582	12978	25466	13762	43206 17573
Concentration of COD exiting anoxic reactor (mg/L)	38	52	86	135	63	148 60
COD of final effluent (mg/L)	**39** 3 4	**35** 5 7	**33** 2 n/a	**35** 6 6	**33** 3 9	**48** 5
% NOx denitrified	28	34	51	100	100	64 15
Observed specific denitrification rate (mgNOx/mgVSS/day)	0.049 (COD limited)	0.115	0.278	0.135 (NOx limited)	0.085 (NOx limited)	0.192 0.083

Note: There is only one data point per operation period for balances on the anoxic reactor, with the exception of operation periods 2 and 6 which had 2 and 3 data points, respectively. Thus, standard deviations are only applicable for operation period 6, and they are displayed as the lower value in that column. More data was collected from the final effluent. The average effluent COD concentration is the top value listed, with the standard deviation given below and the number of samples to the right.

As shown in Figure 27, the percent of nitrates denitrified in the anoxic reactor increased with the COD:NO_x loading ratio until a COD:NO_x ratio of approximately 15 was achieved, at which point complete denitrification was achieved. Thus, although the COD exiting the anoxic reactor was greater than the COD of the final effluent for all operation periods except operation period 1 (see Table 21), denitrification for this wastewater appeared to be COD limited until a COD:NO_x ratio of 15 was reached. The percent denitrification versus the COD:NO_x ratio was apparently independent of the SRT, since data from operation period 1 (the 18 day SRT) appeared to fit the trend as well as the other operation periods (although it is only one data point).

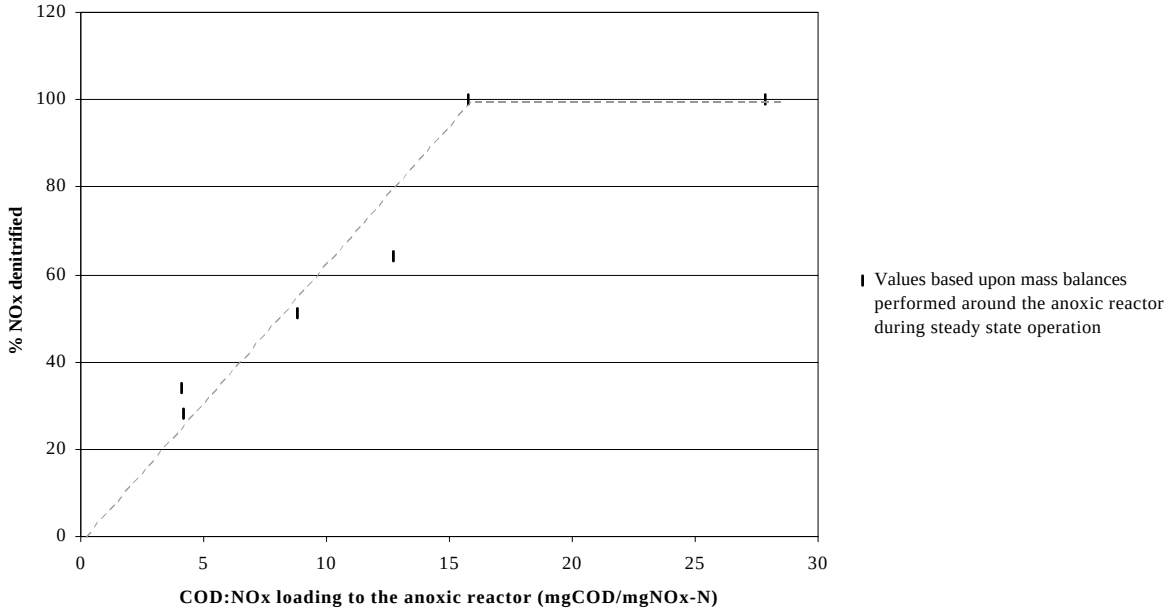

Figure 27. High ammonia side stream treatment train: Using the percent denitrification in the anoxic reactor as a function of the COD:NO_x loading ratio to the anoxic reactor to determine the critical COD:NO_x loading. The dashed curve represents the anticipated performance for this activated sludge and wastewater.

