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REDUCING OPERATING COST WITH ANAMMOX IN WASTEWATER TREATMENT – A SIMULATION STUDY

by

Eliav Jordan Eini, P.Eng Bachelor of Engineering, 2003, Ryerson University

A Major Research Project presented to Ryerson University

in partial fulfillment of the requirements for the degree of Master of Engineering In the Program of Chemical Engineering

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Author's Declaration

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<u>Abstract</u>

A simulation study is presented to investigate the use of the Anammox process for removing nitrogen compounds from wastewater to reduce operating cost. The literature review of technologies for removal of Total Nitrogen includes an overview of the biological nitrogen cycle, the discovery of Anammox, bioreactor design, operational strategies, and start-up of full-scale processes. A facility of 656 MLD with influent loading of 35 mg/L as NH3-N and 250 mg/L as BOD is used as a basis of the simulation study. Preliminary bioreactor sizing calculations are developed for six configurations. Subsequently, eight whole-plant simulation cases are compared to demonstrate operational savings for removal of Total Nitrogen. For the sample facility, this is shown to be on the order of \$460 to \$680K/year while attaining 66-70% Total Nitrogen removal and reduced greenhouse gas emissions by 2 kt CO₂/year. The project concludes with ideas about future development of the Anammox process.

Acknowledgements

I would like to acknowledge the following people for their role in helping complete this project; my project advisor, Dr. M. A. Cuenca for encouraging me to study something new; my colleagues at Toronto Water's Operations Efficiency unit (Abhay Tadwalkar, Susan Atlin, Jessie Cheng, and Dave Scott) for their technical advice and support; staff from CH2M Hill (George Crawford, Tim Constantine, and Tom Johnson) for sending resources on Anammox and trouble-shooting CHEApet.

Dedication

For my wife, family, and many friends.

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List of Acronyms

AerAOB	Aerobic Ammonia Oxidizing Bacteria
AMO	Ammonia Monooxygenase
AnAOB	Anaerobic Ammonia Oxidizing Bacteria
AnMO	Anaerobic Methane Oxidation
AOB/NOB	Ammonia Oxidizing Bacteria / Nitrite Oxidizing Bacteria
AOR	Actual Oxygen Requirements
ASP	Activated Sludge Process
AST	Advanced Secondary Treatment
BOD (COD)	Biological (Chemical) Oxygen Demand
CANON	Completely Autotrophic Nitrogen removal Over Nitrite
CCME	Canadian Council of Ministers of the Environment
CMAS	Complete Mix Activated Sludge
DeAmmon	Nitritation and Anammox in one reactor in a biofilm system
DEAMOX	DEnitrifying AMmonium OXidation
DEMON	DE-amMONification
DENAMMOX	DENitrification-anAMMOX process
DIB	Deammonification in Interval-aerated Biofilm systems
MBR	Membrane Bioreactor
MMO	Methane Monooxygenase
NAS	Novel Activated Sludge
NRR	Nitrogen Removal Rate
OLAND	Oxygen-Limited Autotrophic Nitrification Denitrification
SDNR	Specific DeNitrification Rate
SHARON	Sustainable High rate Ammonium Removal Over Nitrite
SNAP	Single-stage Nitrogen removal using the Anammox and Partial nitritation
STAR	Stirred Tank Anammox Reactor
TAN	Total Ammonia Nitrogen
TKN	Total Kedjhal Nitrogen
WLA	Waste Load Allocations

Executive Summary

The Anammox process is a relatively new biological treatment method for the removal of Total Nitrogen from wastewater. The unique physiology of the Anammox bacterium and its previously-unrecognized role in the global nitrogen cycle mean that novel engineered systems for the simultaneous removal of ammonia and nitrite are now being implemented at full-scale. The Anammox process is particularly well-suited to the treatment of side-streams, such as dewatering centrate that result from the treatment of wastewater. Side-stream loads that have high nitrogen content compared to organic carbon are difficult to treat using conventional biological Total Nitrogen removal methods.

The project entailed a literature review and simulation study of plant design concepts that make use of novel biological Total Nitrogen removal technologies. The research objectives were to:

- Examine the microbial mechanisms of nitrogen converters, including the Anammox bacterium;
- Investigate conventional and emerging biological Total Nitrogen removal technologies;
- Investigate side-stream treatment configurations that use the Anammox process; and
- Estimate the impact of adding side-stream Anammox treatment on the overall plant Total Nitrogen removal, operating cost, and greenhouse gas emissions.

The underlying microbial metabolic pathways of the nitrogen cycle are presented, including the Anammox short-cut. The key design considerations for conventional and emerging biological Total Nitrogen removal technologies are discussed. A short overview is given to clarify the differences between the prevalent technologies for ammonia and nitrate removal, including processes such as SHARON, CANON, DEMON, and DeAmmon.

A simulation study was conducted for a sample facility of 656 MLD with an influent ammonia load of 35 mg/L as NH₃-N and 250 mgl/L as cBOD was used as a basis to develop various configurations for treatment, with and without the side-stream Anammox process. The basis of comparison was an Advanced Secondary Treatment (AST) plant with full nitrification for a baseline performance target of 10 mg/L as NH₃-N and 2.9 mg/L as NO₃-N. The best choice for the removal of Total Ammonia from a municipal wastewater treatment plant effluent was the Biological Nitrogen Removal (BNR) process coupled with side-stream Partial Nitritation-Anammox:

- <u>Total Operating Cost Reduction</u>: The addition of the side-stream Anammox process to a BNR plant expected to save \$1.04 to \$2.30/ML treated. The cost avoidance for baseline treatment is projected be \$460,739 to \$680,652 per year over an AST plant that is fully nitrifying.
- <u>Total Nitrogen Removal</u>: The biological nitrogen plant with side-stream treatment is expected to increase Total Nitrogen removal to 66%-70% for baseline treatment. Optimized treatment can attain 81%-84%, the highest level of Total Nitrogen removal for all treatment configurations that were considered.
- Total Greenhouse Gas: The addition of the side-stream Anammox process is estimated to reduce Total Greenhouse Gas emissions by 2,000 tonnes of CO₂ per year.

1 Introduction

Anaerobic ammonia oxidation, otherwise known as the Anammox process, is a biological treatment method for the removal of Total Nitrogen from wastewater. The unique physiology of the Anammox bacterium and its previously-unrecognized role in the global nitrogen cycle mean that novel engineered systems for the simultaneous removal of ammonia and nitrite are now being implemented at full-scale (Strous, Kuenen, Fuerst, Wagner, & Jetten, 2002). The Anammox process is particularly well-suited to the treatment of side-streams, such as dewatering centrate with high nitrogen content compared to organic carbon (Hippen, Helmer, Kunst, Rosenwinkel, & Seyfried, 2001).

The bacteria responsible for Anammox were unknown until the 1990s, but their importance in regulating the global nitrogen cycle is now recognized (Strous, et al., 1999). The Anammox process was first discovered in marine and freshwater ecosystems, and can be cultivated for the biological nutrient removal (BNR) of nitrate and ammonia from wastewater. When configured as a side-stream treatment reactor, the Anammox process can be used to reduce operating costs and greenhouse gas emissions compared to a conventional BNR process without side-stream treatment (Loosdrecht, 2008). The design of the Anammox reactor must be able to accommodate a long biomass residence time due to the slow growth of the Anammox bacteria (Johnson, Sanjines, Castaneda, & Daigger, 2011).

The development of the Anammox process, from discovery to application as a treatment process for the removal of Total Nitrogen from wastewater, occurred in the span of five decades (Strous & Jetten, Anaerobic Oxidation of Methane and Ammonium, 2004). The existence of anaerobic ammonia oxidation was first postulated 1960s and 1970s. The first evidence of bacterium in nature was demonstrated in the 1980s, leading to identification of the active microorganisms in the 1990s. The Anammox bacteria was successfully cultivated at lab scale in the 2000s, culminating in the present day full-scale implementation of Anammox bioreactors for the treatment of high-ammonia/low-substrate side-streams for wastewater plants in Europe, Asia and North America.

At present, examples of full-scale Anammox reactors are limited to side-stream treatment units. The development of a robust, high-efficiency nitrogen removal process using Anammox for side-stream treatment is an important advance in wastewater treatment by reducing energy consumption when compared to a plant that treats the centrate in the mainstream BNR process (Desloover, et al., 2011).

Although biological removal of Total Nitrogen from wastewater is a preferred method for the removal of nitrogen compounds, the operating costs for oxygen and chemical usage will likely increase as a result of the application of nitrification and denitrification processes. Continuous improvement in engineered systems and investment in construction and renewal needs for infrastructure will open new opportunities to apply more efficient biological processes such as Anammox for removal of Total Nitrogen from wastewater.

1.1 Research Objectives

The primary objectives of this research project were to:

- 1. Examine the fundamental mechanisms of biological removal of Total Nitrogen, with emphasis on the bacterium responsible for the Anammox process;
- 2. Investigate biological Total Nitrogen removal technologies used for wastewater treatment, with emphasis on the considerations for design of an Anammox bioreactor process;
- 3. Investigate side-stream treatment as an option to remove Total Nitrogen from the side-stream before the main plant with focus on novel configurations that use the Anammox process; and
- 4. Estimate the impact of adding side-stream Anammox treatment on the plant's ability to remove Total Nitrogen, operating cost for aeration and chemical demand, and greenhouse gas emissions.

1.2 Structure of the Report

The structure of this report commences with a literature review followed by a presentation of plant design concepts for wastewater treatment using a side-stream Anammox reactor. Next, a short background is provided for basic familiarity with the biological Total Nitrogen removal process for wastewater treatment. The portion of the global nitrogen cycle pertaining to the aquatic environment is described in terms of the various metabolisms of nitrogen-converting microorganisms that are pertinent to wastewater treatment. The Anammox bacterium is introduced, along with its unique physiological characteristics.

From the microscopic world of nitrogen-converters, emphasis is shifted to the full-scale implementation of biological Total Nitrogen removal technologies, both conventional and novel. Wastewater quality parameters and key considerations for the design of mainstream and side-stream nitrogen removal reactors are reviewed. The literature review concludes with an overview of operational strategies for start-up and stable operation of full-scale Anammox reactors.

Various plant design concepts were developed to estimate the impact of different treatment configurations on plant performance in terms of nitrogen removal, cost, and greenhouse gas emissions. The plant design concepts are presented for a comparison between a conventional treatment plant versus one that uses biological nutrient removal and side-stream Anammox treatment. To demonstrate operational savings resulting from side-stream treatment with the Anammox process, the effectiveness was defined by the following indicators:

- 1. Total Nitrogen Removal, as % of Total Nitrogen removal (% TN)
- 2. Operating Cost, as cost per ML treated for power and chemical consumption (t CO₂/ML)
- 3. Facility Greenhouse Gas Emissions, as tones of CO₂ emitted per ML treated (t CO₂/ML)

The plant design concepts were developed in two phases, sizing and simulation. Kinetic models based on growth and decay were used to design reactors heterotrophs, denitrifiers, and Anammox bacteria. Reactors for the main plant stream were developed for carbonaceous and nitrogenous removal. Simplifying assumptions were made to design side-stream reactors. The reactor sizing was followed by software simulation using CHEApet to evaluate the different treatment configurations. As part of this analysis, simulations are presented for baseline and optimized treatment scenarios.

2 Background

2.1 Nitrogen Compounds in Wastewater

Nitrogen compounds naturally occur in wastewater as a product of decomposition of organic matter, in which nitrogen is combined in proteinaceous matter and urea. In addition to natural sources, nitrogen compounds enter the wastewater from a long list of anthropogenic sources, from fertilizers in agricultural run-off, from livestock waste, leachate from solid waste sites, and from industries - particularly textile, health care, and manufacturers of cleansing agents and detergents (Canadian Council of Ministers of the Environment, 2010).

During wastewater treatment, further organic nitrogen is released from the biomass during the anaerobic digestion process. The age of wastewater is indicated by the relative amount of ammonia that is present. The predominance of nitrate over ammonia indicates that that the oxygen demand for aerobic nitrifiers has been satisfied. However this oxygen demand consumes the dissolved oxygen present in wastewater, which can negatively impact the aquatic environment.

Forms of nitrogen compounds in wastewater include free ammonia (NH₃), ammonium ion (NH₄⁺), nitrite (NO₂⁻), nitrate (NO₃⁻), and organic nitrogen. The fractions of total nitrogen are present in both soluble and particulate forms, as well as biodegradable and non-biodegradable portions. Particulate organic nitrogen degrades more slowly than the soluble fraction, and impacts the treatability of the wastewater.

The predominant drivers for nitrogen removal from wastewater are concerns for the aquatic environment and for human health. A strategy for the total removal of nitrogen may be required to meet legislative or other limits for Total Nitrogen release in the effluent of the wastewater plant.

2.2 Impact on the Aquatic Environment

The release of nitrogen compounds has a negative impact receiving body of water and may have consequences for the aquatic habitat. The release of ammonia can be toxic to some forms of aquatic life. Other aquatic organisms, such as algae, may be limited in their growth until a nutrient such as ammonia or nitrate leads to an episode of explosive growth.

Ammonia is acutely and chronically toxic for fish, aquatic plants and benthic invertebrates. The chronic toxicity of ammonia to fish leads to reduced reproductive capacity and reduced growth of young, and acute toxicity may cause death. These definitions are covered in more detail in Section 2.4

Human activity has dramatically increased the release of nitrogen compounds to the environment, resulting in a host of related problems. Increased concern about the negative impacts of nitrogen release on the environment and on human health have led to pollution regulations for ammonia in some places. However, these regulations currently do little beyond the converting ammonia into non-acutely toxic nitrate (Wang, Hamburg, Pryor, Chandran, & Daigger, 2011). Consideration should be given to the indirect impacts of nitrogen removal treatment at wastewater plants. The widespread implications for the wastewater treatment sector in terms of energy consumption and greenhouse gas emissions are briefly considered, if, for example, the legislative discharge limits for nitrogen in wastewater became more stringent.

The reported effects resulting from ammonia and nitrates are for protection from direct toxic effects, and do not consider the indirect effects due to eutrophication. Nitrogen in the form of nitrates and ammonia and organic nitrogen from agricultural, livestock, municipal, and industrial wastes can cause blooms of algae and aquatic plants that can cover the surface of lakes, both small and large.

Indirect effects of the release of nitrates include algal blooms, eutrophication, and "red tides" that results from increased availability of nutrients. The increased global occurrence of algal blooms is a visible indication of the consequences of inadequate environmental stewardship.

Eutrophication is the excessive growth of algae that can lead to the decline of a water body. Nitrates are nutrients for phytoplankton, and can be precursors to rapid algae growth. Eutrophication reduces dissolved oxygen in the water column and can have toxic effects on aquatic organisms. The occurrence of eutrophication depends on a number of factors, so it is not a problem generally associated with every municipal wastewater discharge. Algal blooms may also result in the production of natural toxins that are harmful to other organisms. These "red tides" caused by Harmful Algal Blooms (HABs) are often associated with large-scale marine mortality events.



Figure 1: (a) nitrates lead to eutrophication of a lake¹; (b) satellite image of the worst algal bloom that Lake Erie has experienced in decades (Oct 2011)²

Other side effects of nitrogen released in the form of free ammonia is the formation of chloramines in wastewater effluent, which reacts with natural organic matter (NOM) in the aquatic environment and forms disinfection byproducts such as THMs, in comparison with the free chlorine form (Environment Canada, 2004). Additionally, nitrates exert a chlorine demand on the effluent that may result in chemical cost overruns.

2.3 Human Health Effects

The relevance to human health from nitrogen discharges must also be considered. Ammonia is predominantly an aesthetic concern for drinking water. Nitrate is converted in the human body to nitrite, which interferes with the oxygen-carrying capacity of the blood. Excessive levels of nitrate in drinking water have caused serious illness and sometimes death in infants from a condition called methaemoglobinaemia. This condition is also known as blue-baby syndrome and results from the shortness of breath.

Nitrate and nitrate have not been linked directly to cancer in animal studies. However, the conversion of nitrate in the body to nitrite may react with certain foods containing nitrosamine to form *N*-nitroso compounds, some of which are potent carcinogens in animals and could cause bladder cancer in humans (Canadian Council of Ministers of the Environment, 2010).

The "red tides" attributed to algal blooms affect more than the marine environment; the excessive algae and seaweed that wash ashore during these events rot and release potentially toxic gases such as hydrogen sulfide (H_2S) and methane (CH_4), such as the blooms in Northern France that were attributed to more than 3 livestock deaths (Chrisafis, 2009).

¹ http://environmentalgeography.wordpress.com/2010/01/27/eutrophication

² Adapted from (EarthSky, 2011)

2.4 Indirect Environmental Impact

Human activity has dramatically increased the release of nitrogen to the environment, resulting in a host of related problems. Increased concern about the negative impacts of nitrogen release on the environment and on human health have led to pollution regulations for ammonia in some places. However, these regulations currently do little beyond the converting ammonia into non-acutely toxic nitrate (Wang, Hamburg, Pryor, Chandran, & Daigger, 2011). Consideration should be given to the indirect impact of nitrogen removal treatment at wastewater plants. The widespread implications for the wastewater treatment sector in terms of energy consumption and greenhouse gas emissions are briefly considered, if, for example, the legislative discharge limits for nitrogen in wastewater became more stringent.

In 2000, 80% of the U.S. population was served by centralized wastewater treatment plants, with only 1% of the 80% providing biological nutrient removal. In the U.S., advanced secondary treatment plants treat around 35% of overall wastewater flow. Wastewater treatment plants consume about 3% of the electricity generated in the U.S. If conventional technologies for biological nutrient removal were more widely implemented, the electricity consumed would increase. Therefore, there is good cause to promote the development of novel processes that are energy-efficient.

In terms of the greenhouse gas emissions generated by the wastewater treatment sector, it has been reported that N₂O emissions from WWTPs in terms of percent of influent N may range from less than 1% to as high as 15%. Certain operating conditions have been identified as leading to high emissions, such as non-optimal aeration and insufficient residence times in the bioreactor. There is opportunity to reduce N₂O emissions through retrofits of AST plants at relatively low cost and additional benefits to the environment (Wang, Hamburg, Pryor, Chandran, & Daigger, 2011).

2.5 Regulatory Framework in Canada

The environmental and health impacts are the basis of the regulatory framework that limits the concentration of nitrogen compounds in the effluents of wastewater plants. This section provides an overview of applicable regulations for the release of ammonia and nitrate in wastewater plant effluent, with emphasis in Canada and in Ontario in particular. A brief survey of the regulations around the world reported various discharge limits is presented in Table 1.

Table 1: Survey of Global Total Nitrogen Removal Requirements³

EU countries	70-80% N removal
Dubai	15-40 mg/L TN
Australia	5-15 mg/L TN
US	3-10 mg/L

³ Adapted from (Johnson, Sanjines, Castaneda, & Daigger, 2011).

Canada has regulations for both drinking water quality and discharges to water for ammonia and nitrate. For drinking water, Health Canada has established a Maximum Allowable Concentration (MAC) of and 45 mg/L as NO_3^- (or 10 mg/L as NO_3^-N , and the concentration of nitrite should not exceed 3.2 mg/L as NO_2^- (Health Canada, 1987). Although there is no MAC for ammonia in drinking water, estimated thresholds for ammonia in drinking water are 35 mg/L (as NH_3) for taste and 35 µg/L for odour (Health Canada, 1987).

In 2003 the federal government of Canada published a Proposed Notice regarding upcoming regulations regarding chlorinated effluent, chloramines, and ammonia in wastewater effluent. The federal *Guideline for the Release of Ammonia Found Dissolved in Wastewater Effluent* is applicable for a wastewater system that discharges more than 5,000 m³/day (Environment Canada, 2004). In 2009 the Canadian Council of Ministers of the Environment (CCME) endorsed a Canada-Wide Strategy for the management of Municipal Wastewater Effluent (MWWE) to serve as a national benchmark to ensure baseline level for all jurisdictions in Canada. The proposed National Wastewater Systems Effluent Regulation (NWSER) under the federal Fisheries Act would establish national effluent quality standards for BOD, suspended solids, total residual chlorine, and requires the effluent to be non-acutely toxic for ammonia (Canadian Council of Ministers of the Environment, 2009). At the time of this writing the CCME guideline has not yet been adopted by the provinces. However, the *Canadian Water Quality Guidelines for the Protection of Aquatic Life* has published recommended limits for pollutants including ammonia, nitrite, and nitrate.

The ecotoxicity test for ammonia included in the proposed NWSER is the Environment Canada 96-hour acute lethality test. Rainbow trout are used as the indicator specimen for the assessment of acute toxicity. An effluent is considered to be pass the test if no more than half of the fish die during the 96 hours. If a sample fails a test, investigation into the cause (which may not be related to ammonia toxicity) is initiated (Novak & Holtze, Fall 2010). Ammonia is chronically toxic in concentration of unionized ammonia of more than 0.019 mg/L (Environment Canada, 2004). The water quality guidelines for the nitrate ion from the *Canadian Guidelines* for freshwater is 13 mg/L as NO₃. This is equivalent to 2.9 mg N/L as NO₃.

The federal guideline has not yet been adopted by the provinces. In the province of Ontario, the Ministry of the Environment regulates wastewater effluents. The effluent discharge limits for nitrogen are based on the existing quantity of nitrogen compounds in the receiving body. Whether the federal guideline will be adopted by the provinces and territories remains to be seen. Discharge limits typically vary by watershed, however, the acceptable limits are generally becoming more stringent in response to increasingly impacted water resources and public interest. For example, new regulatory initiatives in the U.S. have established waste load allocations (WLAs) for Total Nitrogen (TN) and total phosphorus for all point sources in Virginia (Johnson, Sanjines, Castaneda, & Daigger, 2011).

It is an interesting exercise to ask if the plant of today will be sufficient to meet discharge requirements in the mid- to long-term (e.g. 10 to 50 years). A comprehensive roadmap for the continuous improvement of wastewater treatment infrastructure should consider the emerging treatment technologies such as Anammox to improve removal of nitrogen from the plant effluent while lowering overall treatment cost and greenhouse gas emissions. The advantage of using the Anammox process is demonstrated in Section 6.

2.6 Technologies for Removal of Nitrogen Compounds

A variety of physiochemical and biological methods for the removal of ammonia are possible. For example, physicochemical methods include air-stripping and breakpoint chlorination for nitrogen removal. However, biological processes are preferred over physicochemical nitrogen recovery on the basis of cost for wastewaters that contain less than 5 g-N/L (Clippeleir, Yan, Verstraete, & Vlaeminck, 2011). The technologies pertaining to nitrogen removal are discussed in the following sections. Conventional Biological Nitrogen Removal (BNR) systems are compared to emerging technologies for nitrogen removal, such as pretreatment of the side-stream using an Anammox process.

2.6.1 Conventional Systems

The conventional system for biological Total Nitrogen removal is a modification of the conventional activated sludge plant (ASP), which includes an aerated reactor for BOD-removal and a subsequent clarifier from which sludge is recycled or wasted. As an alternative to the aerator-clarifier system, a membrane bioreactor (MBR) system may be used. An activated sludge plant that achieves may achieve partial or full nitrification (removal of the ammonia), especially in summer months when nitrifier growth rate is higher. For such an Advanced Secondary Treatment (AST) plant, the effluent of a nitrifying wastewater plant achieves low ammonia concentration, but if the nitrate concentration is considered then it becomes apparent that ammonia has been transformed from to nitrate, but not removed completely.