A ratio above 15 was observed during operation periods 4 and 5, and complete denitrification as defined by zero nitrates being detected exiting the anoxic reactor was achieved during both of those operation periods (see Table 21). The highest nitrate loading to the anoxic reactor that was completely denitrified was 1643 mg-N/day, observed during operation period 4. Although operation periods 1 through 3 corresponded to longer retention times than operation periods 4 and 5, denitrification was not complete during operation periods 1 through 3. Because those operation periods corresponded to lower COD:NO_x ratios, it is assumed that denitrification was COD limited. While operation period 6 corresponded to a shorter anoxic HRT, it too corresponded to a COD:NO_x ratio below 15; and it is thus unknown if denitrification was limited by insufficient contact time or COD. Evidence does suggest that denitrification was limited by contact time, however.

Nitrate breakthrough was observed during operation period 6, which was apparent because the effluent NO_x concentration rose from an average of 9.0 during operation period 5 to an average of 16.6 mg-N/L during operation period 6 (see Table 19). As with nitrification, performance was complete or very near complete on September 8 and 9, but the increase in the wastewater strength after September 9 led to substantial breakthrough. Effluent NO_x concentrations rose from 11.3 and 8.2 mg-N/L on September 8 and 9 to concentrations ranging from 20.4 to 22.6 mg-N/L during the next three days (see Appendix 18). There is no data from the anoxic reactor on September 8 or 9, but effluent nitrate concentrations measured on those days fall within the range of effluent nitrate concentrations

observed during operation periods 4 and 5, when it is known that complete denitrification had been achieved. Anoxic reactor data was collected on September 10 through 12, and a COD:NO_x ratio of 15.4 was observed on September 12. Denitrification was only 49% complete on that day (see Appendix 18). Thus, since denitrification was assumed not to be COD limited for ratios above 15, it appears that there was insufficient contact time to fully denitrify the nitrate loading to the anoxic reactor of 3193 mg-N/day on September 12 at the 0.34 hour anoxic HRT associated with operation period 6. The TKN loading to the overall system that day was 12,177 mg-N/day. Because significant ammonia breakthrough was also observed during operation period 6, it would appear that the anoxic and aerobic basins were well proportioned. Assuming adequate methanol were provided, the effluent nitrates of 9 to 10 mg-N/L observed during operation periods 4 and 5 could possibly be brought down to lower levels with higher nitrate recirculation rates.

A nitrogen balance was performed on both the anoxic reactor and the overall system to determine specific denitrification rate estimates, with higher estimates resulting from the overall system nitrogen balance. The F:M ratio is expressed in terms of the influent TKN loading normalized to the anoxic biomass. The anoxic biomass was estimated to be the biomass present in both anoxic reactors for the MUCT system and the single anoxic reactor biomass for the A2/O system. Despite the apparent nitrate breakthrough, there was not a plateau in denitrification rate curve depicted in Figure 28. The maximum observed rate will be assumed to be the maximum specific denitrification rate of the sludge for the treatment train tested, which was 0.71 mgN/mgVSS/day.

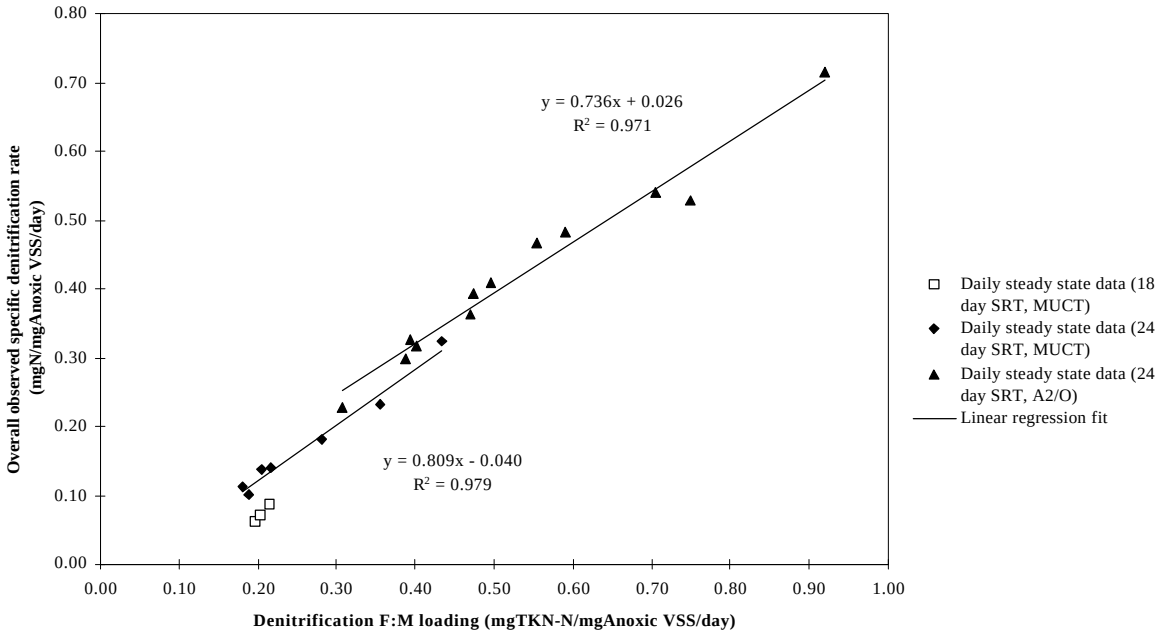

Figure 28. High ammonia side stream treatment train: Overall observed specific denitrification rate as a function of the F:M loading.

Other performance characteristics of the High Ammonia Side Stream treatment train

COD breakthrough also appeared to occur during the final operation period. Similar effluent COD concentrations were measured throughout the experiment, with steady state average soluble effluent COD concentrations ranging from 32 to 39 mg/L for operation periods 1 through 5. The effluent concentration during operation period 6 increased to an average of 48 mg/L, however (see Table 19). As shown in Figure 29, the observed overall specific COD consumption rate did not exhibit an obvious plateau however, suggesting that the

COD loading was only reaching the minimum critical value for the maximum observed overall specific COD consumption rate. Because the last day of operation period 6 corresponded to only one week at the last loading conditions, it is not known if the system could have acclimated and resulted in complete COD removal (or even nitrification and denitrification). As mentioned previously, the fact that the MLVSS was continuing to increase during that operation period (see Figure 25) indicates that acclimation was obviously not complete. Because acclimation to complete or near complete nitrification, denitrification, and COD removal was observed during all other operation periods generally within one week of a change in operating conditions, it will be assumed that the loadings associated with operation 6 were in excess of what the system could adjust to.