The conventional biological Total Nitrogen removal plant is designed for both nitrification and denitrification. By supplying dissolved oxygen and alkalinity to the reactor, the aerobic reactor converts ammonia to nitrate by biological nitrification. The anoxic reactor denitrification system, which must be kept well-mixed and may require a supplemental source of organic carbon, converts the nitrate to nitrogen. Compared to a wastewater plant that is removing only BOD, additional oxygen is required for the oxidation of ammonia to nitrate and results in increased energy consumption. The cost for the extra oxygen is offset by the denitrification process, which uses nitrate as a source of oxygen instead. Ultimately, the increased treatment efficiency increases the treatment cost, but by how much will the operating costs for power and chemical costs will increase as a result of nitrogen removal? This question is explored in greater detail in Section 6 Plant Design Concepts for Side-stream Treatment Using Anammox.

2.6.2 Side-stream Treatment

Side-streams are flows that are generated within a wastewater treatment plant, typically from solids processing, which contain nutrients, solids, organic or inorganic constituents. Anaerobic digestion

concentrates ammonia and organic compounds that are not readily degradable. When the dewatering liquid stream is returned to the mainstream for treatment, this increased load will increase the oxygen transfer rate requirements.

For some plants, it may be more cost-effective to implement a separate side-stream treatment process for loads resulting from anaerobic digestion and dewatering. The side-streams contain higher concentrations of ammonia, TSS, particulate BOD, alkalinity and phosphorus than typical wastewater. For every kilogram of ammonia removed in the side stream, one less kilogram of ammonia enters the main treatment process. Digester supernatant and centrate from sludge dewatering is unsuitable for conventional nitrification/denitrification due to its low organic carbon content compared to the nitrogen mass loading.

Three main options exist to address the impacts of side-stream loads on the main treatment process: (1) Manage the process through side-stream equalization; (2) Export the load to another treatment facility (e.g. sludge water treatment) for further processing; (3) Treat the side-stream load in-plant.

The main focus of this project is the investigation of the impact of the third option, side-stream treatment, on operational costs. It is recommended that side-stream treatment be considered if the side-stream nitrogen load is greater than 15% of the influent nitrogen load (Water Environment Federation and the American Society of Civil Engineers/Environmental and Water Resources Institute , 2010).

A possible advantage of side-stream treatment over treatment in the mainstream is the nitrification efficiency related to the elevated liquid temperature of centrate. At 30°C the rate of nitrification is maximized but above 36-38°C, the rate of nitrification is inhibited.

The main drivers for side-stream treatment include:

- 1. <u>Effluent Nitrogen Removal</u>: Due to the increased cost of aeration for nitrification, a wastewater plant may consider efficiencies resulting from side-stream treatment. Additionally, variable slug loads of nitrogen from intermittent discharges may result in exceedences if discharge limits are stringent, especially at plants with shift dewatering schedules. Therefore a separate system to treat variable slug loads may be operationally preferred.
- 2. <u>Energy Conservation</u>: Side-stream treatment processes may require less energy to operate compared to treating the side-stream load in the mainstream treatment process. For example, by limiting NOB growth, the ammonia is converted to nitrite instead of all the way to nitrate, requiring less oxygen and resulting in reduced blower air demand. This is discussed in further detail in Section 5.5.
- 3. <u>Reduction in Chemical Demand:</u>
 - <u>Alkalinity:</u> Side-stream processes designed to encourage growth of particular microorganisms can reduce chemical demand. For example, AOB selection over NOB

using temperature and pH control requires less alkalinity than if the NOB were allowed to complete nitrification to nitrate.

- <u>Exogenic Carbon</u>: Anammox bacteria can denitrify autotrophically, reducing the exogenous carbon required.
- 4. <u>Reduction in Facility Greenhouse Gas Emissions:</u> The addition of side-stream treatment can lower the overall greenhouse gas emissions for the facility by improving the efficiency of nitrogen oxidation and reduction reactions;
- 5. <u>Reduction in Facility Footprint:</u> By adding a side-stream process to remove nitrogen, the activated sludge reactors of the main liquid stream can be designed to treat a smaller nitrogen load, ultimately resulting in a smaller aerobic reactor volume required for treatment.

As the side-stream nitrogen load may account for 10-20% of the total plant flow, it is a consideration in the design of both new plants and retrofits of existing facilities. Other cost considerations for new construction or retrofits include the investment costs for capital works and equipment such as reactor vessels, heat exchangers, blowers, instrumentation, and controls.

2.6.3 Applications for Anammox

To date, Anammox has seen limited full-scale applications -- but the number of Anammox reactors is starting to grow as the process is better understood. The Anammox process is applicable to a wide variety of high-ammonia wastewater such as generated by agriculture, livestock wastes, semiconductor factory (Tokutomi, 2011), and landfill leachate (Valencia, Zon, Woelders, Lubberding, & Gijzen, 2011).

For example, the Anammox process could be applied for landfill bioreactors to treat the ammonia and nitrate. Modern landfills are designed as bioreactors to enhance their operation. Anammox has been identified in the solid matrix of landfills that could convert ammonium into nitrogen to reduce the nitrogen compounds in the leachate. Efforts to remove the ammonia that accumulates in the leachate of landfills have been focused on ex situ and partial in situ methods such as nitrification, denitrification and chemical precipitation (Valencia, Zon, Woelders, Lubberding, & Gijzen, 2011).

In wastewater treatment, the Anammox process is well suited to high-N side-streams such as centrate and anaerobic digester supernatant. The implementation of Anammox has been limited to the treatment side-stream processes, but in the future the mainstream Anammox process could become a reality (see Section 7.3 Mainstream Redesign).

3 Methodology

3.1 Literature Review

The literature review was conducted in two parts to establish the current state of nitrogen removal technology, with emphasis on the Anammox process for side-stream treatment. In Section 4, Results & Discussion, the microbiology of nitrogen converters was covered, with emphasis on the discovery of Anammox in the context of marine ecology and wastewater treatment. In Section 5, Wastewater Treatment Unit Processes, a discussion of novel treatment technologies for biological Total Nitrogen removal from wastewater with activated sludge treatment is presented. Recent advances in engineered bioreactor systems to make use of microbial metabolic pathways are emphasized.

3.2 Conceptual Designs

The conceptual designs were conducted in two parts, reactor sizing and total plant simulation. Reductions to the overall operational costs are demonstrated through preliminary reactor sizing calculations and by use of modeling software to simulate and compare different treatment scenarios.

- In the first part, preliminary reactor designs were developed for the removal of BOD, ammonia, and nitrate for the mainstream and side-stream. Simplified kinetic models were used determine the reactor volumes to sustain the necessary biomass. Sample calculations are provided to develop a rationale for reactor sizing and overall proof-of-concept for (1) the mainstream reactors for removal BOD, ammonia, and nitrates in order to establish the baseline cost for nitrogen removal; and (2) side-stream reactors for enhanced removal of nitrogen comparison of novel treatment schemes discussed in Section 4 & 5.
- In the second part, the impact of different treatment scenarios was demonstrated using a software simulator. Since wastewater treatment plants simultaneously process liquid, solid, and gas streams, a process change may impact other treatment processes. The effect of a variable on the operating costs, removal efficiency, and greenhouse gas emissions of the WWTP was evaluated. Total plant simulations were developed to compare various treatment scenarios, with emphasis on the addition of Anammox side-stream treatment using the WERF's (Water Environment Research Foundation) web-based software package called CHEApet⁴.

3.3 Objectives

The central goal of this project is to demonstrate operational savings resulting from side-stream treatment with the Anammox process. To evaluate the treatment effectiveness, the Total Nitrogen removal processes were measured against the following objectives:

⁴ http://cheapet.werf.org

- 1. <u>Total Nitrogen Removal</u>: The Anammox side-stream treatment reactor will enable the plant to meet effluent concentrations of 10 mg/L as NH_3 and 13 mg/L as NO_3 or 23 mg/L as Total Nitrogen (match the federal guidelines);
- 2. <u>Operating Cost</u>: The Anammox side-stream treatment reactor will decrease operating cost by reducing aeration and chemical requirements (\$/year and \$/ML); and
- 3. <u>Facility Greenhouse Gas Emissions</u>: The Anammox side-stream treatment reactor will decrease the greenhouse gas emissions by enhancing nitrogen removal efficiency (tonnes CO₂/year)

Other considerations such as reactor surface area are noted, where applicable.

4 Results & Discussion – Nitrogen Converters

If the complexity of the nitrogen cycle in the biosphere is any indication of its overall importance to life on the planet, the nitrogen converters – nitrifiers, denitrifiers, and Anammox -- had a significant impact on the planetary ecosystem over a geological time scale, resulting in the web of nutrient cycles in terrestrial, aquatic and atmospheric that comprises the present global biosphere.

Early in the history of the planet, the first cells must have led a precarious existence. Each new crisis may have wiped out large portions of the first cells on the planet. The most important task may have been to develop a variety of metabolic pathways to extract food and energy from the environment. One of the first pathways may have been fermentation. Some of these fermenters developed the ability to absorb or "fix" nitrogen from the air and convert it into the building blocks such as proteins for living systems. The subsequent emergence of photosynthesizers contributed to the spread of phytoplankton throughout the world's oceans. By the end of the first billion years after the emergence of life, the planet was teeming with bacteria, and these early living systems played a vital role in shaping the planet's climate and atmosphere through feedback loops that were established as stable nutrient cycles (Capra, 1996).

Our understanding of the roles of microbes in global nutrient cycles is extensive but it is not complete, as the recent discovery of a whole new nitrogen biochemistry through Anammox illustrates. The planctomycetes, previously thought to be evolutionarily unimportant, were found to be responsible for the Anammox process, and played a much larger role in the regulation of global nutrient and climate cycles than previously suspected.

An overview of the nitrogen converters is presented in this section, with a review of terminology, microbial metabolism, and a history of the discovery of the Anammox process in marine biology and in wastewater treatment. Further results and discussion are presented with a literature review of emerging ammonia- and Total nitrogen removal technologies in Section 5 - Wastewater Treatment Unit Processes. Sample calculations to demonstrate operational savings are provided in Section 6 - Plant Design Concepts for Side-stream Treatment Using Anammox.

The classical nitrogen cycle as depicted in Figure 2 shows the metamorphoses of elemental nitrogen through the biosphere. Nitrogen-fixers such as plants and algae convert atmospheric nitrogen into biomass, or as molecules such as proteins. At the completion of the cell's lifecycle, organic nitrogen decomposes into ammonia and/or ammonium ion. Nitrification by organisms such as *Nitrosomonas Europaea* and *Nitrobacter* oxidize the ammonia into nitrate. Denitrifying organisms such as *Pseudomonas* are responsible for the conversion of nitrates into nitrite, nitrous oxide, and nitrogen.

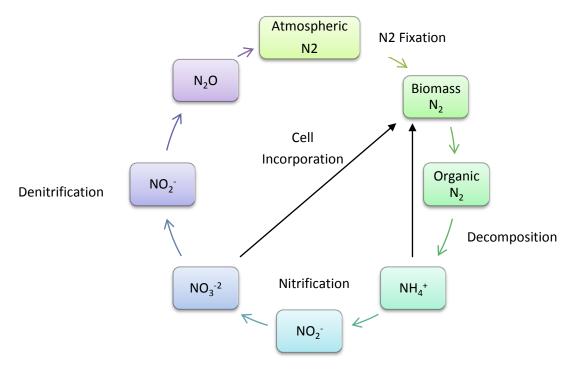


Figure 2: The classical Nitrogen Cycle

4.1 History of the Nitrogen Cycle

The nitrogen cycle and the denitrification reaction has been studied for over 130 years and its intermediates and enzymes by now are well known (Kartal, et al., 2011). The roles of nitrogen fixation, nitrification, and denitrification in the global nitrogen cycle were established by the end of 1890s. Investigations into the inhibition of dissolved oxygen on denitrifying bacteria were reported as early as 1882 by Gayon and Dupetit (Bronk, Mulholland, & Carpenter, 2008). Throughout most of the twentieth century, the general consensus among microbiologists was that ammonium was inert under anoxic conditions and could only be oxidized by aerobic processes. However, by the late 1970s, the picture was no longer so clear-cut. Specialists in the fields of theoretical chemistry and marine biology had independently proposed the existence of ammonia oxidation occurred under anaerobic conditions. The

Anammox process was actually demonstrated experimentally in the late 1980s at Gist-Brocades, Delft, The Netherlands. It is now clear that the Anammox process is a major player in global nitrogen cycling.

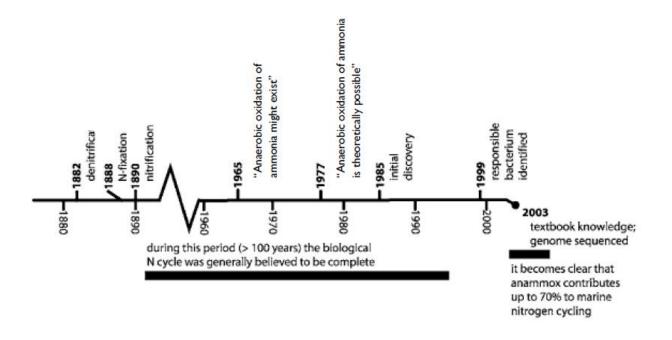


Figure 3: Timelines of discoveries in the field of ammonium oxidation⁵

The discovery of anaerobic methane oxidation (AnMO⁶) followed a parallel course to that of anaerobic ammonia oxidizing bacteria (AnAOB). Studies published in the late 1970s showed that the methane concentrations in marine sediment decreased toward the water column, and that sulfate concentration decreased in the opposite direction, from the water column into the marine sediment, indicating that sulfate might be the electron acceptor for anaerobic methane oxidation. At present, it is clear that anaerobic methane oxidation is responsible for more than 75% of marine methane oxidation (Strous & Jetten, Anaerobic Oxidation of Methane and Ammonium, 2004).

It is interesting to note that in the history of both anaerobic methane oxidation and the Anammox process is that although these two processes are quantitatively important to global nutrient cycles, they remained largely unnoticed for such a long time. Not only were they unnoticed, their discovery was met with strong skepticism by the scientific community. A discussion of further similarities and differences between the anaerobic processes of methane and ammonium oxidation is out of the scope of this project. Investigation of the co-relation of anaerobic microbes in the marine and wastewater treatment environment is recommended for further study.

⁵ Adapted from (Strous & Jetten, Anaerobic Oxidation of Methane and Ammonium, 2004)

⁶ Note that in the literature the acronym AMO is sometimes used to denote anaerobic methane oxidation. Another usage for this acronym is that used for the enzyme AMO (aerobic). In order to avoid confusion, AnMO is used to distinguish the anaerobic methane oxidation process from the aerobic nitrification enzyme AMO.

Since the marine biosphere is strongly coupled to global climate, Anammox may have been an important factor in development of the Earth's ecosystem over the geological time scale. As a major sink for fixed nitrogen, the Anammox process regulates the primary productivity of oceans by limiting the mass of nitrogen compounds available for biomass growth. It is now widely recognized that the Anammox process is responsible for up to 50% of global nitrogen production in the oceans.

Many natural ecosystems such as soils or oceans contain complex communities and it is easy to miss unknown species of microflora such as a specific bacterium responsible for regulating a microbial process. Nonetheless, the identification of the bacteria mediating the Anammox process in 1999 by Strous, et. al, came as a great surprise for the scientific community. Strous reports:

It would be surprising if a planctomycete was responsible for Anammox...Formerly the planctomycetes were considered to be of limited environmental importance. But this view is changing rapidly as molecular microbiology repeatedly provides new evidence showing that these bacteria are ubiquitous and make up a substantial portion of the natural bacterial population. The recognition of the planctomycetes as a major bacterial division may change our notion of what bacteria are (Strous, et al., 1999).

A brief history of the discovery of Anammox in the setting of marine biology, and in the wastewater treatment industry is presented in the sections that follow.

4.1.1 Marine Biology

In 1934, the American oceanographer Alfred Redfield published his observation that the elemental composition of plankton was very similar to the ratio of major dissolved nutrients in the deep ocean. He proposed that the nitrate to phosphate ($NO_3:PO_4$) ratio of 16:1 in the sea was controlled by the requirements of the phytoplankton, which release nitrogen (N) and phosphorus (P) to the environment as this ratio as the dead cells are broken down (e.g. remineralized from organic to inorganic form). This ratio was extended to include carbon (C) C:N:P ratio of 106:16:1 (Arrigo, 2005).

The marine biologist Richards (1963) noted that most of the ammonia that should be produced during the anaerobic remineralization of organic matter was unaccounted for. He proposed that the missing ammonia was anaerobically oxidized to nitrogen by some unknown microbe using nitrate as an oxidant (Arrigo, 2005).

More than a decade later, the Austrian theoretical chemist Eugene Broda predicted (1977) the existence anoxic ammonium oxidation with nitrite or nitrate instead of oxygen as the electron acceptor, was postulated on the basis of thermodynamic considerations (Hippen, Helmer, Kunst, Rosenwinkel, & Seyfried, 2001).

Broda reasoned that if a chemical reaction yields energy, it is likely that there is a chemotrophic bacterium that exploits that niche. The anaerobic ammonium oxidation reaction can yield enough

energy for a chemoautolithotrophic organism to live and the responsible bacterium was dubbed the "lithotroph missing from nature" (Van der Star, et al., 2007).

The discovery of Anammox activity in the marine environment occurred, again more than 10 years later. Anammox activity in the ocean was first investigated in anoxic sediment of different continental shelf regions using ¹⁵N-labeled ammonia. Soon after, evidence of anaerobic ammonia oxidation was found in the suboxic waters of Costa Rica. It was suggested that Anammox may be even more important where the upwelling of anoxic nitrate-rich waters contact sediments that produce significant amounts of ammonia, such as Peru and Chile. Conclusive evidence that the same planctomycetes were responsible for the Total Nitrogen removal in sediments and anoxic waters was achieved during a study of the Black Sea (Arrigo, 2005). A summary of these developments is presented in Table 2.

Table 2: Selected timeline of developments of the nitrogen cycle⁷

1882	Gayon and Dupetit report on the inhibition of denitrifying bacteria by dissolved oxygen
1890	The biological nitrogen cycle including denitrification, N-fixation, and nitrification is considered complete
1934	Redfield Ratio first described the ratios of nutrients (CON) in the marine plankton
1963	Richards proposed that "missing" ammonia was anaerobically oxidized by an unknown microorganism
1972	Cline and Richards propose a reaction mechanism of the anaerobic oxidation of ammonia
1977	Broda predicts anoxic ammonium oxidation based on thermodynamics, the "lithotroph missing from nature"
1989	Anaerobic ammonium activity noted in the water column in the Black Sea
1999	Strous identified microbes responsible for the reaction as members of the bacterial order Planctomycetales
2002	Discovery of Anammox reaction in suboxic waters in Costa Rica
2003	Identification of Anammox in the suboxic and anoxic zones of the Black Sea attributed to Planctomycetales

The 'Redfield ratio' is a fundamental tenet that has shaped the modern understanding of marine ecology, biogeochemistry, and phytoplankton evolution. The ratio links three major biogeochemical cycles (carbon, nitrogen, and phosphorus) through the activities of marine phytoplankton. However, conditions exist under which phytoplankton stoichiometry diverges from the canonical Redfield ratio. Why the Redfield ratio is observed so universally in the deep ocean is not clear but it is important to gain an understanding of microbial C:N:P stoichiometry because of the relevant role biological processes play in the elemental nutrient cycles. These cycles are coupled to processes operating from scales that range from algal photosynthesis to the global climate.

From the human perspective it is essential to understand the variation from the Redfield ratio resulting from increasing anthropogenic inputs of limiting nutrients. Liebig's law of the minimum, which states that only a single resource limits plant growth at any given time, was for many decades the theory that shaped how oceanographers viewed phytoplankton ecology. During the past few decades this view has been superseded by the realization that in some parts of the world's oceans, multiple resources

⁷ Adapted from (Arrigo, 2005), (Strous, Kuenen, Fuerst, Wagner, & Jetten, 2002), (Bronk, Mulholland, & Carpenter, 2008)

simultaneously limit phytoplankton growth, also known as resource co-limitation. The balance between the nutrient inventories and stoichiometric requirements of cell growth controls fundamental aspects of microbial ecology in the ocean and biogeochemistry, such as where and how much N₂ fixation, denitrification and Anammox take place. (Arrigo, 2005).

The Anammox process has been found to occur in a wide variety of $low-O_2$ marine environments. Recent studies have reported Anammox activity in diverse marine ecosystems, such as polar sea ice, oceanic anoxic basins, marine sponges, freshwater ecosystems, river sediment, estuaries (Gao & Y., 2011).

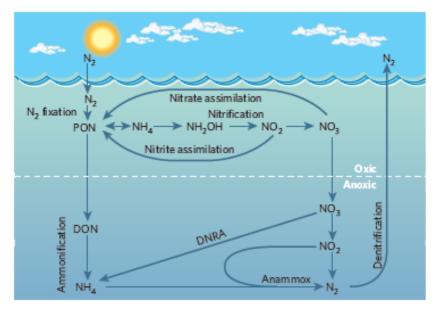


Figure 4: The marine nitrogen cycle including losses of ammonia and nitrite to N2 owing to Anammox⁸

4.1.2 Discovery in Wastewater Treatment

Although there was evidence of the Anammox reaction in the field of marine biology, the relevance of the process to engineer biological systems for wastewater treatment was not immediately evident. In 1989, the Anammox process was first noticed in a wastewater process at Gist-Brocades, Delft, The Netherlands. The Anammox process was discovered "by accident" in a wastewater treatment context in several locations at more-or-less the same time in the late 1990s when it was noted that nitrate was unexpectedly removed simultaneously with ammonia. In 1998, the first full-scale demonstration was conducted in Delft, The Netherlands. In the years following, the unexplained N-losses that were previously noted in nitrifying systems in Switzerland, Germany, and Belgium were attributed to aerobic ammonium oxidation, followed by anoxic ammonium oxidation by Anammox organisms in the deeper layer of the biofilm (Van der Star, et al., 2007). Since that time, the underlying mechanisms for the Anammox process have been explored at lab scale and a number of full-scale installations at wastewater

⁸ Adapted from (Arrigo, 2005), where PON particulate organic nitrogen, including phytoplankton; DON, dissolved organic nitrogen; DNRA, dissimilatory nitrtate reductase to ammonium

treatment plants have been started up in places such as the Netherlands, Austria, Turkey, Sweden, and in the US, as shown in Table 7.

Table 3: Selected Timeline of Anammox Process Development⁹

1989	The simultaneous removal of nitrite and ammonia is reported in an anoxic reactor in Delft, NL
1996	The organism responsible is cultivated in Delft, NL, for the oxidation of ammonium with nitrite as electron acceptor
	and was dubbed the "Anammox" process
1997	The high removal of ammonium from nitrifying RBCs in Mechernich (DE) in the absence of COD is noted, and named
	the apparent aerobic oxidation of ammonium the "aerobic Deammonification" process
1998	High N-losses in Kollikon (CH) are reported and attributed to nitrifiers in the aerobic and anoxic layers in the biofilm.
	Similarly, lab-scale nitrifying rotating disk contactors in Gent (BE) reported nitrogen losses and was dubbed the
	Oxygen Limited Autotrophic Nitrification-Denitrification (OLAND) process.
1999	The Anammox process is linked to the activity of Planctomycetales
2000	Full-scale installations start up in the Netherlands (Rotterdam, Sluisjesdihk, Lichtenvoorde)

4.2 Overview of the Nitrogen Cycle

4.2.1 Terminology

Since the identification of Anammox bacteria in the 1990s, a confusion in terminology has resulted from applications of the process in various configurations, such as two-stage suspended sludge reactors, one-stage sequencing batch biofilm reactors. A standard terminology has been adopted in recent years to include terms to further differentiate specific steps of the nitrogen cycle:

- <u>Nitrification</u>: nitritation plus nitratation
- <u>Nitritation</u> (also, Nitrosification, Ammonia Oxidation, Partial Nitrification): the autotrophic conversion of ammonia to nitrite by ammonia oxidizing bacteria (AOBs)
- <u>Nitratation</u> (also Nitrite Oxidation): the autotrophic conversion of nitrite to nitrate by nitrite oxidizing bacteria (NOBs)
- <u>Denitrification</u>: denitritation plus denitratation
- <u>Denitritation</u>: heterotrophic conversion of nitrite to nitrogen; requires an organic substrate.
- <u>Denitratation</u>: heterotrophic conversion of nitrate to nitrite; requires an organic substrate
- <u>Anaerobic Ammonia Oxidation</u> (ANAMMOX): autotrophic conversion of ammonia to nitrogen, using nitrite as the electron acceptor.
- <u>Deammonification</u>: partial nitritation followed by Anammox reaction

4.2.2 Microbial Metabolism

Activated sludge is a consortium of mixed types of microorganisms in an aerobic or anoxic environment in which different species compete or cooperate with each other. The dominant components of

⁹ Adapted from (Stensel, 2006), (Van der Star, et al., 2007)

activated sludge are carbon degrading heterotrophs and nitrifiers. The microbial ecology is the web of relationships in a community of microorganisms in terms of which cooperate and those which compete. Biotic communities are variously referred to as consortia, guilds, or biocoenosis.