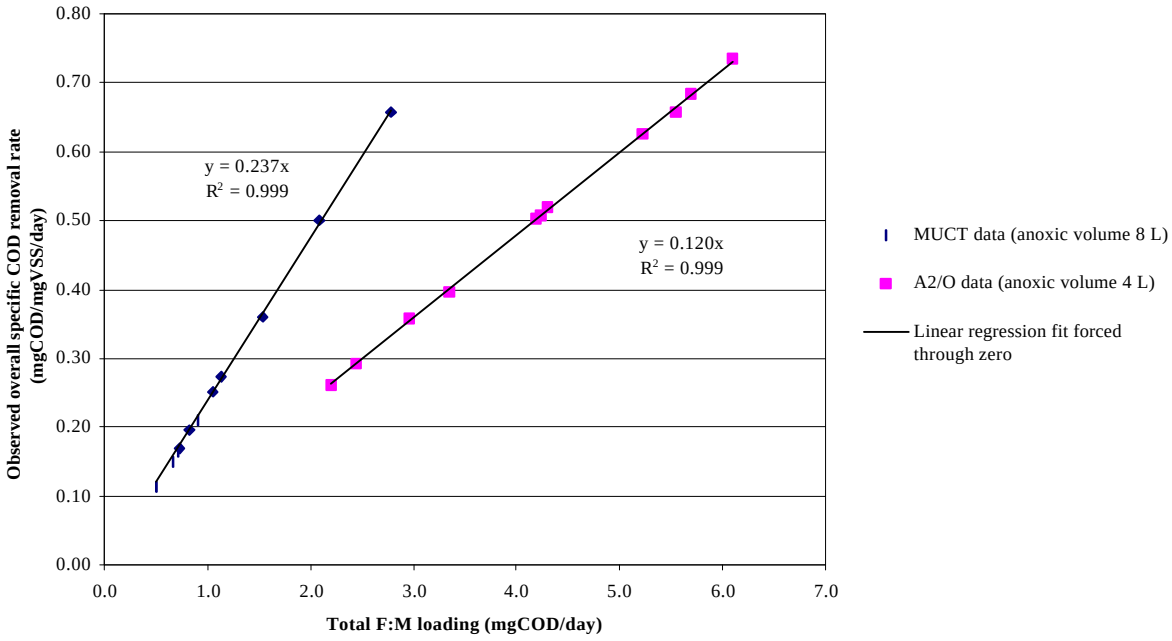

Figure 29. High ammonia side stream treatment train: Plotting the specific COD removal rate against the COD-based F:M loading to determine if COD breakthrough was occurring at higher loadings.

The ETSS concentrations were quite variable, with daily values generally fluctuating between 5 and 60 mg/L, although average ETSS values were in the range of 13 to 26 mg/L. No operation period had a particularly lower or less variable ETSS (see Figure 30 and Table 19), and with only 3 or 4 data points per operation period for most of the operation periods, it was not possible to separate out effects such as COD or TKN loadings or concentrations, or hydraulic loading. Thus, with relatively little difference in the average ETSS between all operation periods, the ETSS was assumed to be independent of factors such as hydraulic loading or nutrient loading, and it appears that both configurations used for the nitrification and denitrification of the high ammonia waste stream will simply tend to produce highly variable, but reasonable ETSS.

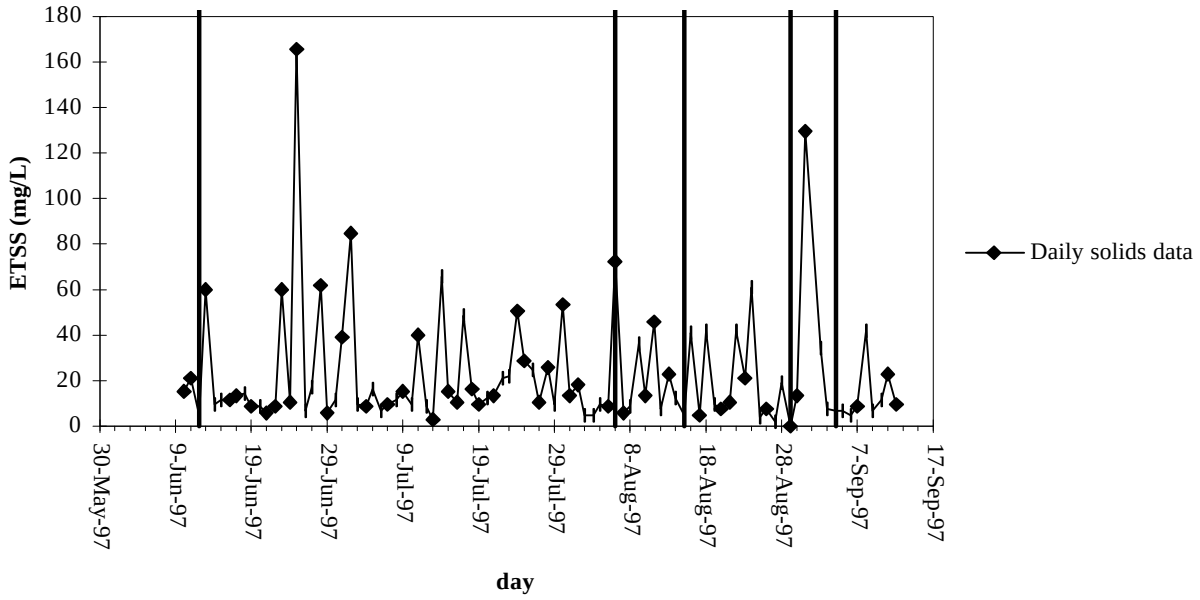

Figure 30. High ammonia side stream treatment train: Daily effluent suspended solids quality The bold lines indicate the last day of a steady state operation period.

High Ammonia Side Stream full scale design

From the data collected, nitrification, denitrification, and COD removal all appeared to fail simultaneously during operation period 6. It is not known if the system would have acclimated, but it will be assumed for the sake of discussion that the maximum specific removal rates achieved by the system were true maximum rates, which will be used for the full scale design. Design will be based upon 80% of the maximum overall observed specific nitrification and denitrification rates. Thus, a specific nitrification rate of 0.14 mgN/mgVSS/day and a specific denitrification rate of 0.57 mgN/mgVSS/day will be used for design.

The system tested tended to operate at an MLVSS of approximately 2700 mg/L, which will thus be assumed for the full scale design. The average TKN concentration of the side stream during this study was 130 mg-N/L. The hydraulic loading for the full scale design only needs to be scaled up to 6.5 MGD, because the industry contributing the high ammonia stream only contributes approximately 13% of the total flow received by HRWTF. Thus, scaling the hydraulic loading up to 6.5 MGD (24.6 million liters per day), an average TKN loading of 3.20 x 10^9 mg-N/day (7060 lb-N/day) is expected. Complete nitrification at the assumed specific nitrification rate would thus require 2.28 x 10^{10} mg of VSS. Assuming the 2700 mg/L MLVSS, an aerobic basin volume of 8.46 x 10^6 L, or 2.24 million gallons would be required. Assuming complete nitrification and complete denitrification, with the assumed denitrification rate of 0.57 mgN/mgVSS/day, an anoxic basin of 0.44 million gallons (1.67 x 10^6 L) would be required.

It was not determined if the anaerobic reactor was necessary for buffering against toxic loads, but it will be assumed that the anaerobic basin is valuable for both buffering against toxic loads and removal of excess nitrates on occasion. Thus, to include the anaerobic basin, scaled up in the same proportion as the ratio of the anaerobic and anoxic reactors in the treatment train tested, would require the addition of a basin 0.66 million gallons in size. Note that further testing would be required before selecting a full scale process, but it is quite likely that the anaerobic basin would not be necessary, and possibly an MLE process with a slightly larger anoxic basin than the one designed here may suffice. For the sake of discussion, the A2/O process will be assumed necessary, as it was the simplest process tested which yielded successful nitrification and denitrification of the high ammonia side stream. Therefore, the total basin volume for A2/O treatment of the 6.5 MGD high ammonia side stream would thus be 3.34

million gallons, with an overall HRT of 12.3 hours (not including final clarifiers). This is a very long retention time, but it is a high concentration of ammonia to be nitrified and denitrified.

With respect to sludge growth and wastage, because this was a low carbon, high ammonia waste stream and much of the COD would be due to methanol addition for denitrification, the amount of VSS grown per day was correlated to the influent TKN concentration, as depicted in Figure 31. Assuming the amount of VSS grown varies linearly with the TKN loading (although it is apparent that the relationship is not linear on all portions of the curve), one can anticipate observing 995 lbs of biomass production for every 10,000 lbs of TKN-nitrogen entering the system. Assuming an average TKN concentration of 130 mg-N/L (as was observed during this experiment) at the full scale 6.5 MGD flow, approximately 7060 lbs of TKN-nitrogen would enter the system per day, resulting in 700 lbs of biomass grown and wasted per day.