Microbial metabolism is the means by which the microbe obtains energy and nutrients to live and reproduce. Catabolism is a set of metabolic pathways that break down molecules into smaller units and release energy. There are three principles used to categorize microbial metabolisms: the means by which microorganisms obtain (1) carbon for cell growth, (2) a reducing equivalent to transfer the equivalent of one electron in redox reactions, such as a lone electron, and (3) energy for living and growing:

- Autotrophs fix carbon dioxide into cell mass and organic compounds;
- Heterotrophs use organic carbon for growth as they are unable to fix carbon dioxide;
- Lithotrophs obtain reducing equivalents from inorganic compounds;
- Organotrophs obtain reducing equivalents from organic compounds;
- Phototrophs obtain energy from sunlight; and
- Chemotrophs obtain energy from external chemical compounds.

Autotrophs such as plants and algae use energy from sunlight (phototrophs) or inorganic compounds (lithotrophs) to fix carbon dioxide and produce organic compounds. The reduced carbon compounds can be used as an energy source by the autotroph or provide energy in the form of food consumed by heterotrophs. Ananmmox bacteria are chemolithoautotrophs which derive energy from reduced compounds of mineral origin ("lithos" (rock) and "troph" (consumer). The electron donor is oxidized within the cell, and the electrons are used to produce ATP (respiration).

Microbes are versatile and numerous distinct metabolic pathways may co-exist within a cell. There may be undetected microbes that can perform anaerobic ammonia oxidation, such as ammonia-oxidizing archaea and undetected strains of Anammox bacteria undetected (Waki, et al., 2009). For example, the role of ammonia-oxidizing archaea (AOA) has been increasingly recognized in the conversion of ammonia (Arrigo, 2005).

i Ammonia Equilibrium

There is an equilibrium between ammonia and ammonium ion in water. It is the free ammonia that is toxic to aquatic life. The dissociation of ammonia to ammonium ion is impacted by pH and temperature. At pH 7.5, 15 °C, about 1% of Total Ammonia Nitrogen (TAN) is NH₃-N (Canadian Council of Ministers of the Environment, 2010).

Equation 1: Speciation of NH_3 to NH_4^+

 $NH_3 + H^+ \leftrightarrow NH_4^+$; pKa = 9.24

ii Nitrification

Nitrification is a two-step reaction for the conversion of ammonia to nitrite and then nitrite to nitrate. The first step, nitritation (also known as nitrosifying or partial nitrification), is shown in Equation 2. Nitritation is the partial oxidation of NH_4^+ to NO_2^- via ammonia oxidizing bacteria (AOBs), activated by oxygen through the use of a catalyst enzyme – ammonium monooxygenase (AMO). Aerobic organisms that are AOBs are typified by the genus *Nitrosomonas*, a common environmental bacterium found in aquatic and terrestrial ecosystems. Ammonia-oxidizing archaea (AOA) have also been reported to be active in the conversion of ammonia.

Equation 2: Nitritation of ammonia to nitrite by ammonia oxidizing bacteria (AOB)

$$NH_4^+ + \frac{3}{2}O_2 \xrightarrow{\text{Nitrosomonas}} NO_2^- + H_2O + 2H^+$$

Nitrosomonas europaea can derive all its energy from AMO catalyzed ammonia oxidation and fixes its carbon from carbon dioxide. Ammonium ion is oxidized by the bacteria to nitrite by AMO to hydroxylamine, followed by a series of reactions which provide the energy for the AMO oxidation reaction. Nitritation consumes $3.22 \text{ g} \text{ O}_2$ (3.43 g not including cell synthesis) for each gram of ammonia to be oxidized to nitrate. This accounts for 75% of the total oxygen consumption for complete nitrification.

Nitratation, shown in Equation 3, involves the partial oxidation of NO_2^- to NO_3^- by nitrite oxidizing bacteria (NOBs), catalyzed by the enzyme *nitrite oxidoreductase*. Aerobic organisms that are NOBs are *Nitrobacter*, etc. Nitratation consumes 1.11 g O_2 (1.14 g not including cell synthesis) for each gram of nitrite to be oxidized to nitrate. This is the remaining 25% of the total oxygen consumption for complete nitrification.

Equation 3: Nitratation of nitrite to nitrate by nitrite oxidizing bacteria (NOB)

$$NO_2^- + \frac{1}{2}O_2 \xrightarrow{\text{Nitrobacter}} NO_3^-$$

The complete conversion of ammonium ion to nitrate ion is given by:

Equation 4: Overall nitrification reaction by AOBs and NOBs

$$NH_4^+ + 2O_2 \rightarrow NO_3^- + H_2O + 2H^+$$

A few key points to consider for nitrifiers:

- Nitrifiers are strict aerobes, and are inhibited by dissolved oxygen concentrations as low as 0.5 mg/L;
- Nitrification consumes 4.57 g O₂ for each gram of ammonia to be oxidized to nitrate. Accounting for cell synthesis, this amount is somewhat less, 4.33 g O₂ for each gram of ammonia oxidized;

- Nitrifiers use alkalinity present in the wastewater as a source of C for growth (i.e. CaCO₃), or 7.14 g alkalinity consumed per g NO₃;
- Aerobic nitrifiers are autotrophic and use either carbon dioxide (CO₂) or bicarbonate (HCO₃-) as their sole source of carbon for cell growth; and
- Nitrification reactions are exothermic, with 80 kJ liberated for every mole of ammonia converted and 27 kJ for every mole of nitrite (350 kJ/mol total).

iii Denitrification

Denitrification is a multi-step reaction that converts nitrate to nitrite then to intermediates of nitrous oxide and nitric oxide before the reaction completes with the production of nitrogen. Denitrification requires a denitrifier, a steady source of nitrates, a well-mixed anoxic zone, and sufficient substrate for carbon source/electron donor. Oxidation-reduction half reactions are presented for nitrate in Equation 5, and nitrite in Equation 6.

Equation 5: Denitrification of nitrate to nitrogen

0.20 NO₃⁻ + e⁻ + 1.2 H⁺
$$\xrightarrow{\text{Pseudomonas}}$$
 intermediates NO and N₂O \rightarrow 0.1N₂ + 0.6 H₂O

Equation 6: Denitritation of nitrite to nitrogen

0.33 NO₂⁻ + e⁻ + 1.33 H⁺
$$\xrightarrow{\text{Pseudomonas}}$$
 intermediates NO and N₂O \rightarrow 0.17N₂ + 0.67 H₂O

The terminal electron acceptor during denitrification is nitrate, rather than oxygen. Nitrate acts as oxygen source for species of denitrifying bacteria, such as *Pseudomonas*. The presence of dissolved oxygen inhibits denitrification because it will be used before nitrate if it is present as it is an electrochemically more favourable electron acceptor.

Nitrogen removal occurs through the reduction of nitrate via one of two routes:

- 1. <u>Assimilating Nitrate Reduction</u>: the nitrate is reduced to ammonia through for use in cell synthesis.
- 2. <u>Dissimilating Nitrate Reduction</u>: in the respiratory electron transport chain, biological denitrification use nitrate or nitrite as an electron acceptor (instead of oxygen) for the oxidation of organic electron donors (i.e. rbCOD).

Typically, the electron donor is (1) the influent bsCOD; (2) the bsCOD produced during endogenous decay; or (3) an exogenous source, such as methanol or acetate. When there is not enough organic carbon to drive the nitrate reduction reaction, the endogenous respiration (assimilating nitrate reduction) occurs at a rate that is 3-8 times slower (Metcalf & Eddy, 2004).

- Nitrification/Denitrification consumes 2.86 mg O₂ for each gram of ammonia to be oxidized to nitrogen as part of the organic substrate in the wastewater is oxidized using NO₃⁻, or 1.72 g COD/g NO₂-N by denitritation.
- Up to 50% of alkalinity lost through nitrification is recovered during denitrification, as one equivalent of alkalinity is produced per equivalent of NO₃-N, or 3.57 g of alkalinity (as CaCO3) produced per g of nitrate nitrogen reduced.
- An important design parameter is the amount of biodegradable soluble COD or BOD to provide sufficient electron donor used for denitrification. The amount of biodegradable soluble COD used per unit of nitrate nitrogen reduced is based on the system biomass yield. As a general rule about 4 g BOD is needed per g NO₃ reduced.
- Methanol, ethanol, acetate, or proprietary products may be added to for carbon-limited (low COD) wastewater

iv Anammox

Until relatively recently, the role of Anammox in the nitrogen cycle was largely unknown. Ammonia is a difficult compound to activate chemically and for many years it was thought that ammonia oxidation could only proceed by *monooxygenase* enzymes produced by strictly-aerobic microbes, so was not possible for anaerobic life (Strous & Jetten, Anaerobic Oxidation of Methane and Ammonium, 2004). However it is now thought that Anammox conversion of ammonium ion directly into nitrogen accounts for 50% of global nitrogen cycling. The Anammox reaction is expressed as:

 $\begin{array}{l} NH_4^+ + 1.32NO_2^- + 0.066HCO_3^- + 0.13H^+ \xrightarrow{\text{Anammox}} 1.02\text{N}_2 \\ + 2.03H_2 O \end{array} \\ \end{array} \\ \begin{array}{l} 0 \end{array} \\ \begin{array}{l} 1.02\text{N}_2 + 0.26NO_3^- + 0.066CH_2O_{0.5}N_{0.15} \\ \end{array} \\ \end{array} \\ \end{array}$

Equation 7: Anammox reaction

The role that Anammox plays in the global nitrogen cycle is depicted in Figure 5. The Anammox reaction was first discovered in an industrial setting at a denitrifying pilot plant reactor in Delft, the Netherlands.

- Anammox consumes ammonium and nitrite in a stoichiometric ratio of 1:1.3;
- The apparent activation energy of the Anammox reaction is 70 kJ/mol. Anammox activity depends on temperature, increasing two times for a temperature increase of 10°C (Strous & Jetten, Anaerobic Oxidation of Methane and Ammonium, 2004);
- Alkalinity consumption per mole of ammonium consumed is 0.066 equivalents or 0.24 g as \mbox{CaCO}_3

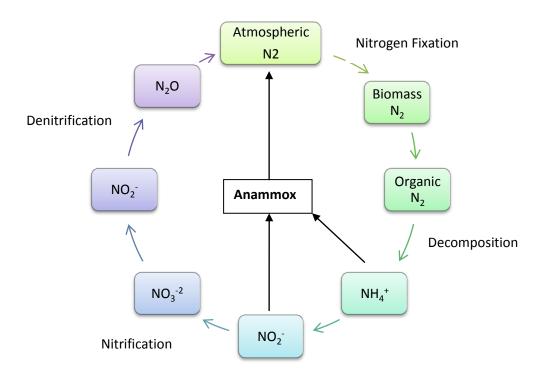


Figure 5: A diagram of the revised nitrogen cycle including the Anammox short-cut

4.3 Characteristics of Anammox Bacteria

This section covers the species of bacteria that are responsible for the Anammox process, and the unique physiology that allows them to metabolize ammonia and nitrite under anaerobic conditions.

4.3.1 Species

Although the existence of the Anammox process was speculated since the 1970s, it was many years before the organisms responsible for the process were identified in the 1990s. The bacteria that perform the Anammox process are of the bacterial phylum *Planctomycetales*, of which *Planctomycetes* and *Pirellula* are the best known genera. Five genera of Anammox bacteria have been defined (Meng & Gu, 2011).:

- Brocadia (fresh water species);
- Kuenenia (fresh water species);
- Anammoxoglobus(fresh water species);
- Jettenia (fresh water species); and
- Scalindua (marine species).

The niche differentiation of Anammox species is unresolved, but there are hypotheses, for example, that *Brocadia* cells are more sensitive to nitrite inhibition, and *Kuenenia* cells have higher affinity for

substrates (Tokutomi, Yamauchi, Nishimura, Yoda, & Abma, 2011) which has been supported by phylogenetic analysis of reactor sludge.

4.3.2 Physiology

Anammox bacteria are characterized by several unique properties, in relation to cell compartmentalization. The Anammox bacteria have a membrane-bound organelle called the Anammoxosome, where Anammox catabolism occurs. Hydrazine (N_2H_4) is produced as an intermediate within the Anammoxosome organelle to catalyze the Anammox reaction. The Anammoxosome membrane is made up of ladderane lipids, which is unique in biology. Ladderane is an organic molecule of serial cyclobutane rings that look like a ladder and are singly bonded like alkanes, as shown in Figure 6(a). Ladderane forms a very tight and dense membrane which prevents the organism from losing the intermediate hydrazine. This is important because hydrazine is poisonous to most organisms – and can be used as high-energy rocket fuel.

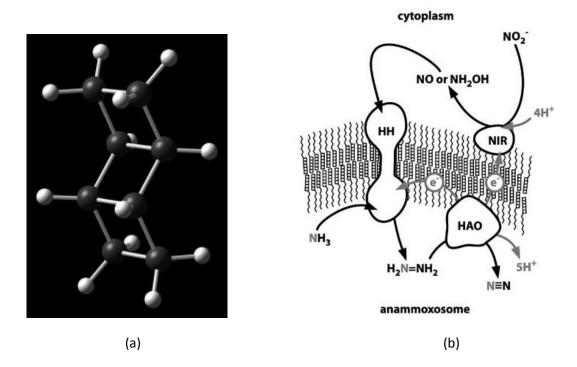


Figure 6: (a) Ladderane molecule [Wikipedia]; (b) possible biochemical pathway of Anammox¹⁰

The Anammox reaction is given in Equation 7. Besides the nitrate that is used for cellular catabolism, Anammox anaerobically oxidizes nitrite to nitrate through the Anammoxosome, as shown in Figure 6(b). The Anammox pathway was established using ¹⁵N-labeling experiments, which showed that hydrazine is

¹⁰ Adapted from (b) (Strous & Jetten, Anaerobic Oxidation of Methane and Ammonium, 2004), in which the proton motive force is general over the Anammoxosome membrane via separation of charges. Black and gray N-atoms highlight the production of $^{29}N_2$ from one of the ^{15}N -labeled substrates, ammonium or nitrite.

an important intermediate. Anammox bacteria were shown to fix CO₂ while anaerobically oxidizing ammonium (Strous & Jetten, Anaerobic Oxidation of Methane and Ammonium, 2004).

A necessary precondition for the anaerobic ammonium oxidizers is a nearby source of nitrogen compounds; ideally, ammonia and nitrate are provided by pretreatment in the correct stoichiometric proportion. That Anammox bacteria have been found in diverse biocoensis across the planet demonstrates their versatility to adapt to widely different environments.

Anammox bacteria have been found in a range of extreme environments. The presence of guilds of Anammox have been identified, with temperatures ranging from 20-43°C to from 36-52°C in hot springs, and 60-85°C at hydrothermal vents located along the Mid-Atlantic Ridge. At the other temperature extreme, Anammox bacteria can also survive at low temperatures, having been found in sea ice as low as $-2^{\circ}C$ (Gao & Y., 2011).

This versatility of Anammox may continue to encourage engineers to find application for the Anammox process under extreme temperatures, reducing the costs of a cooling or heating stage. For example, Anammox acclimated to ambient temperature can be used to treat low-temperature wastewaters (Yang, Zhang, Hira, Fukuzaki, & Furukawa, 2011).

4.3.3 Anammox Detection Methods

Part of the reason for the reticence of the scientific community to accept the new discovery of the Anammox process was the failure to demonstrate the responsible microorganism in a lab setting. In microbiology, Koch's postulate states that to prove that a process is mediated by a bacterium, this bacterium should be isolated in pure culture and still be able to reproduce the process. Anammox bacteria are strictly anaerobes, and difficult to cultivate in a lab. Conventional purification using single-cell isolation met with repeated failure (Strous, et al., 1999). Clearly, a different approach was needed to unambiguously identify the "lithotrophs missing from nature."

The method followed by Strous, et. al to identify the bacteria responsible for the Anammox process was based on a modernization of the Winogradsky/Beyerinck strategy of selective enrichment (1949) with the modern molecular toolbox for detection and bioreactor engineering, as follows:

- (1) postulate an ecological niche based on thermodynamic considerations and ecological field data;
- (2) engineering of this niche into a lab-scale bioreactor;
- (3) black-box physiological characterization of the enriched community in accordance with Koch's postulates; and
- (4) verification of the importance of the detected species in actual ecosystems

Currently, several methods are used to identify Anammox bacteria based on their physiological and biochemical properties, cellular composition, and gene biomarkers (Meng & Gu, 2011):

- Isotope-labeling techniques: The Isotope Pairing Technique (IPT) is a well-establish method for monitoring the ¹⁵N isotope using a series of incubations to detect the contributions of denitrification and Anammox to N₂ production. Another method is Stable Isotope Probing (SIP), a technique with ¹³CO₂ and or ¹⁵N-labeled inorganic nitrogen species to allow the selective recovery analysis of isotope-enriched cellular biomolecules such as DNA, RNA, proteins, and phosphorus lipid fatty acids, and other components of the microbial nitrogen cycle such as ammonia-oxidizing archaea and bacteria in soils.
- <u>Lipid measurement</u>: Unique ladderane lipids in the cellular membrane surrounding the Anammoxosome contain impermeable cyclobutane/cyclohexane molecules that can be used as indicators or biomarkers for Anammox bacteria. The extraction procedures are complicated and may limit the confidence in precision for quantifying Anammox bacteria
- <u>FISH-based techniques</u>: Fluorescence In-Situ Hybridization (FISH) is used to investigate target cells using specific oligonucleotide probes for detection of Anammox bacteria, based on labeling of ¹⁴C to confirm the chemolithoautotrophic biochemical pathway carried out by Anammox bacteria. The FISH technique can be used to quantify the abundance of Anammox bacteria.
- <u>SEM Observation</u>: Scanning electron microscopy is used to observe the surface and inner parts of Anammox granules.
- <u>Molecular techniques:</u> Such techniques include DNA extraction and PCR amplification techniques involve sample DNA templates with specific biomarkers, specific primers, and subsequent phylogenetic analysis of the amplified DNA products, using the 16S rRNA gene, and functional genes based on the Anammox metabolism.

4.3.4 Obstacles to Implementation

Anammox bacteria are known to have a grow rate $1/10^{\text{th}}$ the rate of nitrifiers, with a doubling time of 11 days under optimum conditions (Strous, Kuenen, & Jetten, 1999). Low growth rates have restricted the application of Anammox to few examples of full-scale plants to date, but the number is growing. The yield of Anammox bacteria is 0.07 Cmol fixed/mol NH₄⁺ oxidized, which is approximately the same for AOBs and is consistent with the Gibbs free-energy change of Anammox catabolism. It is notable that the low growth rate is not caused by inefficient energy conservation but by a low substrate conversion rate (Strous & Jetten, Anaerobic Oxidation of Methane and Ammonium, 2004). In the presence of an organic carbon source, heterotrophic bacteria outcompete the Anammox bacteria. In practice, it is common to cover Anammox reactors to prevent the growth of autotrophic algae.

Other than their slow growth rate, the obstacles that have prevented widespread application of the Anammox process are related to the precise control required for:

- (1) Partial Nitritation (i.e. stable nitrite-to-ammonia ratio in effluent of Partial Nitritation process),
- (2) incomplete Total Nitrogen removal (based on the Anammox reaction stoichiometry, and

(3) sensitivity to inhibitors:

- <u>Dissolved Oxygen</u>: Anammox bacteria of the planctomycetes genera are strictly anaerobic. Reversible inhibitory effects for dissolved oxygen have been reported at <0.06 mg/L (Gao & Y., 2011), or < 1 μM (Strous & Jetten, Anaerobic Oxidation of Methane and Ammonium, 2004).
- <u>Methanol</u>: Methanol has been widely used as a hydrogen donor for heterotrophic denitrification. However, it has been shown that methanol is a notable inhibitor of the Anammox reaction, with reported ranges varying from total loss of Anammox activity at 0.5 mM of methanol to 51% activity at 3.3 mM. It would be useful to avoid methanol contamination in the operation of Anammox processes (Isaka, Suwa, Kimura, Yamagishi, Sumino, & Tsuneda, 2008).
- <u>Free Ammonia</u>: Free ammonia concentrations of 13-90 mg/L have been reported to negatively affect the performance of the Anammox process (Li, Ma, Hira, Fujii, & Furukawa, 2011).
- <u>Nitrate</u>: The Anammox reaction inherently produces nitrates that are left over when one mole each of ammonium and nitrite is eliminated in the Anammox process (see Equation 7). Nitrate must be treated through an additional process, such as a heterotrophic denitrification process. In one study, Anammox activity decreased by 37% when nitrate concentration exceeded 430 g-N/m³ (Yang, Zhang, Hira, Fukuzaki, & Furukawa, 2011).
- <u>Nitrite:</u> the presence of more than 10 mM or 0.1 g-N/L as NO₂-N or the Anammox process completely inhibited (Strous & Jetten, Anaerobic Oxidation of Methane and Ammonium, 2004). The nitrite inhibition can be overcome by addition of trace amounts of the Anammox catabolism intermediates (more than 1.4 mg N/L for hydrazine, and more than 0.7 mg N/L for hydroxylamine) (Strous, Kuenen, & Jetten, 1999)

5 Wastewater Treatment Unit Processes

The previous section, the fundamental mechanisms for biological nitrogen conversion was covered. This section covers:

- wastewater treatment technologies used for ammonia- and Total Nitrogen removal, including influencing parameters;
- process control strategies for biological nitrification, denitrification, nitritation, denitritation, and Anammox; and
- reactor configurations for side-stream treatment; and a summary of lessons learned from the literature about the start-up and stable operation of Anammox systems.

The activated sludge process has made use of microbial processes for the removal of carbonaceous BOD for more than a century. In the past decades, the activated sludge process has been modified for biological nutrient removal. In recent years, engineered systems for biological removal of nitrogen compounds have been widely implemented. The biological removal of nitrogen from wastewater can be

conveniently represented by the redox pyramid shown in Figure 7 which depicts the conversion steps of ammonium ion to nitrogen, and the necessary inputs to complete the process.

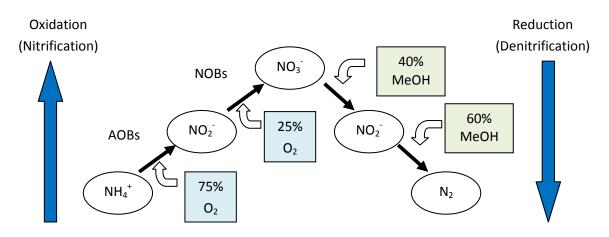


Figure 7: Nitrification – Denitrification Redox Pyramid

5.1 Wastewater Characteristics

An understanding of the characteristic parameters of the raw wastewater and the side-stream that impact the performance of biological Total Nitrogen removal is necessary to prepare an appropriate design and operate a nitrogen removal system, especially one so sensitive to disturbances as Anammox reactors. The activity of nitrifiers, denitrifiers, and Anammox bacteria may be inhibited by the composition of the wastewater, specific compounds, or parameters such as pH and temperature.

Nitrogen Compounds

The forms of nitrogen compound in wastewater include inorganic compounds like ammonia or nitrate, and organic nitrogen fractions that are biodegradable or non-biodegradable, with soluble and particulate portions. The Total Ammonia Nitrogen (TAN) is the sum of the free ammonia and the ammonium ion. The Total Kjeldahl Nitrogen (TKN) includes the sum of ammonia-nitrogen and organic-nitrogen. The Total Nitrogen (TN) is the sum of TKN and salts of nitrogen (e.g. nitrate and nitrite). The fractions of total nitrogen in wastewater are outlined in Figure 8.

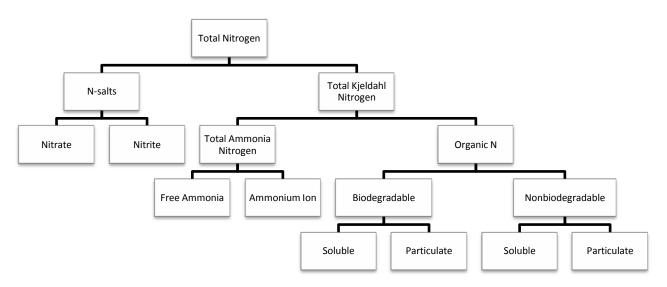


Figure 8: Fractionation of nitrogen in wastewater¹¹

For plants with high nitrogen loading, such as agricultural, livestock wastewater, industrial effluent, and centrate, the low ratio of COD to TN is the primary indicator that it will be difficult to treat by conventional nitrification/denitrification process (Hippen, Helmer, Kunst, Rosenwinkel, & Seyfried, 2001). In wastewater treatment, the raw sewage influent which may be under 50 mg/L TKN and the centrate can carry TKN concentrations of 1,000 mg/L, with comparatively low organic carbon.

Oxygen Demand

The reactor volume, or zone, is characterized by the dissolved oxygen concentration:

- Anoxic: dissolved oxygen concentration of less than 0.5 mg/L with salts of nitrogen such as nitrite and nitrite providing a source of oxygen for denitrifying organisms;
- Aerobic: dissolved oxygen concentration, typically greater than 0.5 mg/L; and
- Anaerobic: an environment that exists in the total absence of dissolved oxygen or oxides such as nitrate or nitrite.

The biochemical oxygen demand (BOD) is widely used to characterize wastewater in terms of the consumption of oxygen from the decay of organic matter (carbonaceous BOD, or cBOD), and nitrification of ammonia (nitrogenous BOD, or nBOD). Comparison of cBOD vs nBOD is in shown on Figure 9. Dissolved oxygen is essential to life in the aquatic environment. The difference between BOD and cBOD is assumed to be nBOD, which represents the portion of oxygen demand attributed to nitrogenous matter in the wastewater (Metcalf & Eddy, 2004)

¹¹ Adapted from (Metcalf & Eddy, 2004)

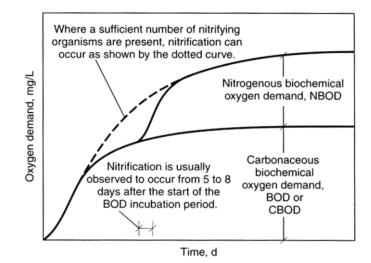


Figure 9: The carbonaceous and nitrogenous biochemical oxygen demand in a sample of wastewater¹²

Although BOD is a useful parameter to characterize the carbonaceous and nitrogenous matter in wastewater, COD is the parameter that is primarily used to determine the amount of carbonaceous material either oxidized or converted to biomass (see Figure 10). To establish the availability of substrate for biomass production, the characterization of wastewater for the design of activated sludge process include:

- the biodegradable (bCOD) and non-biodegradable (nbCOD) portions of chemical oxygen demand as organic carbon;
- the soluble readily biodegradable chemical oxygen demand (rbCOD);
- the non-biodegradable portion of volatile suspended solids (nbVSS)
- the effluent sCOD assumed to be non-biodegradable (nbsCOD); and
- the inert TSS (iTSS) entering the reactor.

Of these the rbCOD and nbVSS have the greatest impact on process design. For activate sludge processes, the rbCOD concentration can be used to evaluate oxygen demand. Also, the fraction of rbCOD affects the denitrification rate, which increases with the amount of rbCOD. The extent of fractionation of rbCOD is measurable in lab tests or estimated from experience or literature. The nbVSS affects sludge production and aeration reactor volume, and can be estimated from other constituents of the wastewater.

¹² Adapted from (Metcalf & Eddy, 2004)

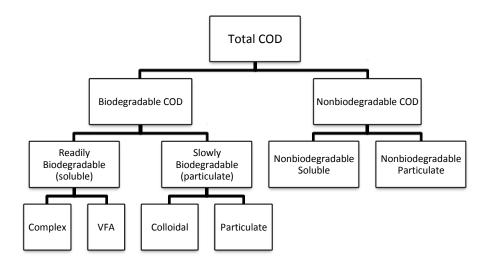


Figure 10: Fractionation of COD in wastewater¹³

Heterotrophic bacteria that consume organic carbon substrate have growth rates that are 5 times and yields of 2 to 3 times that of autotrophic nitrifying bacteria (Ozdemir, Mertoglu, Yapsakli, Aliyazicioglu, Saatci, & Yenigun, 2011), as shown in Table 10.

Other Parameters

In addition to readily measured parameters such as pH and temperature, there must be adequate alkalinity concentration available for the biological nitrification reaction to reach completion.

5.2 Biological Nitrification

The mechanisms for removal of ammonia by conversion to nitrite by AOBs and/or nitrate by NOBs have been described in Section 4.2.2. This section describes the application to wastewater to remove ammonia biologically. Ammonia and nitrite are relatively difficult to oxidize, and is made biologically possible by *monooxygenase* enzymes available only to aerobic organisms. Ammonia oxidizing bacteria utilize the *AMO*-mediated reaction to oxidize ammonia to nitrite, and subsequently NOBs use the enzyme *nitrite oxidoreductase* to oxidize nitrite to nitrate.

An Advanced Secondary Treatment (AST) is a modification of the activated sludge process to aims for complete conversion of ammonia to nitrate, otherwise known as full nitrification. The operational conditions to encourage nitrification in addition to BOD removal include:

- Nitrifiers are strict aerobes, and are inhibited at very low DO levels (e.g. approximately 0.2 mg/L).
- The optimal pH for nitrification is between 7 and 8.5, with nitrification inhibited below pH 6.
- Alkalinity is consumed as the carbon source for nitrifier growth, and it is good practice to ensure that there is at least 80 mg/L as HCO₃⁻ remaining to ensure pH neutrality. The total

¹³ Adapted from (Metcalf & Eddy, 2004)

alkalinity demand can be estimated from stoichiometric ratios presented in Equation 2 & Equation 3. Further, nitrifier activity is reduced by high concentrations of inhibitors such as free ammonia and nitrous acid.

Design considerations noted in the following sections include kinetics for nitrifier growth and decay, estimates of sludge residence and nitrate production, and aeration energy demand.

5.2.1 Nitrifier Kinetics and Sludge Retention

Nitrifying organisms grow more slowly than heterotrophic organisms that remove the organic carbon. The nitrification rate therefore governs the reactor design. The concept of minimum solids residence time (SRT) is crucial in the development of a nitrification reactor to ensure that nitrifying organisms are able to grow and are retained in the reactor biomass.

Nitrifiers are more sensitive to temperature than heterotrophic denitrifier bacteria. In colder climates, the reactor sizing is controlled by the coldest water temperature that will occur during the year. Typical design rates for SRT range from 10 to 20 days at 10°C to 4 to 7 days at 20°C. Below 28°C ammonia-oxidation (*Nitrosomonas*, AOB) kinetics are rate limiting compared to nitrite-oxidation (*Nitrobacter*, NOB). Above 28°C the kinetics of both AOBs and NOBs should be taken into consideration (Metcalf & Eddy, 2004).

The mean cell residence time or solids residence time, θc is affected by the mixed liquid temperature. A first approximation to SRT as a function of temperature (°C) only is given by Equation 8 (Reynolds & Richards):

Equation 8: The minimum mean cell residence time as a function of temperature

$$\theta_c = 2.13e^{0.098(15-T)}$$

A better estimate for solids residence time for a nitrification system based on kinetics is a modified form of Monod's expression, which includes nitrifier growth, endogenous decay and the effects of dissolved oxygen (Metcalf & Eddy, 2004), as given in Equation 9. Kinetic parameters include growth and decay rates for aerobic heterotrophs, AOBs, and NOBs:

Equation 9: Nitrification growth rate as Monod function

$$\mu_n = \mu_{n,max} \left(\frac{N}{K_n + N} \right) \left(\frac{O_D}{K_o + O_D} \right) - k_{dn}$$

Where:

 μ_n = specific growth rate of nitrifiers (g new cells/g cells-time)

- $\mu_{n, max}$ = the maximum specific growth rate for nitrifiers
- K_n = the half-saturation constant for nitrogen, or the substrate concentration when $\mu = \frac{1}{2} \mu_{max}$
- K_o = the half-saturation constant for oxygen (0.5 mg/L)

- *N* = target effluent nitrogen concentration (mg/L)
- *O*_D = target dissolved oxygen concentration (mg/L)
- k_{dn} = endogenous decay term for nitrifiers (g VSS/g VSS-d)
- $\mu_{n, max}$ $K_{n\nu}$ $k_{dn\nu}$ are corrected for temperature by $X_2 = X_1 \cdot \tau^{(T_2 T_1)}$ where T_1 is the reference temperature, T_2 is the temperature of the mixed liquor, and τ is the temperature correction coefficient

Although the use of this model will generally over-predict nitrification rates at high organic loadings, it provides a basis to estimate the specific growth rate for the nitrifiers so that minimum SRT can be determined in Equation 10. The specific growth rate of nitrifying bacteria is much lower than for aerobic heterotrophic bacteria at the same temperature (see Table 10), requiring much longer SRT values to achieve biomass growth for nitrification. Further, the growth rate for *Nitrobacter* is higher than the specific growth rate of *Nitrosomonas;* the growth rate of *Nitrosomonas* will control the overall conversion rate, and there will be no accumulation of nitrite in the reactor.

Equation 10: Minimum Solids residence time

$$\theta_c = \frac{\Phi}{\mu_n} \cong \frac{\Phi}{\mu_{max,n} - \mathbf{k}_{dn}}$$

Where:

 Φ = Safety Factor (assume = 2.5 unless otherwise noted);

When operating with a residence time greater than the minimum SRT, nitrification takes place, otherwise nitrification does not occur due to washout of the microbes. The US EPA recommends a safety factor of 2.5 be used for design such that ammonia breakthrough does not occur during peak loading periods (Reynolds & Richards). As seen in Figure 11: The effect of wastewater temperature on minimum SRT, in the optimal temperature range of 25 to 35°C, the minimum SRT predicted by both Equation 8 and Equation 9 is lower than 1.5 days. At the design temperature range of 10 to 14°C for a typical wastewater treatment plant, nitrifiers are washed out at solids residence times lower than 4 days.

The design volume for the activated sludge reactor to achieve nitrification is based on the rate of growth of nitrifiers, which is a function of the influent wastewater temperature. The design for the aerobic reactor SRT is therefore based on the coldest wastewater temperature that will be experienced.

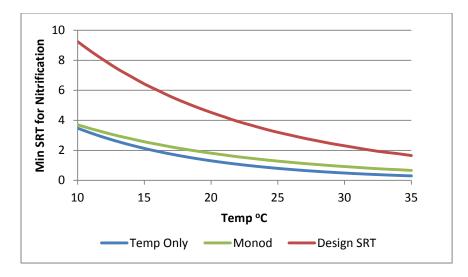


Figure 11: The effect of wastewater temperature on minimum SRT¹⁴

As the solids residence time increases the required oxygen transfer and consequently the blower power consumption will also increase towards a maximum, as in Figure 12. The optimal mixed liquor suspended solids concentration is between 2,500-3,000 mg/L in the aeration reactors to prevent excessive solids loading to the secondary clarifiers (Metcalf & Eddy, 2004).

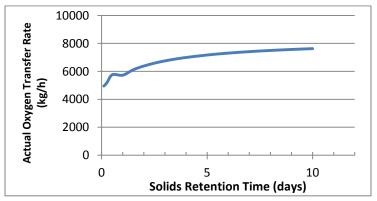


Figure 12: Aeration blower air required vs. Solids Retention Time¹⁵

5.2.2 Nitrate Production

The rate of nitrate production is needed to find the oxygenation requirements for the aerobic reactor. The rate of nitrate production is proportional to the activity of ammonia- and nitrite- oxidizing bacteria suspended in the mixed liquor of the aerobic reactor. The biomass production in the reactor is calculated using Equation 11, and comprises of four parts:

¹⁴ Based on Equation 8

¹⁵ Developed from the nitrification reactor presented in Section 6.4.2.

- (A) heterotrophic biomass,
- (B) cell debris from endogenous decay,
- (C) nitrifying bacteria biomass, and
- (D) non-biodegradable VSS in the influent.

If the SRT is greater than the minimum number of days needed to retain nitrifying organisms in the biomass, then the (C) term is included in the biomass calculation, otherwise it is assumed to be negligible.

Equation 11: Mixed Liquor Volatile Suspended Solids Concentration and Solids Production

$$P_{X,VSS} = \frac{QY(S_o - S)(\frac{1kg}{10^3g})}{1 + (k_d)\theta_c} + \frac{(f_d)k_dQY(S_o - S)\theta_c(\frac{1kg}{10^3g})}{1 + (k_d)\theta_c} + \frac{QY_n(N_{OX})(\frac{1kg}{10^3g})}{1 + (k_{dn})\theta_c} + Q(X_{nbVSS})\left(\frac{1kg}{10^3g}\right)$$

Where:

 $P_{X,VSS}$ = daily biomass production (kg VSS/d)

Q= flow rate (m³/d) N_{Ox} = nitrogen oxidized (mg/L) Y, Y_n = yield of heterotrophs and of nitrifiers (g/g-d) f_d = fraction of biomass that remains as cell debris X_{nbVSS} = nonbiodegradable VSS (mg/L)

The effluent soluble substrate concentration (sCOD) for a complete-mix activated sludge (CMAS) reactor is expressed in Equation 12 as a function of the SRT and the kinetic growth and decay coefficients (Metcalf & Eddy, 2004); it is not a function of the influent soluble substrate concentration (sCOD), although the influent concentration affects the biomass concentration in Equation 11, with all terms as previously defined.

Equation 12: Effluent Substrate Concentration Mass Balance with Biomass Growth Kinetics

$$S = \frac{K_s[1+(k_d)\theta_c]}{\theta_c(\mu_n-k_d)-1}$$

The amount of nitrogen oxidized to nitrate is found by performing a nitrogen balance, considering that some of the nitrogen is assimilated into cell tissue of the biomass (Metcalf & Eddy, 2004):

Equation 13: Nitrogen oxidized to nitrate

$$(N_{OX}) = \left(N_{TKN,o}\right) - (N_e) - 0.12 \frac{P_{x,bio}}{Q}$$

Where:

N_e = effluent nitrogen concentration (mg/L)

 $N_{TKN,O}$ = effluent TKN concentration (mg/L) $P_{x,bio}$ = biomass as VSS wasted (kg/day)

The total mass solids in the reactor per day is based on TSS, which is the VSS plus the inorganic solids. The biomass typically contains 10-30% inorganic solids which are not soluble and are captured in the mixed liquor solids and removed through the waste sludge line. The mixed liquor total solids production is given by Equation 14 (Metcalf & Eddy, 2004).

Equation 14: Mixed Liquor Total Solids Production

$$P_{X,TSS} = \frac{A+B+C}{V} + D + Q(X_{TSS,o} - X_{VSS,o})$$

Where:

A,B,C,D = As defined previously

V = percent of volatile solids in total solids

 $P_{x,TSS}$ = net waste activated sludge produced each day (kg/day)

From the design data obtained above, the concentration and mass of VSS and TSS in the aeration reactors can be readily determined. The aeration reactor volume and hydraulic residence time is determined by selecting the desired MLSS for operation. Other parameters such as the F/M ratio, the BOD loading rate, and the expected yield can be found from standard design procedures. An example of an aerobic reactor design for BOD-removal and for BOD-and-Ammonia removal in given in Section 6.4 Aerobic Activated Sludge Main Process Design.

5.2.3 Aeration Energy

Oxygen requirements for the activated sludge process increase with the extent of nitrification reactions, therefore blower air usage and consequently power consumption increase. The oxygen demand, *R*, for nitrification in

Equation 15 includes the oxygen required for COD removal (1.42 g COD/g biomass VSS) and for the conversion of ammonia-nitrogen to nitrate-nitrogen (4.33 g O_2 / g TKN) using the amounts for substrate, biomass, and oxidized nitrogen found by previous Equations (Metcalf & Eddy, 2004).

Equation 15: Oxygen Requirements $R = Q(S_o - S) - 1.42P_{x,bio} + 4.33Q(N_{OX})$

The design of the oxygen delivery system is overall a complex process and is beyond the scope of this project, however some simplifying assumptions can be made to use Equation 16 to ascertain the impact of treatment on the oxygen demand and air usage (Metcalf & Eddy, 2004).

Equation 16: Standard Oxygen Transfer Rate

$$SOTR = R\left(\frac{C_{s,20}}{(\alpha)(F)(\beta C_{s,T,H} - C_{L})}\right) \left(1.024^{20-T}\right)$$

Where:

SOTR = Standard Oxygen Transfer Rate (kg O_2/hr)

 α, β = oxygen transfer correction factors

F = diffuser fouling factor

$$C_{s,T,H}$$
 = the average dissolved oxygen saturation concentration in clean water in the aeration basin at temperature T and altitude H

 C_L = the average dissolved oxygen saturation concentration in liquid bulk phase

The increased air demand for nitrification can be clearly seen in Figure 13. As the SRT increases, the nitrifier population grows and consumes more oxygen for nitrogenous removal in addition to the removal of organic carbon by heterotrophs.

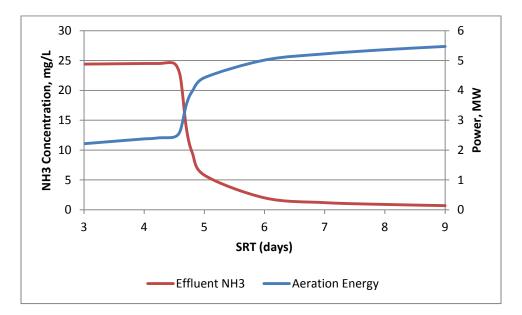


Figure 13: The effect of SRT on effluent ammonia concentration and aeration energy for a fine-bubble aeration system¹⁶

Air is produced by blowers whose motors account for a substantial portion of a plant's total demand. The air demand is a function of the SOTR and the diffuser efficiency. Therefore the efficiency of the diffusers is a major consideration in the lifecycle cost of the facility. The diffuser efficiency is reported in terms of standard oxygen transfer efficiency (SOTE) and may be reported in terms of % or %/basin depth.

¹⁶ Developed from the CHEApet simulation for a fine-bubble activated sludge plant with DO = 2.0 mg/L, presented in Section 6.86.8.

By increasing the rate of mass transfer of oxygen to the bulk fluid from the blower air, the air consumption can be reduced, resulting in operational cost savings as energy demand (kW) and consumption (kWh) is lowered. Oxygen transfer efficiency is proportional to the surface area of the bubbles, and smaller bubbles produced by fine-bubble diffusers have a greater surface area. Coarse-bubble aeration systems range between 3-7% OTE (e.g. 0.5%/ft), and fine-bubble aeration operate in the range of 20-35% OTE (e.g. 2%/ft) (Crawford, Johnson, Johnson, Krause, & Wilner, 2011).

As seen in Figure 14, fine bubble diffusers result in lower power demand for aeration compared to coarse bubble diffusers at the same depth. Deeper basins increase the residence time of the bubble in the water column. Fine bubble diffusers installed in deep basins (10 m) deliver oxygen to the wastewater more efficiently than shallower (e.g. 4 m) coarse-bubble reactors.

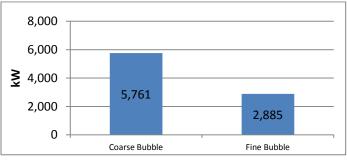


Figure 14: Aeration requirements for biological nitrification with coarse and fine bubble diffusers in basins with 4-m depth¹⁷

However, over time OTE for fine bubble diffusers decreases since they are prone to biofouling that reduces performance, and should be cleaned periodically to recover OTE (US Environmental Protection Agency, 1977). The fine bubble diffusers must be properly selected and subsequently maintained to keep the high oxygen transfer efficiency by removing excessive fouling and preventing diffuser damage. As seen in Figure 15, when the fouling factor is kept close to 1 the aeration energy consumption is at its minimum. For example, the difference between a fouling factor of between 0.9 and 0.7 could amount to 23 MWh/day. The rate of diffuser biofouling is site specific but there can be a big impact on energy usage. This is not a peripheral issue – fouling can become a major issue for a plant that is nitrifying.

¹⁷ Developed from the CHEApet simulations for activated sludge treatment, presented in Section 6.8.

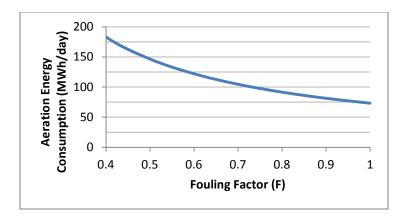


Figure 15: Impact of Fouling on Aeration Efficiency¹⁸

5.3 Biological Denitrification

In the previous section, first principles for carbonaceous BOD removal and nitrification were covered. This section presents an overview of biological denitrification as a subsequent treatment step, with focus on single-sludge (e.g. one solids clarifier) systems that can be extended to other type of anoxic system, such as denitritation. Heterotrophic denitrifiers play a major role in moving nitrogen from wastewater into the atmosphere on its journey through the global nitrogen cycle. The denitrification step converts the nitrite and nitrate produced during nitrification to nitrogen.

For denitrification to occur, an anoxic zone is required so that denitrifiers use nitrates as their oxygen source. The presence of dissolved oxygen can inhibit the nitrate reduction by repressing the nitrate reduction enzyme. Denitrifiers are strictly anoxic and typically the dissolved oxygen must be lower than 0.5 mg/L. Inhibition of a *Pseudomonas* culture has been reported at dissolved oxygen concentration as low as 0.2 mg/L (Metcalf & Eddy, 2004). Nitrate is used as the oxygen source for denitrifying bacteria so the denitrification process step provides an oxygen credit, reducing the actual oxygen requirements by 60%. Alkalinity is also recovered through the denitrification step. About 50% of alkalinity that was consumed during nitrification is produced during the denitrification reaction.

Denitrification is usually coupled with a nitrification process to ensure that nitrates are continuously fed to the anoxic denitrification reactor. There are three typical configurations (shown in Figure 16) for the placement of the anoxic zone in relation to the aerobic reactor:

1. <u>Anoxic/Aerobic</u> (Preanoxic): Nitrate is recycled to the pre-anoxic reactor. The rate of recycle impacts the amount of dissolved oxygen returned to the anoxic zone, and how much nitrate is recycled to the denitrification reactor;

¹⁸ Developed from aeration energy consumption for the nitrification reactor equipped with fine bubble diffusers, presented in Section 6.4.2.

- 2. <u>Aerobic/Anoxic</u> (Postanoxic): Since BOD (or rbCOD) is consumed in the aerobic process, denitrification will occur by endogenous respiration unless a source of exogenous carbon can be added to increase the denitrification rate; and
- 3. <u>Simultaneous Nitrification/Denitrification</u>: Aerobic and anoxic zones occur simultaneously, such as in a biofilm or granular sludge reactor.