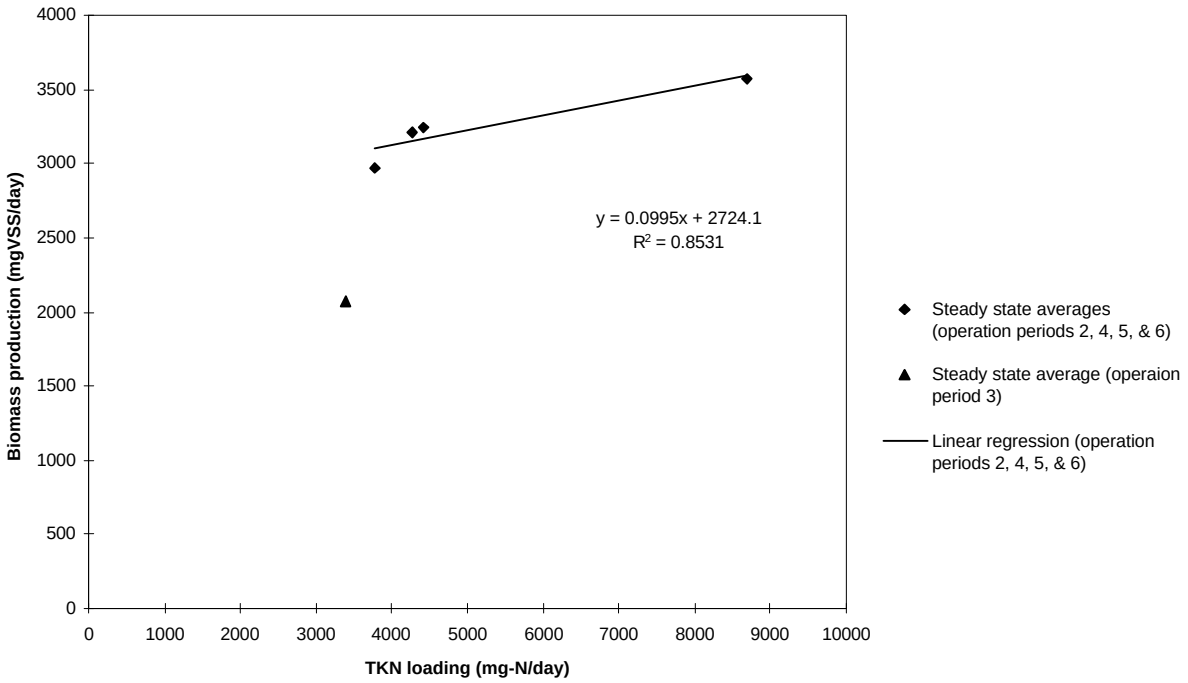

Figure 31. High ammonia side stream treatment train: Estimating biomass production for the 24 day SRT sludge as a function of the TKN loading.

V. SUMMARY OF RESULTS

The purpose of this bench scale research was to quickly determine the most feasible means of nitrifying and denitrifying the wastewater at HRWTF, with its present conditions and difficulties, in order to select the best options to test at the pilot scale. While the mixed stream (both the primary effluent and secondary effluent of HRWTF) was shown to be amenable to complete nitrification and denitrification at ambient temperatures, testing at the high temperatures indicated that either nitrification performance or effluent biosolids were likely to be unstable. The high ammonia side stream could also be fully nitrified, but denitrification down to a total nitrogen concentration of 10 mg-N/L or less was difficult to maintain due to the fact that a post nitrification process was used for a high ammonia wastewater.

For the sake of comparison, preliminary sizing of the three ambient temperature systems was performed, based upon observed specific nitrification and denitrification rates, although further testing is required to obtain performance estimates accurate enough for design purposes. The resulting systems are compared below in Table 22. The high temperature systems were not sized because further testing is required to determine if satisfactory and stable results can be obtained at the high temperatures of approximately 40^{o}C and above.

Table 22. Comparison of preliminary designs of the three ambient temperature systems.