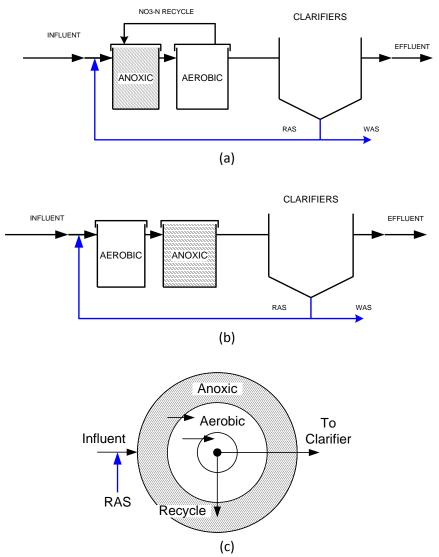


Figure 16: Schematic diagrams of process configurations for Total Nitrogen removal: (a) pre-anoxic, (b) post-anoxic, ad (e) simultaneous nitrification/denitrification

A supplemental carbon source may be needed if the wastewater is poor in compounds useable as electron donor, as such is the case for the postanoxic process. Where the carbon (i.e. rbCOD) has been previously consumed in the aeration step or is otherwise insufficient (such as centrate), providing supplemental carbon as an electron donor can vastly improve denitrification rates.

Adding denitrification to nitrifying WWTPs can reduce the energy that is required for treatment by 30% when compared to nitrifying-only AST plants (Wang, Hamburg, Pryor, Chandran, & Daigger, 2011). Firstly, less aeration is required to oxidize organic matter (60% could be recovered from the oxygen credit). Replacing part of the reactor volume of with an anoxic denitrification zone reduces the volume that needs to be aerated by increasing the mass loading to the aerobic section, if the nitrification reactor is overdesigned. Further, denitrification increases OTE by removing organic surfactants that surround air bubbles during nitrification.

Denitrification performance is a key element to reduce greenhouse gas emission potential. Incomplete denitrification will result in the production of NO and N_2O . Ensuring that the denitrification reactions are taken to completion may require additional carbon to be added.

5.3.1 Preanoxic Design

The steps for the design procedure are elaborated for the anoxic/aerobic configuration, including determination of the internal recycle ratio, the nitrate load, and the anoxic volume.

i Internal Recycle Ratio

The internal recycle ratio represents internal recycle flow as a fraction of the influent flow. A mass balance for the nitrates produced in the aerobic zone, including the return activated sludge recycle rate (*RAS*), effluent, and internal recycle flows, is given by Equation 17 (see Figure 17). An IR ratio range of 2 to 4 is typical; above *IR* of 4, the incremental removal of NO_3 -N is low versus the increased amount of DO recycled from the aeration zone in to the anoxic zone (Metcalf & Eddy, 2004). The equation can be used to find what internal recycle ratio must be to meet the target effluent nitrate concentration.

Equation 17: Internal Recycle Ratio

$$Q \times N_{OX} = N_e (Q + IR \times Q + R \times Q)$$
$$IR = \frac{N_{OX}}{N_e} - 1.0 - R$$

Where:

IR = Internal Recycle Ratio;

 N_{OX} = $TKN_{in} - NH_3 - N_{out}$ = Amount of ammonia to nitrate oxidized (mg/L);

 N_e = Total Nitrogen in the effluent (mg/L); and

R = Return Activated Sludge Recycle Ratio

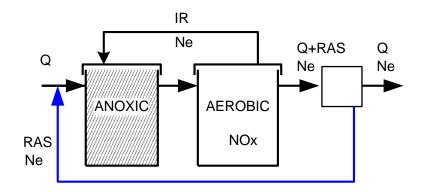


Figure 17: Schematic of mass balance for preanoxic Internal Recycle rate

The recirculation rate expressed as a percent of influent flow is typically 250-400% influent flow. The effect of the recirculation rate on the nitrate removal can be seen in Figure 18:

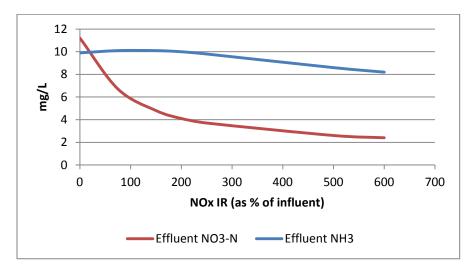


Figure 18: The impact of the internal recirculation rate on the effluent quality¹⁹

ii Anoxic Reactor Loading

In the anoxic reactor design, the nitrate load from the nitrification reactor is used to size the anoxic basin volume (V_{nox}) and find the hydraulic residence time.

The active biomass entering the anoxic volume is found from a mass balance of the substrate utilization in the aeration reactor shown in Equation 18, assuming steady state conditions.

Equation 18: Active biomass in the mixed liquor

$$X_b = \left[\frac{Q(\theta_c)}{V}\right] \left[\frac{Y(S_o - S)}{1 + (k_d)\theta_c}\right]$$

¹⁹ Developed from the CHEApet simulations presented in Section 6.8

Where:

- X_b = Active biomass in the mixed liquor (mg/L);
- k_d = kinetic decay term for heterotrophs;
- Y = Yield term for heterotrophs; and
- S, S_o = Substrate S is bCOD.

The food-to-microorganism (F/M_b) ratio is found from a function of BOD loading to the anoxic volume and the active heterotrophic biomass concentration, given in Equation 19 (all terms previously defined). The detailed design procedure includes the determination of the specific denitrification rate (SDNR in g/g-d) from literature, and corrected for the wastewater temperature such that the amount of NO₃-N that can be reduced with a 20 percent factor of safety.

Equation 19: Food-to-Microorganism ratio based for the anoxic volume

$$\frac{F}{M_b} = \frac{QS_o}{(V_{nox})X_b}$$

iii Oxygen Credit

The oxygen credit resulting from the addition of denitrification is found from the difference between the oxygen required for nitrification calculated using

Equation 15 versus the oxygen credit for denitrification using Equation 20.

Equation 20: Oxygen credit for nitrification/denitrification

$$O_C = O_{C,Nitrate}(N_{ox} - N_e)Q$$

Where:

 O_c = Oxygen credit $O_{C,Nitrate}$ = Oxygen credit for nitrate conversion (2.86 g O₂/ 1 g NO₃-N)

From Equation 15, the net oxygen required (R_o) after the oxygen credit is given by Equation 21. As a result, the aeration rate (m³/min) will decrease proportionally.

Equation 21: Net oxygen demand for nitrification/denitrification

$$R_0 = R - O_C$$

iv Mixing Energy

Adequate mixing of the anoxic zone is an important design consideration since mechanical mixers or jets will be needed to keep the biomass in suspension, ensure the uniformity of the reaction, and remove nitrogen and other gases from suspension. The power demand for mechanical mixing is between 5 W

per m³ of reactor volume (Water Environment Federation and the American Society of Civil Engineers/Environmental and Water Resources Institute , 2010) to 10 W/m³ (Metcalf & Eddy, 2004).

5.3.2 Postanoxic Design

For the design of a postanoxic denitrification system, the BOD (or sCOD) is already depleted by the aerobic process and denitrification will proceed slowly by endogenous respiration unless an exogenous source of carbon is provided. The amount of bsCOD required per mass of nitrate removed can be estimated from Equation 22, where the net biomass yield and the ratios of 1.42 g O_2/g VSS, and 2.86 g O_2/g NO₃-N.

Equation 22: COD required for Nitrate Reduction

$$\frac{bsCOD}{NO_3 - N} = \frac{2.86}{1 - 1.42Y_n}$$

As a rule of thumb, additional carbon may be required if COD/N Ratios are less than 4, the denitrification reactors are enlarged by a factor of 1.5 to 1.7. If less than 2.5, sufficient denitrification cannot be achieved without the use of exogenous carbon source (Hippen, Helmer, Kunst, Rosenwinkel, & Seyfried, 2001).

Methanol is commonly added to provide this extra source of carbon for denitrification. The required carbon demand found from Equation 22 is used to calculate the carbon dose (and resulting impact on operating cost), as shown in Table 4. An example is provided in Section 6.5.2 Suspended Growth Aerobic/Anoxic Denitrification Process.

 Table 4: Supplementary Carbon Sources²⁰

Carbon Source (cBOD)	Chemical Formula	COD (g/L)
Methanol	CH₃OH	1,173
Acetic Acid	CH ₃ COOH	604
50% Sugar Water (as Sucrose)	C ₁₂ H ₂₂ O ₁₁	487
Glycerol	CH ₃ H ₈ O ₃	1,536

In addition to the conventional liquid sources of readily biodegradable organic carbon, methane gas (CH₄) has been reported to enhance the denitrification process as a carbon source for heterotrophic denitrification. Methane is contained in the biogas produced during the anaerobic digestion process. As a potential source of inexpensive carbon, it would be an advantageous option for treatment plants to reduce operational costs (Waki, et al., 2009). Some amount of methane is typically dissolved in the sludge water. The mechanisms by which the methane is metabolized as a carbon source are varied. For example, the simultaneous occurrence of methane oxidation, aerobic ammonium oxidation, and

^A Adapted from (Crawford, Johnson, Johnson, Krause, & Wilner, 2011)

Anammox by a consortium of methanotrophs, nitrifiers, and planctomycetes has been observed (Waki, et al., 2009).

Recently other mechanisms have been elucidated by which methane is partially oxidized to methanol by the AMO enzyme used by ammonia oxidizing bacteria to normally used to metabolize ammonia. For example, Dr. Kartik Chandran from Columbia University received 2011 Paul L. Busch award from the Water Environment Research Foundation (Water Environment Research Foundation, 2011) for his work to turn methane in biogas into liquid methanol fuel. Although out of the scope of this discussion, the production of a liquid fuel via the novel application of an existing microbial pathway is an exciting development and a topic that is recommended for future study.

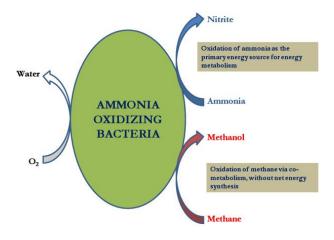


Figure 19: Schematic of co-metabolism in ammonia oxidizing bacteria using ammonia and methane, respectively²¹

5.4 Process Variations for Biological Nitrification/Denitrification

In general terms, the process consists of an aerobic reactor zone for nitrification to occur and an anoxic reactor zone (nominal HRT = 4 hrs) for denitrification to occur. Nitrification and denitrification processes have been excellently and thoroughly reviewed elsewhere and are included here for comparison to Anammox processes.

Variations of the conventional nitrification/denitrification process of the mainstream activated sludge reactor include:

- <u>Wuhrmann</u>: The Wuhrman Process places the anoxic basin after the nitrification zone (i.e. postanoxic). This process relies on nitrate produced in the preceding aeration basin as the oxygen source. Facultative bacteria that make up a majority of the MLSS perform the work of denitrification.
- <u>Modifier Ludzack-Ettinger (MLE)</u>: Flow enters the anoxic zone and then aerobic zone (i.e. preanoxic). An optional internal recycle from the aerobic zone to the anoxic zone may be used. In

²¹ From (Water Environment Research Foundation, 2011)

the MBR configuration, the RAS may enter the aerobic zone. For non-MBR configurations, RAS enters the anoxic zone.

• <u>Bardenpho</u>: a four-stage process Influent flows into the preanoxic zone and then into an aerobic zone. An internal recycle draws from this zone to the pre-anoxic. Following the aerobic zone is an anoxic zone which allows for the addition of supplemental methanol. Mixed liquor is then reaerated prior to entering the secondary clarifier. RAS enters the bioreactor at the first aerobic zone for an MBR configuration; otherwise it enters in the pre-anoxic zone

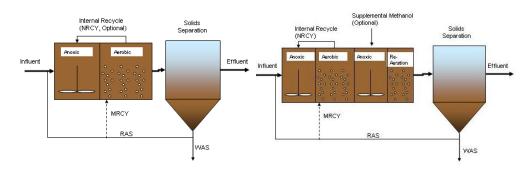


Figure 20: (a) Modified Ludzack-Ettinger (b) Bardenpho²²

The waste activated sludge (WAS) from the BNR treatment is processed by anaerobic digestion then dewatered; it is proposed in this project that the centrate or sludge-water side-stream be treated with a Partial Nitritation-Anammox process in a side-stream reactor prior to recycle back through the mainstream activated sludge process, described in Section 6.6 Side-stream Treatment Facilities Sizing.

5.5 Biological Nitritation

The previous sections provided a review of the essential design considerations for a conventional biological Total Nitrogen removal process that makes use of nitrification for ammonia removal followed by denitrification for nitrate removal. The advantage to controlling the extent of nitrification can be seen in the Redox pyramid in Figure 21, such that nitrite is produced but is halted before the nitrite is converted to nitrate by NOBs is that ammonia is converted to nitrite with 25% less oxygen is used over complete nitrification to nitrate, and 60% less exogenous carbon.

²² Adapted from (Crawford, Johnson, Johnson, Krause, & Wilner, 2011)

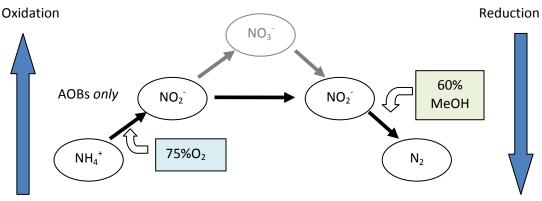


Figure 21: Nitritation- Denitritation Redox Pyramid

Nitrite is typically a short-lived intermediate product that is consumed by NOBs in nitrification. Therefore, depending on the control strategy, the nitrite can be (1) consumed by NOBs for further nitratation (with further oxygen and NOB enrichment) or (2) by denitritation, or by Anammox. Nitrite is an important precursor to each of these processes, and can be a controlling factor in their operation. Strategies for successful nitritation and denitritation are discussed with respect to control strategies and inputs (e.g. chemical and energy).

5.5.1 Nitritation Control Strategies

To achieve nitritation without nitratation, the growth of NOBS must be suppressed to prevent the conversion of nitrite to nitrate. Terminating aeration prior to complete ammonia oxidation is a key factor to sustained partial nitritation. The growth of NOBs can be suppressed based on certain physiological differences from AOBs (Liang, Shangyuan, Liang, Du, Liu, & Yang, 2011). Online control of ammonium oxidation can be based on various instrumentation strategies, such as DO, ammonium, conductivity sensors, oxygen uptake rate (OUR), and pH measurements.

Stability of the nitritation reaction has historically been a challenge. Stability is an important precondition to a downstream Anammox reactor. Stable control has been achieved with pH 7.8 +/- 0.2, temperature of 30+/-1 °C and DO of 0.5 to 0.8 mg/L (Liang, Shangyuan, Liang, Du, Liu, & Yang, 2011).

A number of different control strategies have been discussed in the literature. A overview of nitritation control strategies includes: (1) Suppressed Dissolved Oxygen; (2) Temperature Control; (3) Real-time pH Adjustment; (4) Retention Time Control; (5) Alkalinity to Ammonia Ratio Control; (6) Nitrite to Ammonium Ratio; and (7) Inorganic Carbon to Ammonia Ratio, as discussed in Liang, Shangyuan, et. al (2011). These are described in more detail:

1. <u>Suppressed Dissolved Oxygen</u>: In the suppressed dissolved oxygen control strategy, controlled aeration is used to select for AOBs, which have higher affinity for DO than NOBs, since *Nitrobacter* have a lag phase that is longer than *Nitrosomonas*. A low DO level (less than 1.0

mg/L) is used to inhibit the activity of NOBS. Blower operation is used to control the DO in the required range with a proportional-integral-derivative (PID) controller. Dissolved oxygen control with intermittent aeration is a feasible strategy to enhance ammonia conversion to nitrate and limit NOB activity and also saves on energy costs. The DO level is similar to that under anoxic conditions is crucial for achieving a stable partial nitritation reaction as a pretreatment of the side-stream to the Anammox reactor. Dissolved oxygen concentration lower than 0.5 mg/L has been shown to be suitable for stable partial nitritation reactor (Zhang, Yang, Hira, Fujii, & Furukawa, 2011). Using the DO control approach, intermittent aeration of a nitrification reactor will lead to nitrite enrichment if aeration time is adjusted. This method of discontinuous aeration is used at the full-scale plant at Hattingen to achieve nitritation (Rosenwinkel, Cornelius, & Thole, 2005)

2. <u>Temperature Control</u>: Control based on temperatures above 30°C enable AOBs to prevail over NOBs due to AOB's faster growth rate under elevated temperatures, as seen in Figure 22. At low temperature of up to 12°C, the growth rate of *Nitrobacter* is higher than *Nitrosomonas*. From 12°C to 25°C, the growth rate of *Nitrosomonas* is higher than *Nitrobacter*. Above 25°C the rate becomes higher, to the maximum rate at 33°C. At temperatures above than 36°C, nitrite production is inhibited. Therefore the optimal temperature is 31-34°C. A detailed heat balance should be performed to determine if there are cooling or heating requirements for the reaction. For side-stream treatment, the temperature of the recycle stream from the mesophilic anaerobic digestion process is already in this range.

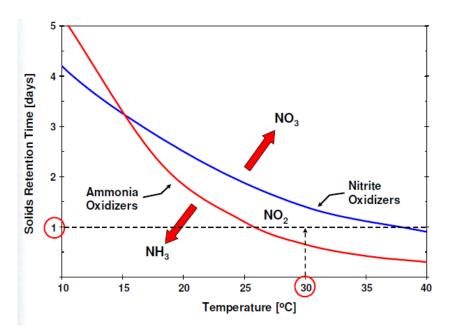


Figure 22: SRT versus temperature for AOB and NOB²³

²³ Adapted from (Constantine, 2008)

- 3. <u>Real-time pH Control</u>: Real-time pH control can be used to suppress the NOBs using the alkaline condition to limit the conversion of nitrite into nitrous acid (which inhibits AOB) and equilibrates with concentrations of free ammonia to selectively inhibit NOB. Inhibition of AOB (*Nitrosomonas*) occurs at 0.10 mg HNO₂-N/L and 10 to150 mg NH₃/L, while inhibition of NOB (Nitrobacter) occurs at 0.011 mg HNO₂-N/L and 0.1 to 10 mg NH₃/L. This method of control is used to obtain a suitable free ammonia concentration to inhibit NO₂-N oxidation to NO₃-N. However, malfunction of the pH meter would affect the stability of the Nitritation process. Another drawback is that the cost for real-time pH adjustment can be high.
- 4. <u>Residence Time Control</u>: Residence time (HRT and SRT) can be used to prevent the oxidation of nitrite to nitrate or to facilitate NOB washout in the activated sludge system. In conditions that favour AOB over NOB, such as higher temperature, the NOB can be washed out of the biomass by reduction of the SRT to 2-3 days, and HRT above 16-h and below 8-h might result from the prolonged and shortened reaction times (depending on the growth kinetics).
- 5. <u>Alkalinity Ratio Control (Alk/NH₄-N):</u> The reaction rate of AOB is accelerated by a high Alkalinity to Ammonium Ratio. With alkalinity ratio control, influent HCO₃/NH₄ ratio should be 1.0 to achieve 50% ammonium oxidation to control the effluent. However, the continuous adjustment of alkalinity can be costly (e.g. calcium carbonate). Nitrification consumes alkalinity and can become a limiting factor in the reaction. Influent alkalinity is closely related to the production and accumulation of nitrite in the Nitritation reactor. An Alkalinity/NH₄-N ratio of around 4.8 was optimal for the Partial Nitritation process (described in Section 5.7.1), with the rate of NH₄-N removal at a maximum (90 mg NOx-N/L/h). Controlling the Alk/NH₄-N influent ratio was shown to be a feasible and effective way to control the Partial Nitritation reactor (Zhang, Yang, Hira, Fujii, & Furukawa, 2011).
- 6. <u>Nitrite to Ammonium Ratio Control(NO₂-N/NH₄-N)</u>: The most frequently cited control strategy for partial nitritation was control over the Nitrite to Ammonium Ratio (NAR). Effluent Nitrite Ammonium Ratio (NAR) can be determined by online sensors and/or mass balance. For example, if nitrite is measured with sensor, a mass balance approach can be used to estimate the concentration of ammonia. Alternatively, two different sensors can be used to measure the NAR. Aeration blowers are used to control the Nitrite to Ammonium Ratio in the required range with a proportional-integral-derivative (PID) controller. The stoichiometric ratio for NAR is 1:1.3 but the empirical evidence suggests that 1:1 is better to prevent the build-up of toxic levels of NO₂-N. When influent T-N concentration was high, the variation of NAR would have a greater effect on reactor performance than if the T-N concentration was low (Li, Ma, Hira, Fujii, & Furukawa, 2011).
- 7. Inorganic Carbon Control (IC/NO₃-N): Inorganic carbon control can be used to eliminate NOB from the biofilm as they are outcompeted by the AOB, (Tokutomi, Yamauchi, Nishimura, Yoda, & Abma, 2011). In the nitritation phase, IC in the bicarbonate form can be used as a carbon source or to neutralize hydrogen ions produced during partial nitrification. Therefore, the ratio of inorganic carbon to nitrate-nitrogen is a crucial parameter for reactor stability. A ratio of

Inorganic carbon (IC) to nitrate-nitrogen of 0.8 was found to be favourable. (Zhang, Yang, Hira, Fujii, & Furukawa, 2011)

5.5.2 Process Variations

A variety of process configurations for nitritation have been developed to implement the concept for full-scale applications. For example, the SHARON (Single reactor for High activity Ammonium Removal Over Nitrite) process developed in Delft, the Netherlands, is used to remove ammonia from wastewater.

The SHARON process is operated with minimal solids residence time (equal to hydraulic residence time) and at an elevated temperature to select *Nitrosomas* (nitrite-forming AOBs) over *Nitrobacter* (nitrate-forming NOBs) (Rosenwinkel, Cornelius, & Thole, 2005). Although high rates of ammonia removal are possible, the SHARON reactor can be tuned to different nitrogen conversion rates depending on downstream operational needs (such as if operated upstream of an Anammox reactor).

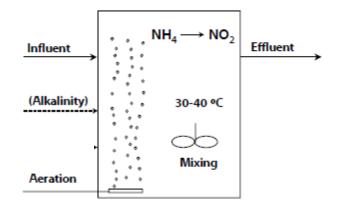


Figure 23: Schematic of SHARON Reactor for nitritation²⁴

When used as a pretreatment to the Anammox process, the target for nitritation is conversion of approximately half of the ammonia to nitrite to feed the Anammox bacteria with a mixture that is suitable for their metabolism. This is discussed in further detail in Section 5.7 Biological Anammox.

5.6 Biological Denitritation

Following the nitritation process, biological denitritation is a novel treatment method that converts the nitrite directly to nitrogen. Denitritation saves 40% of carbon over the conventional biological denitrification process since it skips the reaction steps to convert nitrate back into the nitrite (see Figure 21). Design and operational considerations for denitritation are similar to those developed for the full denitrification reaction, except that the amount of nitrogen oxidized (NO_x) is composed of nitrite instead of nitrate, which has a lower stoichiometric demand for organic carbon than the full denitrification

²⁴ Adapted from (Pennsylvania Department of Environmental Protection, 2002)

reaction. Therefore, the oxygen credit is lower for denitritation than for denitrification, or 1.72 g O_2 per g NO₂-N.

Equation 23: Oxygen credit for nitritation/denitritation

$$O_C = O_{C,Nitrite}(N_{ox} - N_e)Q$$

Where:

 $O_{C,Nitrite}$ = Oxygen credit for nitrite conversion (1.72 g O₂/ 1 g NO₃-N)

5.7 Biological Anammox

The biological Anammox process is a novel alternative to denitritation or conventional denitrification. This section briefly touches on some of the pertinent design considerations for Anammox reactors, such as common process control strategies, and biomass residence. A more detailed discussion of the implementation of Anammox is given in the following section on side-stream treatment reactor configurations.

The physiology of Anammox bacterium has several advantageous implications for nitrogen removal. Similarly to denitrification, the process is anaerobic and by definition does not require aeration. As chemolithotrophs, Anammox bacteria do not require a source of organic carbon for energy or cell growth. In contrast to denitrification, there is no oxygen credit resulting from nitrogen removal through Anammox. As discussed in Section 4.3 Characteristics of Anammox Bacteria, several obstacles exist in the full-scale implementation of biological Anammox process related to the slow growth rate, and sensitivity of the Anammox bacteria to dissolved oxygen. For example, the Zürich WWTP experienced a significant loss of Anammox activity due to toxic shock during start-up that caused a set-back lasting over 1 month (Joss, et al., 2009).

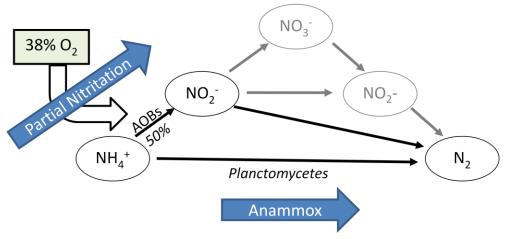


Figure 24: Partial Nitritation– Anammox pathway

5.7.1 Process Control for Anammox Bioreactor

General guidelines for an Anammox bioreactor process control strategy include:

- (1) Keep inhibitors such as dissolved oxygen and methanol out of the reactor;
- (2) Keep the organic carbon source limited. Organic carbon or sources of light increase the growth rate of heterotrophic and autotrophic bacteria that will out-compete the Anammox bacteria;
- (3) Feed the Anammox bacteria with the correct ratio of nitrites and ammonia using Partial Nitritation; and
- (4) Retain the Anammox biomass for SRT > 11 days and more. This can be accomplished through the use of suspended growth processes such a as granular sludge reactors or membrane bioreactors, or fixed film processes such as fixed- or moving- bed reactors, or rotating biological contactors (RBCs).

The main challenges for the control of the Anammox bioreactor involves the stability of the ammonium and nitrite feed and the residence of the slow-growing Anammox biomass. The influent to the Anammox reactor must be monitored and controlled to ensure process stability. Partial Nitritation is used to pretreat the feed to the reactor. The goal of the Partial Nitritation process is to supply an ideal proportion of nitrite and ammonia to the Anammox process. It is important to distinguish between partial nitrification, and the *Partial Nitritation* control strategy. Partial nitrification refers to the incomplete removal of ammonia and nitrite by AOBs and NOBs. By contrast, the target of *Partial Nitritation* is to achieve 50% conversion of ammonium to nitrite as a pretreatment for the Anammox process. Therefore, the stability of the partial nitritation process is a key component of successful control of the Anammox process.

The optimum pH range of the Anammox reaction was reported to be 7.0 to 8.0 (Strous, Kuenen, & Jetten, 1999). The pH shifts to the alkaline side after the Anammox reaction so the lower side of the optimum is selected as the operational range (Tokutomi, Yamauchi, Nishimura, Yoda, & Abma, 2011).

For the lab-scale reactors described in the literature, the performance of the Anammox reactor is impacted by the hydraulic residence time (HRT). The nitrogen removal rate increases as the HRT is increased, with the maximum occurring removal at 8 hr (Tsushima, Ogasawara, Kindaichi, Satoh, & Okabe, 2007).

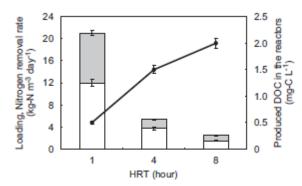


Figure 25: Nitrogen removal rate as a function of HRT²⁵

Another consideration of note is that of the heat balance for the Anammox reactor. In general, nitrogenreducing oxidation reactions are exothermic. However, the Anammox is not strongly exothermic. A heat balance model should be developed to determine the impacts inside the reactor and to size the heat removal mechanism. Temperatures within the reactors need to be maintained below 37°C and above 20°C to ensure process stability. Maintaining a process temperature of between 30 and 35°C optimizes the biological kinetics. Optimal conditions for the Anammox process are summarized in Table 5.

Table 5: Optimum Anammox Parameters

Temperature (oC)	30-35
рН	7.0-8.0
SRT (days)	11+
HRT (hours)	8
Nitrite to Ammonia Ratio	1-1.3

5.7.2 Biomass Retention

Wastewater processes for activated sludge treatment are categorized as fixed- or suspended-growth processes. Biomass residence is one of the main challenges to the successful engineering application of the Anammox process. The slowly-growing Anammox necessitate a long SRT in order to retain the biomass. The Anammox bacteria require an environment that is strictly anaerobic environment, with low organic carbon so they are not out-competed, and supply of approximately half ammonia and half nitrite. It is no wonder that in some cases it took many years to successfully cultivate Anammox sludge in an enrichment reactor, before bringing the Anammox process to a full-scale process (Tokutomi, Yamauchi, Nishimura, Yoda, & Abma, 2011).

²⁵ From (Tsushima, Ogasawara, Kindaichi, Satoh, & Okabe, 2007), where the whole bars represent loading rates and the white parts represent the nitrogen removal rates. The line plots represent the produced dissolved organic content produced in the reactor

i Fixed Growth

Fixed growth processes rely on the growth of a layer of biofilm, such as the structure shown in Figure 26, as the mechanism to retain biomass in the reactor. The conversion rate of the reactor is defined by penetration depth in the biofilm and thus by the surface area of the biomass, biomass thickness, and nitrite level. Biofilm deammonification is an example of a biofilm Anammox system, in which a two step process is performed by two groups of organisms sequentially. Both steps can occur within the same biofilm structure, simultaneously.

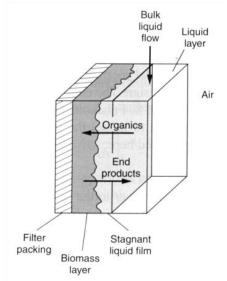


Figure 26: Example of a biofilm structure²⁶

Fixed growth reactors include fixed bed up-flow reactors, moving bed reactors, and rotating biological contactors:

- <u>Rotating Biological Contactors (RBC)</u>: A layer of biofilm builds on rotating discs that are partially submerged in wastewater. The depth of the biofilm is affected by operational factors such as the speed of the rotors, the depth of submergence, air flow rate, and size of the anoxic zone.
- <u>Fixed Bed</u>: In a fixed bed up-flow reactor, biogrowth attachment occurs on a fixed bed of carrier material. Polyester non-woven material, acrylic resin material, gel beads and other materials were developed for immobilization of the Anammox sludge (Ma, Hira, Li, Chen, & Furukawa, 2011). Polyethylene sponge was used with success at lab scale as a biomass carrier for Anammox bacteria (Zhang, Yang, Fujii, Hira, Zhang, & Furukawa, 2011), but is otherwise an obsolete technology for full-scale implementation.
- <u>Moving Bed</u>: Moving bed reactors use a carrier material that is suspended by mixing jets and mechanical mixers. The mixing also prevents the localized buildup of reaction products that would inhibit the reaction or have toxic effects, such as hydrogen sulfide. Examples include the

²⁶ Adapted from (Metcalf & Eddy, 2004)

KALDNES[®] reactor, which uses an engineered plastic carrier media for biomass growth (Figure 36).

It is first necessary to build up a biofilm structure in which the Anammox bacteria can be settled afterwards. Members of the planctomycetales group seem not be able to build up their own biofilm structure but need an existing biofilm to enrich. Therefore, the first step is an aerobic operation with high substrate load and adequate aeration which will result in a mixed biocoenosis containing aerobic and anoxic ammonia oxidizers (Rosenwinkel, Cornelius, & Thole, 2005).

The thickness of a biofilm is limited by the shear forces caused by mixing. The biofilm thickness of 1 to 2 cm for a rotating biological contactor allows for separation into outer aerobic and inner anoxic zones so that there is sufficient habitat for the nitrifiers, the denitrifiers, and Anammox bacteria. In contrast, there are higher shearing forces on the carrier material in a moving bed reactor. Depending on the carrier material and conditions, a biofilm thickness of 100-500 μ m can be achieved. This is not sufficient to serve as habitat for both aerobic nitrifiers and the anaerobic Anammox bacteria. Since the diffusion of oxygen through the biofilm defines the habitat for the aerobic or anoxic/anaerobic organisms, separation of the process into two steps can be more efficient (Hippen, Helmer, Kunst, Rosenwinkel, & Seyfried, 2001).

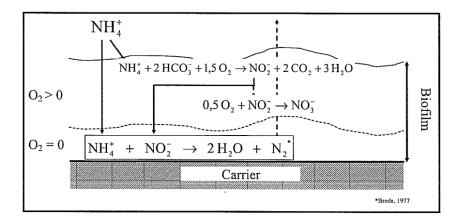
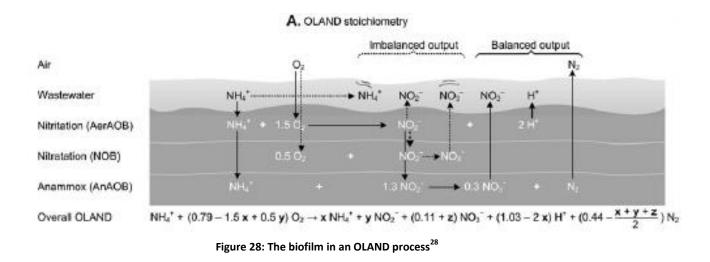


Figure 27: Deammonification in a biofilm²⁷

²⁷ Adapted from (Rosenwinkel, Cornelius, & Thole, 2005)



In an Anammox biofilm system, FISH analysis showed that AOBs and Anammox bacteria were distributed throughout the biofilm, with Anammox accounting for more than 70% of total bacteria, as shown in Figure 27 (Tsushima, Ogasawara, Kindaichi, Satoh, & Okabe, 2007).

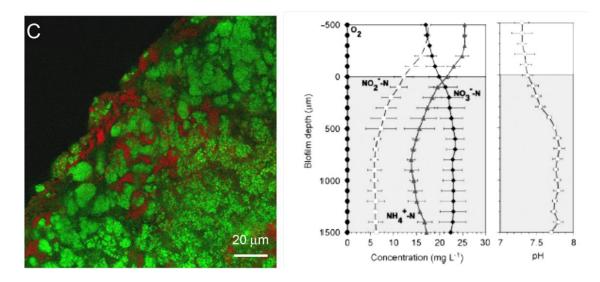


Figure 29: (a) FISH image with red markers specific to Anammox bacteria in a 30 μm thick section of Anammox biofilm; (b) steady-state concentration profiles in the Anammox biofilm²⁹

ii Suspended Growth

Biomass residence in a suspended growth system is exemplified by the conventional activated sludge process with floccular sludge that is settled in the secondary clarifier and recycled or wasted. However, due to the extended SRT required for cultivation of Anammox bacteria, the anoxic reactor volume required for a conventional process with flocky sludge would be impractically large (as shown in Section

²⁸ Adapted from (Clippeleir, Yan, Verstraete, & Vlaeminck, 2011)

²⁹ Adapted from (Tsushima, Ogasawara, Kindaichi, Satoh, & Okabe, 2007)

6.6.3 Side-stream Partial Nitritation/Anammox). A higher density of biomass is therefore required. Suspended growth processes that can support the high sludge residence age required for Anammox growth include granular sludge reactors and membrane bioreactors.

Granular sludge is characterized by high density agglomerations of biomass with high surface area, seen in Figure 30. In a granular sludge, the depth of the agglomerated sludge particle is such that it can support both an aerobic and an anoxic zone within its volume, as shown in Figure 30(a). Suspended growth processes for advanced biological removal of Total Nitrogen may rely on the simultaneous occurrence of aerobic nitrifiers on the outside of the particle and denitrifiers or Anammox bacteria within the particle's anoxic zone.

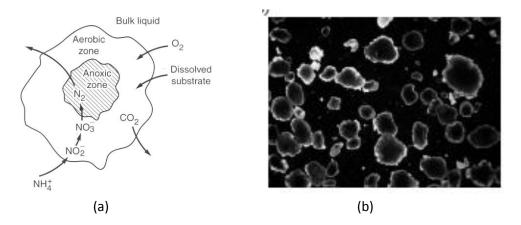


Figure 30: (a) Activated sludge with aerobic and anoxic zones³⁰; (b) Anammox granules³¹

Granules must be produced of sufficiently high quality such that good biomass residence and stable operation is guaranteed (Abma, et al., 2007). Granule size varies, tending to increase in average size during operation to a range of 0.8 to 1.5 mm in diameter, observed in Figure 31. The granule biomass population may include a mixed population of heterotrophs and methanogens or consist of Anammox bacteria only. The characteristic dark red colour of Anammox has been noted in both methanogenic and Anammox-only granules (Tokutomi, Yamauchi, Nishimura, Yoda, & Abma, 2011).

³⁰ Adapted from (Metcalf & Eddy, 2004)

³¹ Adapted from (Abma, et al., 2007)

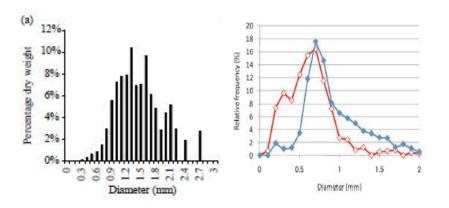


Figure 31: (a size distribution of the biomass³²; (b) size distribution of Anammox granule³³

The volumetric loading capacity for the bioreactor is increased proportionally to the available contact surface area. The effective surface area of a moving bed biofilm reactor (depending on the packing type) is around 200 m²/m³. By comparison, this can be up to 3,000 m²/m³ in granular sludge reactors. Additionally, by using a granular sludge the cost of carrier material for fixed-growth processes can be avoided. For example, the Rotterdam WWTP converted the existing Moving Bed reactor to a Granular Sludge reactor at full-scale, with removal rates of 10 kg-N/m³ day (Abma, et al., 2007).

Membrane bioreactors (MBR) are a high-biomass residence technology for wastewater treatment that couples membrane filtration with biological treatment. Membrane-based nitrogen removal systems can achieve TKN removal efficiency >99%, comparable to conventional systems. The application of MBR to the Anammox process has been successfully demonstrated in facilities such as the Pasakoy Advanced Biological Waster Treatment Plant municipal plant in Istanbul, Turkey with microfiltration MBR (Ozdemir, Mertoglu, Yapsakli, Aliyazicioglu, Saatci, & Yenigun, 2011).

There are several advantages of an MBR system over a conventional aeration basin and clarifier system:

- High mixed liquor suspended solids concentration (>10,000 mg/L) can be maintained without affecting settling properties of the sludge;
- The SRT can be controlled independently of HRT to retain slow-growing microorganisms. At high SRT, complete nitrification and nitrate removal by the Anammox process can be accomplished; and
- A lower footprint area required compared to conventional systems.

A major operational drawback is the issue of membrane fouling. Fouling of the membrane is caused by the production of extracellular polymeric substances at high SRT. This can be mediated by reducing the

³² Adapted from (Abma, et al., 2007)

³³ Adapted from (Tokutomi, Yamauchi, Nishimura, Yoda, & Abma, 2011)

SRT from 20 days to 10 days and operating with higher MLSS concentration by lowering HRT (Ozdemir, Mertoglu, Yapsakli, Aliyazicioglu, Saatci, & Yenigun, 2011). The applications of Anammox systems using MBR are recommended for further investigation.

5.8 Configurations for Side-stream Treatment

The previous sections provided a discussion of both conventional and novel nitrogen removal technologies for wastewater treatment from the main treatment stream. In this section, it is shown that the biological processes for nitrogen removal that were applied to the mainstream activated sludge reactor can be modified for use in side-stream treatment.

For removal of nitrogen from a side-stream such as dewatering centrate or anaerobic digester supernatant, novel reactor configurations are possible due to the unique characteristics of sludge water -- namely that it has a low carbon to nitrogen ratio, and elevated temperatures resulting from mesophilic anaerobic digestion for enhanced growth kinetics. These configurations are demonstrated in Section 6.6 Side-stream Treatment Facilities Sizing.

5.8.1 Nitrification/Denitrification

Side-stream loads can be treated by conventional nitrification/denitrification. However, the low COD/N ratio of side-stream flow would require the addition of supplemental carbon by methanol or acetate, such as for a postanoxic denitrification configuration.

The elevated temperature of the side-stream lends itself to faster growth kinetics so there is a slight advantage in terms the incremental reactor volume reduction for treating the side-stream load separately from the mainstream BNR process. The aeration requirements and sludge production would be similar to that if the side-stream load were routed to the main plant.

5.8.2 Nitritation/Denitritation

Nitritation coupled with denitritation for side-stream treatment is a potentially cost-effective alternative to conventional nitrification/denitrification. The operational strategy is to limit nitrite oxidation to nitrate. The combination of (1) nitritation to oxidize ammonia to nitrite, and (2) denitritation to reduce nitrite to nitrogen, is used to remove the nitrogen load from the side-stream.

Nitritation/denitritation systems will typically have smaller volumes, require less process mechanical energy for mixing, and consume 25-30% less electrical energy for blower air use and about 40% less exogenous carbon than an equivalent nitrification/denitrification system (Johnson, Sanjines, Castaneda, & Daigger, 2011).

Multiple designs for reactor configuration, both proprietary and non-proprietary, have been developed. For example, use of the SHARON technology has achieved ammonia removal efficiencies of greater than 95%. There are multiple full-scale SHARON plants currently operating in the cities of Rotterdam, Utrecht, Zwolle, Beverwijk, with a facility under construction at New York City's Wards Island Water Pollution control plant (Johnson, Sanjines, Castaneda, & Daigger, 2011).

The two-stage SHARON process shown in Figure 32 is a one-reactor suspended-growth high-rate nitritation/denitritation process. The process is run in aerobic mode alternating with anoxic mode to accomplish both oxidation and reduction from nitritation and denitritation, without full nitrification. The two-stage SHARON process can be operated in an intermittent aerated reactor or a continuous flow staged reactor. Alternatively, a two-step configuration can be used to separate the reactions into two stages.

The two-stage SHARON reactor operates with SRT equal to the HRT (i.e. no sludge residence), is controlled by dissolved oxygen concentration, and operates at elevated temperature. There is no need to develop large clusters of microbial floc since the reactor is run with small clusters of bacteria. No sludge residence allows the selection of *Nitrosomas* (i.e. AOBs) bacteria over *Nitrobacter* (i.e. NOBs). The oxidation of ammonia is stopped at nitrite, rather than left to run all the way to nitrate. In the anoxic mode, denitritation bacteria in the SHARON reactor convert the available nitrite to nitrogen, as in Equation 6.

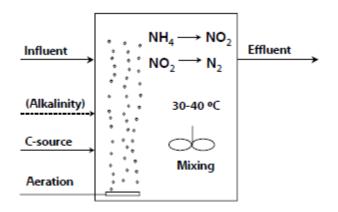


Figure 32: Schematic of Two-Stage Nitritation/Denitritation SHARON Reactor³⁴

5.8.3 Partial-Nitritation/Anammox

As previously described, the Partial-Nitritation/Anammox process uses ammonia oxidizing bacteria for nitritation, followed by an autotrophic Anammox process. Half the ammonia is oxidized to nitrite which is consumed by Anammox bacteria along with the remaining ammonia to produce nitrogen. Since nitrite serves as the electron donor for the formation of biomass from carbon dioxide, the formation of nitrate is coupled to the growth by stoichiometric ratios. Ammonia is used by the biomass to reduce the nitrite, so supplemental organic carbon is not required. The combination of Equation 2 and Equation 7 results in nitrogen removal according to Equation 24 (Third, Sleikers, Kuene, & Jetten, 2001):

³⁴ (Pennsylvania Department of Environmental Protection, 2002)

Equation 24: Nitrogen Removal by Nitritation/Anammox

 $NH_4^+ + 0.85O_2 \rightarrow 0.435N_2 + 0.13NO_3^- + 1.3HO + 1.4H^+$

5.8.4 Reactor Configurations for Anammox Processes

Biological nitrogen removal is a two-step process, and can be implemented one step at a time in sequential reactors or both accomplished in a single reactor. In the literature there has been much discussion, research and design on both approaches. Which is better? It could be that the difference in strategy boils down to implementation and eventual operability. The sludge cultures could be kept separate in a two-reactor configuration, or together in mixed sludge in a single-reactor system.

There are pros and cons for both one- and two- reactor systems:

- <u>Two-Reactor</u>: Separate reactors entail higher investment costs related to the construction of different reactors, but this configuration allows the user to attune and optimize the conversion stages individually. The reaction steps are separated into two separate reactors in sequence. In the first reactor, Partial Nitritation (PN) feeds an ideal mixture of ammonia and nitrite to the Anammox bacteria in the downstream reactor. With such a reactor is it possible to control HRT and SRT independently for each reactor. Operating the Anammox process in a separate reactor allows up to 10 times higher turnover rates for the anaerobic step (Joss, et al., 2009).
- <u>One-Reactor</u>: A single reactor configuration combines both treatment steps into one reactor. A reactor such as an SBR (suspended growth) can be run under an alternating regime of aerobic and anoxic modes. A fixed growth reactor such as an RBC can support both aerobic nitrifiers and anaerobic Anammox in the same biofilm structure.

Since two-reactor configurations are conceptually easier to follow, they are presented first, followed by a description of some of the existing one-reactor systems for nitrogen removal from the side-stream using the Anammox process.

i Two-Reactor Configurations

Variations of two-reactor configurations for side-stream nitrogen removal are briefly described. They include (1) the SHARON-ANAMMOX process and (2) the Novel Activated Sludge process.

SHARON-Anammox was described as an "embryonic technology" by the US Environmental Protection Agency (US EPA, 2008). The SHARON-Anammox reactor configuration entails the Partial Nitritation of ammonia by SHARON and the subsequent anoxic oxidation of the residual ammonia and nitrite to nitrogen by Anammox. About half the amount of ammonia is oxidized to nitrite and then residual ammonia and nitrite is anoxically reduced to elementary nitrogen. The total nitrogen removal is accomplished using a stoichiometric oxygen demand of only 40 percent compared to conventional nitrification/denitirifcation, with no requirement for supplemental carbon.

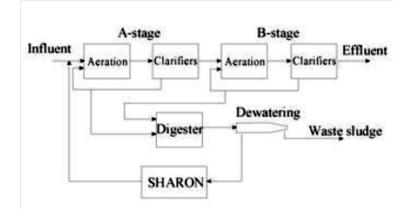


Figure 33: Schematics of SHARON-Anammox Process³⁵

Novel Activated Sludge (NAS) is a hybrid floc-based nitrogen removal system that was built at an industrial WWTP (in Bergen op Zoom, The Netherlands) without carbon addition or pH adjustment, based on the control of SRT and DO levels, , to follow the production cycle of a potato processing plant The NAS system is multiple-stage nitrogen removal process, consisting of (1) partial nitritation; (2) Anammox; (3) denitrification; (4) nitrification; and a (5) final settler. In effect, the nitrate produced by the Anammox process reduced by the nitrification/denitrification reaction to polish the final effluent. An overall total nitrogen removal efficiency of 95% was reported with 40% savings in carbon costs (Desloover, et al., 2011).

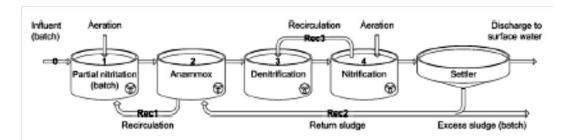


Figure 34: Schematic overview of the examined four-stage hybrid NAS nitrogen removal process³⁶

ii One Reactor Configurations

The one-reactor process relies on the interaction of two groups of autotrophic bacteria under oxygenlimited environment that perform two sequential reactions, simultaneously, using either a two-phase growth of biofilm with sufficient depth to support both aerobic and anaerobic organisms, or granules with an aerobic surface and anoxic/anaerobic centers. Such reactors are operated with low-DO

³⁵ Adapted from (Pennsylvania Department of Environmental Protection, 2002)

³⁶ Adapted from(Desloover, et al., 2011)

concentration. The oxygen is consumed by nitrifiers on the surface of the biofilm or granule, which in turn provide nitrite to the Anammox bacteria in the anoxic environment.