Parameter	Aerobic/Anoxic Treatment of the Secondary Effluent	A2/O Treatment of the Primary Effluent	A2/O Treatment of the High Ammonia Side Stream*
Expected effluent SKN (mg-N/L)	1.0 or less	3.0 or less	4.0 or less
Expected effluent NH_x (mg-N/L)	0	2.0 or less	2.0 or less
Expected effluent NO_x (mg-N/L)	0.5 or less	6.0 or less	10.0 or less
Expected effluent COD (mg/L)	50	50	35
Maximum observed overall specific nitrification rate (mgN/mgVSS/day)	0.18	0.078	0.18
Maximum observed overall specific denitrification rate (mgN/mgVSS/day)	0.42	0.26	0.71
Design MLVSS (mg/L)	2500	3800	2700
Design SRT (days)	6	12	24
Total additional basin volume (not including final clarifiers) (million gallons)	8.39	6.85**	3.34***
Design total HRT (not including final clarifiers) (hours)	5.0	6.1	12.3

* The primary and secondary effluent systems treat the entire wastewater mixture, while the side stream treats only the one stream contributing 13% of the total flow at HRWTF. Therefore, the contribution to the mixed flow final effluent would only be approximately 13% of the expected effluent concentrations listed for the high ammonia side stream treatment train.

** The design incorporates using existing basins at HRWTF, the total basin volume required to treat the primary effluent of HRWTF is 12.61.

*** Treatment of the side stream is for 6.5 MGD rather than 50 MGD.

VI. CONCLUSIONS AND RECOMMENDATIONS

From this bench scale study, several facts were learned about the treatability of the wastewaters tested. Both the primary and secondary effluent of HRWTF could be consistently and completely nitrified at ambient temperatures. The high ammonia side stream could be fully nitrified at extant, moderate temperatures and did not require a MUCT detoxification process due to there no longer being MEKO in the effluent of the industry of interest. Nitrification of the high ammonia side stream was not as consistently completed to zero effluent ammonia concentrations, however, due to the wide daily variations in the wastewater strength.

Denitrification of the secondary effluent of HRWTF could be achieved to near zero effluent nitrates and total nitrogen concentrations well below 10 mg-N/L at ambient temperatures, but the addition of methanol was required for this carbon-deficient waste stream. Denitrification of the primary effluent of HRWTF could be achieved to total nitrogen concentrations less than 10 mg-N/L on a consistent basis at ambient temperatures. Although the primary effluent at HRWTF may occasionally be slightly carbon deficient for complete denitrification, effluent nitrogen concentrations were still sufficiently low during testing, and the occurrence of low carbon concentrations appeared to be rare. The high ammonia side stream could be successfully denitrified to a great extent with methanol addition, but total effluent nitrogen concentrations were often above 10 mg-N/L. Because the anoxic reactor was generally nitrate limited, the higher final effluent nitrates were most likely due to the fact that the system was a post nitrification process treating a high ammonia wastewater. A nitrate recirculation rate greater than the 300% tested might have alleviated the problem to a limited extent.

The primary effluent could be consistently completely nitrified at extant higher temperatures (of 45^{o}C and higher) with an activated sludge which had been gradually acclimated from 30^{o}C to 45^{o}C over the course of two months. The high temperature study conducted on the secondary effluent treatment train did not entail the use of an activated sludge acclimated to high temperatures, but, rather, simulated the existing sudden increase in temperature that occurs every spring at HRWTF. Nitrification was quickly lost from the system and could not recover during the two-week high temperature study despite the addition of biomass on two occasions. Nitrification of the heated secondary effluent treatment train did appear to begin recovery within 24 hours of reducing the temperature back to ambient conditions, suggesting that temperatures of 40^{o}C and higher suppress nitrification activity but do not necessarily kill the organisms responsible for nitrification. A longer time of testing might have shown a shift toward an acclimated culture capable of nitrifying at the high temperatures, but several weeks of high effluent nitrogen concentrations would not be an acceptable occurrence in an actual treatment plant.

Although no attempts were made to isolate out the organisms responsible for nitrification in each of the systems during this project, it is possible that different organisms were involved with the ambient temperature nitrification and the high temperature nitrification. Thus acclimation to the high temperatures might have been due not to acclimation by the cooler nitrifiers, but rather to a population shift from cooler temperature nitrifiers (such as, perhaps *Nitrosomonas* and *Nitrobacter*) to higher temperature nitrifiers (such as, perhaps, methanotrophic organisms).