Operating a one-step reactor simplifies reactor control, saving space and provides in-built prevention of toxic build-up of intermediate products. During regular operation, nitrite is continuously depleted, avoiding potentially toxic feed concentrations required if the second step if fed with NAR > 1. Control strategies vary, but may include pH- or NAR-based controllers to trigger blower operation.

One reactor configurations that support the simultaneous processes of nitritation and deammonification include: (1) DEMON; (2) DeAmmon; (3) CANON; (4) OLAND; and (5) Hybrid Reactors.

(1) DEMON[®] (DE-amMONification) allows both nitrifying and deammonifying bacteria to co-exist in a single reactor despite different growth demands of the group through controlled aeration sequenced batch reactor (SBR).

Full-scale DEMON is used in two full-scale plants in Strass WWTP, Austria (since 2004) and in Glarnerland WWTP, Switzerland (since 2007). The vendor that is promoting the process is World Water Works in the US, or M2T Tech in Europe. In Virginia in the US, WWTPs at Upper Occoquan and Alexandria will use this process (Johnson, Sanjines, Castaneda, & Daigger, 2011). DEMON is also used to treat waste from a potato factory in Olburgen, Denmark since 2006 (Joss, et al., 2009).



Figure 35: The DEMON[®] Process (a) aerobic conditions; (b) red Anammox biomass is clearly seen³⁷

(2) DeAmmon[®] is a one step process that incorporates nitration and Anammox in a biofilm system with suspended plastic carriers kept in suspension by aeration or mixing (Ling, 2008). Nitrite is produced

³⁷ Figures (a) and (b) adapted from (Wett, B., 2007)

in the outer aerobic layer, and in the inner layer nitrogen is produced under anoxic conditions. At a certain point the further growth of the biofilm will no longer enhance performance in the process because diffusion into the thickening biofilm will be a limiting factor. The accumulation of biofilm on the carrier is controlled through mixing and aeration.

The DeAmmon process with a moving bed system is in use at the Himmerfjärden WWTP, near Stockholm, Sweden (Ling, 2008), and in Hattingen WWTP, Germany (Rosenwinkel, Cornelius, & Thole, 2005).

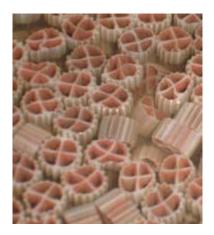


Figure 36: The DeAmmon[®] Process carrier material (KALDNES)³⁸

(3) Completely Autotrophic Nitrogen removal Over Nitrite (CANON) is a single-reactor suspended sludge system which relies on the interaction of autotrophic AOB and Anammox under low dissolved oxygen to perform two sequential reactions – nitritation then Anammox – simultaneously. The CANON process is completely autotrophic, making use of nitrosifying (AOB) bacteria and Anammox & denitritation bacteria under oxygen-limited control to achieve high levels of nitrogen removal in a single reactor. The autotrophic process consumes 63% less oxygen and 100% less reducing agent (COD) than conventional nitrogen removal systems.

Nitrite produced by aerobic ammonia oxidizers, such as *Nitrosomonas* and *Nitrosospira* can be subsequently be used by plactomycete-like anaerobic ammonia oxidizers, the Anammox bacteria. The molar ratio of NO₃-N(produced):NH₄-N(consumed) was about 0.13 (Third, Sleikers, Kuene, & Jetten, 2001). Dense biomass granular flocs create physical and biotic gradients allowing for coexistence of different groups of microorganism, and the performance of microbial processes and populations with complimentary and/or opposed metabolic requirements. The *Nitrosomonas* bacteria are limited by the oxygen supply, and the Anammox bacteria depend on AOB activity for their nitrite supply (Third, Sleikers, Kuene, & Jetten, 2001).

³⁸ Adapted from (Wett, B., 2007)

CANON has the ability to withstand ammonium limitation for up to one month without irreversible damage. It is therefore a feasible industrial system to remove ammonium from wastewater with very low organic load and variable nitrogen loading (Third, Sleikers, Kuene, & Jetten, 2001).

- (4) Oxygen-Limited Autotrophic Nitrification Denitrification (OLAND) is a biofilm process such as rotating biological contactor or sequencing batch reactor. OLAND uses aerobic ammonium oxidizing bacteria (AerAOB) to oxidize about half the ammonium to nitrite in the aerobic outer layers of the biomass (partial nitritation) while Anammox (AnAOB) convert the nitrite and remaining ammonium to nitrogen (89%) and nitrate (11%) in the anoxic inner layer (Clippeleir, Yan, Verstraete, & Vlaeminck, 2011).
- (5) By definition, hybrid reactors are those that defy a simply classification since they may make use of more than one mechanism for nitrogen removal. One type of hybrid reactors for anaerobic wastewater treatment combines the approach of both sludge blanket and fixed-bed reactors by developing Anammox bacteria in both matrix-free and support matrix regions. Biofilm growth on the support matrix and the suspended sludge retained in the volume occupied by the matrix treat the wastewater through the activity of the biomass. The hybrid reactor has not yet been implemented at full-scale. In future studies, the application of this hybrid reactor to CANON process will be explored (Ma, Hira, Li, Chen, & Furukawa, 2011).

Another type of hybrid reactor design is the "Universal SBR" concept, which is designed with flexibility to operate in different modes, allowing the plant operators to gain experience with the system and move on to more advanced modes of operation that require tighter modes of process control. The more advanced modes require more monitoring, they present opportunities for savings in chemical and electrical power. It also allows the operators to fall back to operate the system in a conventional mode in case of upset (Johnson, Sanjines, Castaneda, & Daigger, 2011).

5.9 Greenhouse Gas Emissions from Wastewater Processes

Next to energy- and cost-efficiency, the CO₂ footprint of a wastewater treatment plant is evolving into one of the benchmarks for sustainability. Greenhouse gas emissions from direct and indirect sources can be reduced through increased treatment efficiency using novel microbial pathways such as nitritation, denitritation and Anammox.

By comparison, I kg CH_4 has a global warming potential of 25 kg CO_2 on a 100-year timeline, whereas 1 kg N_2O has a potential of 298 kg CO_2 (Desloover, et al., 2011). The production of these gases during wastewater treatment may be more than previously thought.

Direct sources of emission in the wastewater treatment process are varied. Nitrogen gas (N_2) is produced by complete denitrification and the Anammox reaction. Incomplete denitrification results in

nitrous oxide emission (N₂O). Also, methane is released by methanogenic microorganisms from anaerobic processes (e.g. anaerobic digestion, denitrification, Anammox).

Biological methane production is well understood and the direct impact of methane emissions has been accounted for. Methane emissions observed from the nitrification or nitritation stages are likely derived from stripping of residual dissolved methane from the anaerobic digester. In an anaerobic reactor such as the Anammox process, observed methane emissions are likely derived from in-situ production through anaerobic methanogenesis within the Anammox reactor.

However, N₂O emissions are more difficult to establish and rely on the interplay of many parameters that determine N₂O production from nitritation, nitrification, and denitrification. Incomplete denitrification will produce higher greenhouse gas emissions than conventional activated sludge treatment for BOD removal. A well-performing denitrification process will reduce the total greenhouse gas emissions from the process compared with nitrification-only. Further, the addition of the Anammox side-stream process will further reduce the total greenhouse gas emissions from the plant since N₂O is a byproduct of denitrification rather than the Anammox process.

Therefore the impact of variability in nitrification and denitrification performance on N_2O emissions should be considered in more detail. It remains to be determined what modifications to the plant must be made to render the wastewater treatment process carbon dioxide emission neutral, for example, through the use of partial nitritation to reduce N_2O emissions by 50%.

There are also indirect emissions from various sources. Greenhouse gas emissions are attributed to the production and delivery of chemical additives like methanol. Methanol used for heterotrophic denitrification in the mainstream produces 1.16 moles of CO₂ for every mole of methanol produced. Further, by reducing the blower air usage, the greenhouse gas emissions due to power production are reduced.

5.10 Lessons Learned From Implementation

The preceding sections have demonstrated that not only is the Anammox process feasible for treatment for the removal of nitrogen from wastewater, it has been successfully applied at full-scale in several instances. Experiences gleaned from the literature were noted as "lessons learned." Studies done at labscale and full-scale were considered, as were the issues commonly experienced during start up and commissioning of the Anammox bioreactor.

A listing of lab-scale reactors described in the literature is compiled in Table 6. A partial listing of fullscale installations that make use of the Anammox process for nitrogen removal is provided in Table 7. Generally, the conversion for these reactors is limited by the availability of the substrate nitrogen rather than an inherent limitation in the bioreactor design. Substrate-limited reactors are over-designed (by definition), so it is not possible to directly compare the bioreactor types based on the observed conversion rates, but the listing provides an overview of the state of the technology's deployment.

Table 6: Lab-scale Anammox reactor p	erformance rates
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Location	Ref	Goal	Stage 1	Stage 2 - Anammox	WW Source	Performance (kg N/m ³ day)	Size	Year
Kumamoto University, Japan	(Yang, Zhang, Hira, Fukuzaki, & Furukawa, 2011)	Ambient temperature Anammox process	None	Spiral gas-solid separator	Synthetic	17.5	7.0 L	2011
Kumamoto University, Japan	(Zhang, Yang, Fujii, Hira, Zhang, & Furukawa, 2011)	Detection using DNA method	None	Up-flow fixed bed Anammox reactor with polyethylene sponge + granules	Digester supernatant	7.6	22 L	2011
Kumamoto University, Japan	(Zhang, Yang, Hira, Fujii, & Furukawa, 2011)	Partial nitritation as pretreatment before Anammox	Swim-bed partial nitritation reactor	Swim-bed reactor with biofringe material	Sludge water	3.1	5 L	2011
Kumamoto University, Japan	(Ma, Hira, Li, Chen, & Furukawa, 2011)	Hybrid reactor	None	Hybrid fluidized bed and fixed bed	Synthetic	20	6 L	2011
Kumamoto University, Japan	(Li, Ma, Hira, Fujii, & Furukawa, 2011)	-	Partial nitritation (biomass carrier)	Anammox (STAR)	Municipal		3L + 1.4 L	2011
Hokkaido University, Japan	(Tsushima, Ogasawara, Kindaichi, Satoh, & Okabe, 2007)	High-rate Anammox biofilm reactors	None	Anammox (Upflow fixed bed biofilm column reactors)	Synthetic	26	-	2007
Zhejiang University, Hangzhou, China	(Liang, Shangyuan, Liang, Du, Liu, & Yang, 2011)	Biofilm nitritation reactor	Partial nitritation (biofilm)	Anammox	Synthetic	-	-	2011

Table 7: Full-scale Anammox reactor performance rates

Location	Ref	Goal	Stage 1	Stage 2	WW Source	Performance (kg N/m ³ day)	Size	Year
Japan	(Tokutomi, Yamauchi, Nishimura, Yoda, & Abma, 2011)	Retrofit of existing nitrification denitrification plant	Nitritation (moving bed))	Anammox (upflow anaerobic sludge blanket (UASB)	Semi- conductor factory	1.04-3.29	30 m ³ (nitritation) + 60 m ³ (Anammox) + 30 m ³ (reaeration)	2011
Himmerfjärden, Sweden	(Ling, 2008)	Commissioning of side-stream reactor	DeAmmon process (biofil series in two pa -		-	-	1400 m3	2008
Bergeb op Zoom (NL)	(Desloover, et al., 2011)	New Activated Sludge retrofit of existing plant	Partial nitritation (floc)	Anammox (floc) followed by Nitrification/ Denitrification	Potato	0.5	2370 (Nitritation) + 1650 (Anammox) + 1600 (Denitrification) + 2300 (Nitrification)	2011
Hattingen (DE)	(Rosenwinkel, Cornelius, & Thole, 2005)	-	۔ Deammonification (KAL Reacto		Sludge water	0.16 (160 kg/L day)	319 m ³ (five basins)	2005
Rotterdam (NL)	(Abma, et al., 2007).	Retrofit of existing SHARON- Anammox reactor to granular sludge reactor	SHARON	Granular sludge reactor	Sludge water	10	-	2007
Strass, Zürich, St. Gallen, Niederglatt, Sweden	(Joss, et al., 2009)	Control strategy	Partial nitration and Anammox in one SBR		Digester supernatant	0.5	1400 m ³ (Zürich), 300 m ³ (St. Gallen) 150 m3 (Niderglatt)	2009
Pakasoy, Turkey	(Ozdemir, Mertoglu, Yapsakli, Aliyazicioglu, Saatci, & Yenigun, 2011)	Membrane bioreactor	Microfiltration modules (10-20,000 mg/L MLSS)	Sludge water	-	3	100 m ³ /day	2011
Mechernich/Germany, Kolliken/Switzerland, Pitsea/UK	(Hippen, Helmer, Kunst, Rosenwinkel, & Seyfried, 2001).	Leachate treatment with RBC Reactor	Rotating contactors for ni removal by Anammox Deammonif	in biofilm (aerobic	Leachate	-	5225 m ² (Mechernich), 2090m ² (Kolliken)	2001
Sluisjesdijk, Rotterdam, The Netherlands	(Van der Star, et al., 2007)	Startup of first Anammox reactor in Rotterdam	SHARON nitritation reactor	Granular sludge reactor	Sludge water	7.1	70 m3	2007
Lichtenvoorde, The Netherlands	(Van der Star, et al., 2007)	-	-	Granular sludge reactor	-	1	100	2007

Location	Ref	Goal	Stage 1	Stage 2	WW Source	Performance (kg N/m ³ day)	Size	Year
Hattingen (DE)	(Van der Star, et al., 2007)	-	-	Moving bed	Industrial	0.1	67	2007
Stockholm (SE)	(Van der Star, et al., 2007)	-	-	Moving bed	Municipal	2	2	2007
Zürich (CH)	(Van der Star, et al., 2007)	-	-	SBR	Municipal	1.2	2.5	2007
Olburgen (NL)	(Van der Star, et al., 2007)	-	-	Bubble Column	Municipal	0.6	600	2007
Strass (AT)	(Van der Star, et al., 2007)	-	One-reactor nitration	-Anammox SBR	Municipal	0.6	500	2007
Glanerland (CH)	(Van der Star, et al., 2007)	-	One-reactor nitration-Anammox SBR		Municipal	0.4	400	2007
Pitsea (GB)	(Van der Star, et al., 2007)	-	One-reactor nitration	-Anammox RBC	Industrial	1.7	240	2007
Hattingen (DE)	(Van der Star, et al., 2007)	-	One-reactor nitration-An	ammox moving bed	Municipal	1	102	2007
Mechernich (DE)	(Van der Star, et al., 2007)	-	One-reactor nitration	-Anammox RBC	Municipal	0.6	80	2007
Kollikon (CH)	(Van der Star, et al., 2007)	-	One-reactor nitration	-Anammox RBC	Municipal	0.4	33	2007
Stockholm (SE)	(Van der Star, et al., 2007)	-	One-reactor nitration-An	ammox moving bed	Municipal	0.5	4	2007
Upper Occoquan, North Virginia, USA	(Johnson, Sanjines, Castaneda, & Daigger, 2011)	-	DEMON ®	Sludge water	Municipal	-	-	2018
Alexandria, North Virginia, USA	(Johnson, Sanjines, Castaneda, & Daigger, 2011	-	DEMON ®	Sludge water	Municipal	-	-	2013

5.10.1 Considerations on Start-Up of the Anammox Reactor

Drawing from the lessons of experiences reported in literature, some of the "teething problems" associated with the start-up of a new technology can be anticipated. For example, the Strass plant in Austria took more than 3 years to scale-up from pilot through full-scale (Van der Star, et al., 2007). Since the Strass and Rotterdam plants were amongst the first to start up, the lessons learned from these startups have undoubtedly shortened the startup time for Anammox reactors built since then.

At the time of start-up, the reactor is typically fed stepwise with synthetic feed water consisting of the nitrites and ammonia. The synthetic feed to used to prevent nitrite overloading the reactor at start-up (Tokutomi, Yamauchi, Nishimura, Yoda, & Abma, 2011). At first, the flow regime is characterized with high influent flow rate and low nitrite concentration. The concentration of nitrite during the startup is a key factor for growth – too little will result in slow growth due to substrate limitation, and too much (above 50-150 mg-N/L) can lead to inhibition (Van der Star, et al., 2007).

Specifics of the start-up strategy will depend to a large extent on whether the system is designed for two-stage or single-stage operation. The precondition for Anammox activity is the stable supply of nitrite and ammonia. It is therefore possible to attain high rates of ammonia removal during the start up of the Anammox reactor by a nitritation/nitrification process before the Anammox process gains sufficient biomass and begins to consume nitrite and ammonia simultaneously.

By analyzing the biomass growth and Anammox activity, the startup period can be separated into two phases: one in which there is biomass growth but no detectable activity, and the period from which there is a notable increase in ammonium conversion and nitrate production. During the start-up phase, the simultaneous removal of ammonia and nitrite indicates the occurrence of the Anammox reaction. The day on which the first evidence of Anammox activity is noted may vary, having been reported in range of 40 days, with the characteristic red colour of Anammox biomass appearing after 60 days, with increasing total nitrogen removal rate (NRR) as the culture matures, barring issues during startup (Tsushima, Ogasawara, Kindaichi, Satoh, & Okabe, 2007).

The start-up phase at the Himmerfjärden WWTP in Sweden occurred in two phases. During the startup of the nitritation reactor, intermittent aeration was used to control the nitrite oxidizing bacteria. To start up the denitritation reactor, a heterotrophic biofilm was created with a dosage of external carbon operated at first with continuous aeration and then later without any air for denitritation (Ling, 2008).

To apply the process at a large scale, it is important to know the limitations of the system. At full-scale, the Anammox reactor is initially inoculated with nitrifying sludge. The lab-scale reactor (10-100 L) is useful for both cultivation and enrichment of Anammox bacteria sludge to be used as the inoculum for full-scale start-up. Once a stable running state is achieved for Partial Nitritation, Anammox seed culture sludge from an Anammox enrichment reactor can be added in portions to start up the Anammox process.

To save time and resources, Anammox reactors have been scaled up from lab scale to full-scale. It was found that the costs to operate a pilot plant (i.e. personnel) were approximately the same as when running a full-scale pilot. (Van der Star, et al., 2007).

i Seed Culture

Anammox bacteria are found in most activated sludges, but in very small proportions. In the absence of an existing seed culture, the start-up of a full-scale reactor with a microbial process that is known for its long growth period can be a difficult task. However, the Anammox process has been successfully started at full scale at numerous plants. An enrichment culture of sludge containing a high quantity of Anammox bacteria increases the chances of success for the start-up of a full-scale anammxox reactor:

- <u>Selection</u>: The activated sludge from a number of different treatment plants can be obtained to find the one best suited to the intended wastewater application. For example, of the 11 Anammox bacteria samples obtained different treatment plant sludges, the one with the highest activity was selected (Tsushima, Ogasawara, Kindaichi, Satoh, & Okabe, 2007).
- <u>Enrichment</u>: Once the appropriate seed culture is selected, the culture is enriched at lab-scale to produce a volume of Anammox sludge for startup of the full-scale reactor, as in Figure 37. The length of time to successfully enrich an Anammox culture varies, with some researchers having reported enrichment culture ages in terms of years (Tokutomi, Yamauchi, Nishimura, Yoda, & Abma, 2011).
- <u>Inoculation</u>: The enrichment culture is used to inoculate the full-scale reactor with the effluent biomass. The volume of inoculum required depends on the volume of the receiving reactor.



Figure 37: Enrichment culture of Anammox³⁹

³⁹ From <u>http://en.wikipedia.org/wiki/Anammox</u>

ii Commissioning Issues

From a review of experiences with commissioning of full-scale system in the literature, issues commonly reported to occur during the start up and commissioning of the reactor included:

- Washout due to sudden changes in the hydraulic regime, such as a sudden increase in upstream velocity due to shock loading.
- Toxicity, such as breakthrough of toxic methanol from nitritation reactor (methanol slip) into the Anammox reactor. Another source reported was toxic shock from chemical toilet discharges into sludge line strongly inhibits nitrification and adversely affect the Anammox process. Incidental nitrite toxicity may be caused by too high ammonium loading rate and occurs when NAR is higher than 1.0-1.3.
- Mechanical failures, such as freezing of pipes and other equipment failures. In cold climates, the freezing of lines may lead to stoppage of feed lines, such as the recycle stream. Incidental failure of pumps and blowers for air delivery may impede mixing and loading.

6 Plant Design Concepts for Side-stream Treatment Using

Anammox

Plant design concepts for the removal of nitrogen compounds from wastewater are presented in this section in order to illustrate the ideas developed in the preceding sections. The plant design concepts for nitrogen removal were developed in two phases, sizing and simulation.

Simplified models based on Monod-type growth kinetics were used to size the activated sludge reactors required for the removal of carbon and nitrogen. A baseline scenario for activated sludge treatment for BOD-only removal only was developed first. In the subsequent scenarios, treatment steps were added for the removal of ammonia and nitrogen salts from the main treatment stream, with a side-stream treatment process. Key process equipment, and energy and chemical requirements are also discussed.

In the simulation phase, the effect of the various reactor configurations was investigated on the overall plant. The goal of the simulation effort was to investigate the impacts on (1) Total Nitrogen Removal from the plant effluent, (2) Operating Cost, including chemical and energy costs, and (3) Greenhouse Gas Emissions from direct and indirect sources.

6.1 Facility Basis / Assumptions

A wastewater treatment plant with average daily flows of 656 ML/d serving a predominantly residential and commercial area with a population of over 3 million is considering upgrades to nitrogen removal technology with side-stream treatment using the Anammox process.

The existing plant's liquid processing train includes primary treatment and conventional activated sludge treatment with diffused coarse-bubble aeration. The solid processing train includes waste activated sludge (WAS) thickening and anaerobic digestion, followed by dewatering of the digested sludge with centrifuges. The influent flow and loadings are presented in Table 8. For this system, centrate recycled to the primary treatment process accounts for 20% of the influent ammonia load. Therefore, it meets the recommendation of at least 10-15% of the influent ammonia load to consider side-stream treatment.

	Raw Sewage Centrate		trate		Raw Sewage + Centrate		
Design flow, Q (m3/day)	65	6,000	4,523		% Influent	660	,523
	mg/L	kg/day	mg/L	kg/day		mg/L	kg/day
TSS	270	177,121	4,099	18,540	10%	296.2	195,661
cBOD5	250	164,001	1,786	8,079	5%	260.5	172,080
COD	535	351,217	3,914	17,705	5%	558.5	368,922
TKN (as N)	35.0	22,960	1,123.6	5,082	22%	42.5	28,042
NH3 (as N)	22.0	14,432	887.2	4,013	28%	27.9	18,445
Alkalinity (as CaCO3)	225	147,600		0	0%		0
Sustained Temperature	1	4°C	35°C			14°C	
COD/N Ratio	9.4		1.1			8.3	
BOD/COD Ratio	0.42		0.29			0.42	
Nitrogen Loading Rate (kg-N/m3)	C).04	1.1	12		0.04	

Some further assumptions:

- The typical coldest water temperature is 14°C. (Typical coldest temperature in Ontario is 10°C);
- TSS is approximately equal to cBOD;
- Primary clarifiers reduce influent BOD by 30%
- The side-stream is recycled to the headhouse
- Fractionation of Influent: BOD:COD=0.42, VSS=70% of TSS;
- Primary treatment efficiency is 60% removal for TSS and 35% removal for BOD;
- The average blower performance is assumed to be 0.018 kWh per m³ air consumed;
- The target MLSS for operation of the activated sludge process is 1,750 mg/L.
- Dewatering operates 24 hours per day, 7 days a week;

- Power consumption for RAS and recycle pumps not considered;
- Sludge production and wasting rates for the side-stream reactors were not considered;
- Coarse Bubble SOTE = 0.5%/ft, Fine Bubble SOTE = 2.0%/ft (fouling rate of 0.9 / no fouling effects); and
- Depth of the reactor basin is 4.5 m and the diffusers are mounted 0.5 m from the bottom of the basin.
- Activated Sludge concentration of 5,120 mg/L (range 6,000 to 12,000 mg/L).