Denitrification of the heated primary effluent treatment train could be achieved to effluent total nitrogen concentrations less than 10 mg-N/L on a consistent basis shortly after the addition of the anoxic reactor to the process. Denitrification of the heated secondary effluent treatment train was consistently at or near zero, although far fewer nitrates were being produced in the aerobic process. The continued complete denitrification after the sudden increase does indicate that the denitrifying population could acclimate at least as well as the nitrifying population and probably better than the nitrifying population given that the relative percent of nitrogen nitrified decreased after the temperature increased while the percent of nitrates denitrified remained constant at 100%.

A better approach for both high temperature studies, which was not tested, would have been to acclimate the cultures to the high temperatures and operate them for several months at the high temperatures, followed by reducing the temperatures for three weeks and then increasing them to the high temperatures, to simulate existing conditions at HRWTF as best as possible. Limitations on time and resources prior to selecting pilot scale strategies prevented such testing. From the existing data, with the sporadic ETSS of the heated primary effluent treatment train and the quick disappearance of nitrification activity in the heated secondary effluent treatment train, successful nitrification and denitrification at the present high temperature conditions at HRWTF seems unlikely.

While all three waste streams could be consistently nitrified and denitrified, several factors should be considered and tested before a final concept for HRWTF should be selected. High temperature studies as described above should be conducted on the primary and secondary effluents. Given that the primary effluent was not inhibitory to nitrification and that substantial savings in land space, capital costs, and chemical (methanol) costs would be realized by treating the primary effluent, rather than adding a tertiary treatment to HRWTF, it seems likely that treating the primary effluent is a better option. It is also necessary to determine if year-round nitrification and

denitrification could be maintained with the present high temperatures and annual temperature fluctuations, or if construction of a cooling tower would be necessary. Possible problems with effluent solids might also require a specially designed or larger clarifier, or even an effluent filtration process.

Another option which might yield the desired effluent nitrogen goals for HRWTF would be to treat the moderate-temperature high ammonia side stream separately. Although total effluent nitrogen concentrations could not be kept below 10 mg-N/L as consistently, there may be solutions to such a problem. Simply increasing the nitrate recirculation rate above 300% may suffice. Given the wide fluctuations in the ammonia concentration of the plant, a better solution is likely to involve mixing flows. Combining the high ammonia side stream with the domestic wastewater which enters HRWTF (contributing approximately 15% of the total flow at HRWTF), for instance, would cut ammonia spikes roughly in half and may even provide enough carbon to eliminate the need for methanol addition. Carbon and nitrogen data for all the waste streams received by HRWTF are obviously necessary to determine the best flows to combine with the high ammonia side stream. Diluting the side stream with another stream such as the domestic wastewater received by HRWTF would roughly double the basin size requirements, yielding similar land space and capital cost requirements as those for treatment of the primary effluent. The significant benefit is the elimination of the high temperature issue, however. If the combined nitrogen loading of all other waste streams is low enough, mixing the high ammonia side stream with another stream for more complete denitrification might be a non-issue, because diluting out the nitrified and denitrified side stream with the combined total flow at HRWTF might result in a total effluent nitrogen concentration of less than 10 mg-N/L. Data to determine if such a strategy is possible were not collected, but if such a strategy is possible, treatment of the high ammonia side stream alone (without mixing with another flow), would result in further land space and capital cost savings by requiring the addition of only approximately half the basin volume required to treat the total primary effluent (although methanol addition would definitely be required).

It was not determined if the anaerobic zones incorporated in the primary effluent and high ammonia side stream systems were truly necessary for detoxification, but they are certainly desirable to keep in the designs because of the potential to buffer occasional toxic spikes or to act as extra anoxic volume during high ammonia (and therefore nitrate) loads. Thus, although consistent nitrification and denitrification down to effluent TN concentration of 10 mg-N/L or less at HRWTF is definitely possible, it is not clear with the data presented in this thesis what the best strategy is. The optimum solution, in terms of both effluent quality and cost considerations, will most likely involve either treatment of the primary effluent of HRWTF with an A2/O process and possibly a cooling tower or A2/O treatment of the high ammonia side stream possibly combined with one or more other streams. The elimination of high temperature concerns suggests that the latter option may be the most desirable one.

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