6.2 Treatment Configurations

A simplified complete-mix activated sludge system using microbial kinetics to model different treatment plant configurations using reactors in series to obtain preliminary sizing data such as reactor volumes and air requirements.

The base case is an activated sludge treatment system for carbonaceous BOD removal only using coarse bubble diffusers. Design examples are provided for the base case with coarse bubble diffusers to demonstrate the relative impact on blower air usage. The remainder of the design examples use fine bubble diffusers:

- (1) BOD-removal only;
- (2) BOD-removal and nitrification for ammonia removal;
- (3) Conventional Biological Nitrification / Denitrification;
 - (a) preanoxic
 - (b) postanoxic
- (4) Biological Nitrification / Denitrification with Side-stream treatment, using:
 - (a) nitrification/denitrification;
 - (b) nitritation/denitritation;
 - (c) partial nitritation/Anammox.

Following the design concepts, a set of total-plant simulations was developed using CHEApet in which the scenarios are compared in terms of (1) Total Nitrogen Removal, (2) Operating Cost, and (3) Greenhouse Gas Emissions.

6.3 Preliminary Sizing Calculations

The design approach was to determine the required volumes by selecting the solids residence time to provide a target effluent ammonia and nitrate concentration. Preliminary calculations were developed in order to estimate the reactor volumes for the mainstream nitrification system and the side-stream treatment reactor. In addition, the supplemental carbon needed and the aeration requirements in order to adequately size the system were developed as a basis of process comparison. The calculations are

based on the design outlined in "Wastewater Engineering: Treatment & Reuse", Fourth Edition, Metcalf & Eddy Inc.

The wastewater fractions required for design are presented in Table 9 and were estimated from the equations provided in Section 5.1 Wastewater Characteristics.

Parameter	mg/L	Notes
COD	558.1	
bCOD	297.7	Biodegradable COD = 1.6 x cBOD
nbCOD	260.5	Non-biodegradable COD = COD - bCOD
sCOD	251.2	Soluble COD = 45% COD
cBOD	260.5	
scBOD	143.3	Soluble cBOD = 55% cBOD
sCODe	21.9	Non-biodegradable COD in effluent = sCOD - 1.6 x scBOD
bpCOD/pCOD	0.61	
nbVSS	80.7	Non-biodegradable VSS = (1-bpCOD/pCOD)x VSS
iTSS	53.3	Inert TSS = TSS - VSS

Table 9: Wastewaster fractions

6.4 Aerobic Activated Sludge Main Process Design

The design for the aerobic activated sludge process of the mainstream was conducted as a "desktop design" exercise for preliminary sizing calculations of 3 different complete mix activated sludge (CMAS) systems:

- (1) Activated Sludge Treatment with BOD-removal only (coarse- and fine- bubble)
- (2) Activated Sludge Treatment with BOD- and nitrogen removal (coarse- and fine bubble)
- (3) Activated Sludge Treatment with BOD- and nitrogen removal (fine bubble) with pretreatment of the recycle stream

The model was based on Monod-type growth rate kinetics rather than substrate utilization. Kinetic parameters used as the design basis for the subsequent sections are presented in Table 10. These parameters are used to develop reactor volumes for nitrifier and denitrifier activity at the design temperature of 14°C (temperature of the main stream reactor). These preliminary sizing calculations are used to support the simulations presented later in this section.

The secondary clarifier design is similar for the three options presented. From Equation 25, where X_s is the sludge concentration of 5,120 mg/L and MLSS is 1,750 mg/L, the sludge recycle ratio (Metcalf & Eddy, 2004) was found to be 0.52:

Equation 25: Return Activated Sludge Ratio

$$R = \frac{X_{MLSS}}{X_S - X_{MLSS}}$$

For an assumed clarifier overflow rate of 22 m^3/m^2 -d, the required area for the clarifiers was 29,818 m^2 . If one clarifier is provided for each of the 11 aeration reactors, there would be 11 clarifiers of 2,711 m^2 each.

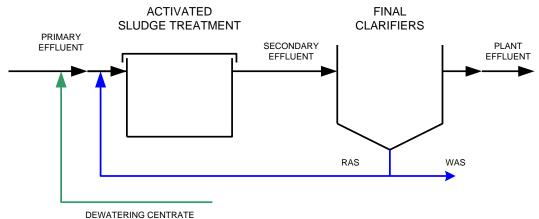




Figure 38: PFD for Carbonaceous BOD Removal and Nitrification

Design goals for this stage were to determine:

- Effluent concentration of BOD and Total Nitrogen;
- Biomass residence, mixed liquor concentration, and aerobic reactor volume;
- Oxygen requirements, blower air usage, power consumption; and
- Ammonia removal, Nitrate production (NO_x).

The maximum growth rate of the nitrifiers is established first. The design SRT for nitrification to occur is a function of the specific growth rate of the nitrifying bacteria using kinetic theory. Cell yield and specific growth rates, μ (new cells/cells-time) for heterotrophs, nitrifiers and denitrifiers are given in Table 2. The growth rate of nitrifiers from Equation 9 (p.32), with the assumed conditions and values for kinetic coefficients $\mu_{n,m,14oc}$ =0.60 g/g-d; $K_{n,14oc}$ =0.513 mg/L; $k_{dn, 14oc}$ = 0.095 g/g-d; with effluent TKN = 10 mg/L and DO = 2.0 mg/L;

$$\mu_n = \mu_{n,max} \left(\frac{N}{K_n + N}\right) \left(\frac{O_D}{K_o + O_D}\right) - k_{dn} = 0.60 \left(\frac{10}{0.513 + 10}\right) \left(\frac{2.0}{0.50 + 2.0}\right) - 0.06 = 0.36 \text{ g/g-d}$$

The theoretical SRT for nitrifier growth was found from Equation 10 (p.33),

$$\theta_c = \frac{1}{\mu_n} = \frac{1}{0.36} = 2.77 \text{ days}$$

Considering a peaking factor of 2.5 for TKN, the design SRT for nitrification in the main plant stream is **6.92 days.**

	Coefficient	Unit	Range at 20 °C	Typical Value	θ values ⁴¹	Typical Value	Adjusted Value @ 14°C
	μ_{m}	g VSS/g VSS-d	3.0-13.2	6.0	1.03-1.08	1.07	4.0
	Ks	g bCOD/m3	5.0-40.0	20.0	1.03-1.08	1.04	20
Heterotrophs	Y	g VSS/g bCOD	0.30-0.50	0.40	-	-	-
	k _d	g VSS/g VSS-d	0.40-0.60	0.12	1.00	1.00	0.095
	f _d	n/a	0.08-0.20	0.15	-	-	-
	μ _{mn}	g VSS/g VSS-d	0.20-0.90	0.9	1.06-1.123	1.07	0.60
	Kn	g NH4-N/m3	0.5-1.0	0.7	1.03-1.123	1.053	0.513
Nitrifiers	Y _n	g BSS/g NH4-N	0.10-0.15	0.12	-	-	-
	k _{dn}	g VSS/g VSS-d	0.05-0.15	0.08	1.03-1.08	1.04	0.095
	Ko	mg/L	0.40-0.60	0.50	-	-	-
	Y	g VSS/g bCOD used	-	0.30	-	-	-
Denitrifiers @ 15°C	k _d	g VSS/g VSS-d	-	0.05	-	-	-
13 C	к	g VSS/g VSS-d	-	6.7	-	-	-
	Ks	mg/L	-	10.9	-	-	-

Table 10: Activated sludge kinetic coefficients for heterotrophic bacteria and nitrifiers at 14°C⁴⁰

where μ is the maximum specific bacteria growth grate; K is the half-velocity degradation coefficient; Y is yield; k_d is the endogenous decay coefficient; f_d is fraction of biomass that remains as cell debris

6.4.1 Complete-Mix Activated Sludge Process for BOD removal only

As a first step, a complete-mix activated sludge process (CMAS) for carbonaceous BOD removal only was designed for the influent loading noted in Table 8 (p.73). The plant effluent concentration is required to meet BOD and TSS of less than 15 mg/L, with no requirement for ammonia or nitrate removal.

i Reactor Sizing – BOD-removal only

To size the reactor, the daily biomass production and total biomass expected in the basin was found using the effluent substrate concentration. A solids residence time (SRT) of 3 was selected to suppress the growth of nitrifiers for BOD removal because it is lower than the minimum SRT required for nitrifier growth, so only BOD is removed.

Biomass Retention in Aerobic Reactor:

• The influent bCOD concentration (S_o) was given in Table 9 (p.75). The effluent substrate concentration (bCOD) was calculated using Equation 12 (p.35), based on the selected SRT and

⁴⁰ Adapted from (Metcalf & Eddy, 2004), (Reynolds & Richards)

⁴¹ Correction for temperature $\mu_{m,T} = \mu_m \theta^{T-20}$; $k_{d,T} = k_2 \theta^{T-20}$

kinetic parameters for heterotrophs given in Table 10 (where $K_s = 5 \text{ mg bCOD/L}$, $\mu_m = 2.13 \text{ g/g-d} k_d = 0.49 \text{ g/g-d}$):

$$S = \frac{K_{S}[1+(k_{d})\theta_{c}]}{\theta_{c}(\mu_{m}-k_{d})-1} = \frac{20[1+(0.49)3]}{3(2.13-0.49)-1} = 3.14 \text{ mg bCOD/L}$$

• With the influent and effluent bCOD, the biomass in the reactor basins was calculated by following the mass balance approach. A prediction of biomass production and sludge production was made using Equation 11 (p.35), considering heterotrophic biomass (A), cell debris from endogenous decay (B), with no nitrification (C) and neglecting the non-bidedegradable VSS (D) in the influent, where Y = 0.40 VSS/g bCOD; S_o = 280 mg bCOD/L:

$$P_{X,BIO} = \frac{QY(S_o - S)\left(\frac{1kg}{10^3g}\right)}{1 + (k_d)\theta_c} + \frac{(f_d)k_d QY(S_o - S)\theta_c\left(\frac{1kg}{10^3g}\right)}{1 + (k_d)\theta_c};$$
$$P_{X,BIO} = 31,503 + 6,946 = 38,449 \ kg \ VSS/d$$

- The amount of biomass retained in the reactor was found from the daily mass loading and the solids residence time (SRT). The mass of the MLVSS and MLSS in the aeration basins was found calculated as follows:
 - The total mass of VSS in the reactor includes VSS from biomass production, nonbiodegradable VSS entering the reactor from the primary clarifier effluent.

$$\begin{split} P_{X,VSS} &= P_{X,BIO} + Q(X_{nbVSS}) = 38,449 + 660,523 \text{ x (81)}; \\ P_{X,VSS} &= 36,202 + 53,273 = 91,723 \text{ kg/day}; \\ (X_{VSS})(V) &= (P_{X,VSS})\text{SRT} = (91,723 \text{ kg/day})^*(3 \text{ day}) = 275,168 \text{ kg as MLVSS} \end{split}$$

The total mass of solids in the rector included TSS associated with the biomass (e.g. 70% biomass VSS and 30% inorganic solids), non-biodegradable VSS, and inert TSS entering the reactor from the from the primary clarifier effluent.

$$P_{X,TSS} = \frac{P_{X,BIO}}{V} + Q(X_{nbVSS}) + Q(X_{TSS,o} - X_{VSS,o}) = 38,449/0.70 + 660,523 \times (81 + 53);$$

$$P_{X,TSS} = 54,927 + 53,273 + 35,219 = 143,420 \text{ kg/day};$$

$$(X_{TSS})(V) = (P_{X,TSS})SRT = (143,420 \text{ kg/day})^*(3 \text{ day}) = 430,260 \text{ kg as TSS in the reactor}$$

Aeration Basin Volume and HRT:

 Select the design MLSS mass concentration and determine the aeration basin volume and detention time using the TSS mass computed. At MLSS = 1,750 mg/L:

• The aeration basin residence time, 11 basins are used at 22,349 m³ each. The required residence time is:

V/Q = (245,835/660,523) x24 hr/day= 8.93 hours

Reactor loading characteristics:

- The food-to-microorganism ratio (F/M_b) ratio was calculated to be 0.72 g/g-d.
- The BOD loading is 0.80 kg/m³-d.
- The observed yield was 0.78 g TSS/g bCOD, or 0.55 g VSS/g bCOD.

ii Process Inputs-BOD-removal only

Oxygen demand:

• The actual oxygenation requirements for carbonaceous BOD removal was calculated using Equation 15 (p.36):

$$R = Q(S_o - S) - 1.42P_{x,bio}$$
= 5,830 kg O₂/hr

• The actual oxygen required for the aeration process was found from Equation 16 (p.37). The density of air at 14°C and pressure of 101.3 kPa. The corresponding amount of oxygen by weight is 1.23 kg/m³. With the values of the parameters for air density, obtained from literature, the oxygen demand was found:

$$SOTR = R\left(\frac{C_{s,20}}{(\alpha)(F)(\beta C_{s,T,H}-C_L)}\right) (1.024^{20-T}) = 11,339 \text{ kg O}_2/\text{hr}$$

• The air flowrate was found as follows, where *E* is the diffuser efficiency and ρ_{air} is the density of air:

Air flowrate,
$$m^3/min = \frac{\left(SOTR\frac{kg}{h}\right)}{E\left(60\frac{\min}{h}\right)\rho_{air}\left(kg\frac{O_2}{m^3}\right)}$$

For a fine-bubble configuration with 25% SOTE, the air flow rate is 2,492 m³/min compared with 9,734 m³/min for coarse-bubble configuration with 6.4% SOTE. If the average blower performance is assumed to be 0.018 kWh/m3 air, the daily power consumption to meet the calculated air demand was 69 MWh/day for the fine-bubble diffuser system, versus 269 MWh/day for the coarse-bubble diffuser system.

Chemical demand:

- In this scenario it is assumed that there is no nitrification occurring, so the alkalinity demand is zero.
- Similarly, there is no denitrification occurring so the methanol demand is zero.

Sludge production:

• The daily sludge production was found in order to design the secondary clarifiers. Based on the sludge recycle ratio of 0.52, a wasting rate of 143,420 kg/day as WAS would result.

6.4.2 Complete-Mix Activated Sludge Process for BOD removal with nitrification

The complete-mix activated sludge process (CMAS) designed in the previous section was modified for removal of both carbonaceous BOD and ammonia. In this example, the plant effluent now must meet an effluent limit of 10 mg/L NH₃-N in addition to the existing requirements for TSS and BOD removal.

i Reactor Sizing – BOD and Ammonia Removal

The nitrification rate will control the design because the nitrifying organisms grow more slowly than the heterotrophic organisms that remove organic carbon. The design SRT for nitrification was found to be \sim 7 days (see calculation on page 75). A similar approach was followed to the BOD-removal only design, with nitrification

Biomass Retention in Reactor:

• The effluent substrate concentration (bCOD) was calculated using Equation 12 (p.35):

$$S = \frac{K_s[1+(k_d)\theta_c]}{\theta_c(\mu_m - k_d) - 1} = \frac{20[1+(0.49)3]}{7(2.13 - 0.49) - 1} = 2.12 \text{ mg bCOD/L}$$

The biomass and sludge production was estimated using Equation 11 (p.35), considering heterotrophic biomass (A), cell debris from endogenous decay (B), with nitrification (C) and neglecting the non-bidedegradable VSS (D) in the influent, where Y = 0.40 VSS/g bCOD; S_o = 280 mg bCOD/L:

$$P_{X,BIO} = \frac{QY(S_o - S)(\frac{1kg}{10^3g})}{1 + (k_d)\theta_c} + \frac{(f_d)k_dQY(S_o - S)\theta_c(\frac{1kg}{10^3g})}{1 + (k_d)\theta_c} + \frac{QY_n(NO_x)(\frac{1kg}{10^3g})}{1 + (k_{dn})\theta_c}$$
$$P_{X,BIO} = 17,792 + 9,044 + 2,032 = 28,868 \ kg \ VSS/d$$

- The mass of the MLVSS and MLSS in the aeration basins was found calculated as follows:
 - The total mass of VSS in the reactor:

 $P_{X,VSS} = P_{X,BIO} + Q(nbVSS) = 28,868 + 660,523 \times (81);$ $P_{X,VSS} = 28,868 + 53,273 = 82,141 \text{ kg/day};$ $(X_{VSS})(V) = (P_{X,VSS}) \theta_c = (82,141 \text{ kg/day})^* (7 \text{ day}) = 897,189 \text{ kg as MLVSS}$

• The total mass of solids in the rector:

$$P_{X,TSS} = \frac{P_{X,BIO}}{V} + Q(X_{nbVSS}) + Q(X_{TSS,o} - X_{VSS,o}) = 28,868/0.70 + 660,523 \times (81 + 53);$$

 $P_{X,TSS} = 42,240 + 53,273 + 35,219 = 129,732 \text{ kg/day};$ (X_{TSS})(V) = (P_{X,TSS}) θ_c = (129,732 kg/day)*(7 day) =897,189 kg as TSS in the reactor

Aeration Basin Volume and HRT:

• Select the design MLSS mass concentration and determine the aeration basin volume and detention time using the TSS mass computed. At MLSS = 1,750 mg/L:

• The aeration basin residence time, 11 basins are used at 41,494 m³ each. The residence time is: $V/Q = (492,344/660,523) \times 24 hr/day = 18.6 hours$

Reactor loading characteristics:

- The food-to-microorganism ratio (F/M_b) ratio was calculated to be 0.35g/g-d.
- The BOD loading is 0.38 kg/m3-d.
- The observed yield was 0.71 g TSS/g bCOD, or 0.49 g TSS/g bCOD.

ii Process Inputs- BOD and Ammonia Removal

Oxygen demand:

• The amount of nitrogen oxidized to nitrate is found by performing a nitrogen balance, Equation 13 (p.35):

$$(N_{OX}) = (N_{TKN,o}) - (N_e) - 0.12 \frac{P_{x,bio}}{o};$$

$$(N_{OX}) = (42) - (10) - 0.12 \frac{28,868 \times 1000 g/kg}{660,523} = 27.2 \text{ mg/L as NO3-N}$$

• The actual oxygenation requirements for carbonaceous BOD removal was calculated using Equation 15 (p.36):

 $R = Q(S_o - S) - 1.42P_{x,bio} + 4.33Q(N_{OX}) = 9,668 \text{ kg O}_2/\text{hr}$

• The actual oxygen required for the aeration process was found from Equation 16 (p.37):

$$SOTR = R\left(\frac{C_{s,20}}{(\alpha)(F)(\beta C_{s,T,H}-C_L)}\right) (1.024^{20-T})$$
= 21,153 kg O₂/hr

• For a fine-bubble configuration with 25% SOTE, the air flow rate is 4,948 m³/min compared with 19,328 m³/min for coarse-bubble configuration with 6.4% SOTE. The daily power consumption

to meet the calculated air demand was **128 MWh/day** for the fine-bubble diffuser system, versus **501 MWh/day** for the coarse-bubble diffuser system.

Chemical demand:

- Alkalinity required for nitrification is 7.14 x 27.5 g NO3-N/m³ = 196.3 mg/L as CaCO3, resulting in a residual alkalinity concentration of 28.7 mg/L as CaCO3. Alkalinity needed to maintain neutral pH is ~80 mg/L as CaCO₃. Therefore there is an alkalinity demand of 51.3 mg/L as CaCO₃ to ensure complete nitrification.
- There is no denitrification occurring so the methanol demand is zero.

Sludge production:

• The daily sludge production was found in order to design the secondary clarifiers. Based on the sludge recycle ratio of 0.52, a wasting rate of 129,732 kg/day as WAS would result.

6.4.3 Activated Sludge Process with Pretreatment of Recycle Stream

What would be the impact on the main liquid process stream if the nitrogen load from the recycle stream was removed in a previous step? This would immediately achieve a 20% reduction in ammonianitrogen, with an ultimate impact on the design for the nitrification and denitrification reactors.

In this example, an identical design process was followed for reduced nitrogen loading from the centrate stream, using a biological nitrification and preanoxic denitrification. Results are presented in Section 6.7 Summary of Reactor Design Concepts:

6.5 Anoxic Activated Sludge Main Process Design

Design goals for this stage are to determine:

- Effluent concentration of TN
- Biomass residence, mixed liquor concentration, Anoxic reactor volume
- Nitrate removal, total nitrogen removal
- Methanol requirements
- Mixing energy requirements

6.5.1 Suspended Growth Anoxic/Aerobic Denitrification Process

In this example, the plant effluent now must meet an additional limit of 2.9 mg/L NO_3 -N in addition to the existing requirements for TSS, BOD, and Ammonia removal. An anoxic denitrification system was added to the system previously described for the removal of nitrate produced by the nitrification reactions. A pre-anoxic configuration was provided as a pre-treatment step to the aerobic reactor, shown in Figure 39.

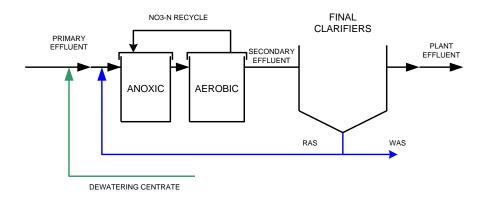


Figure 39: PFD for Preanoxic Nitrification/Denitrification with No Side-stream Treatment

Internal Recycle Rate:

• From the NOx concentration determined in the nitrification reactor design and the target effluent NO3-N concentration, the Internal Recycle (IR) ratio was found using Equation 17 (p.41):

$$IR = \frac{N_{OX}}{N_e} - 1.0 - R = \frac{27.2}{2.9} - 1.0 - 0.52 = 7.86$$

- At higher ratios, the influent COD is diluted more in the anoxic reactor by mixed liquor from the aerobic reactor, resulting in a lower denitrification rate.
- The amount of nitrate fed to the anoxic basin assumes that most of the nitrate fed to the anoxic zone will be reduced, although some residual concentration may remain. The influent TKN is assumed to be biodegradable. The mass balance approach yields:

Flowrate to anoxic basin = $IR \times Q + R \times Q = 5,537,052 \text{ m}^3/\text{d}$

Anoxic Basin Volume and HRT:

- To determine the optimal volume for the anoxic reactor, a trial-and-error procedure was used to estimate the detention time for the anoxic zone to minimize the excess nitrate removal capacity of the reactor.
- Using a detention time of **6.8 hours**, the anoxic volume was found:

- The anoxic reactor volume would be situated upstream of the aerobic reactors. The anoxic zone at the head of 11 basins would comprise a volume of 17,013 m³ each.
- The active biomass concentration in the MLSS was found using Equation 18 (p.42):

$$X_b = \left[\frac{Q(\theta_c)}{V}\right] \left[\frac{Y(S_O - S)}{1 + (k_d)\theta_c}\right] = 235 \text{ mg/l}$$

The selection of the anoxic volume affects the F/M_b ratio, which is used to determine the SDNR from standard curves in literature and the fraction of rbCOD to bCOD (assumed to be 51%). The F/M_b ratio is found based on the active biomass concentration from the aerobic nitrification reactor:

$$\frac{F}{M_b} = \frac{QS_O}{V_{nox}(X_b)} = 4.4 \text{ g/g-d}$$

• From the literature, the SDNRb was determined to be 0.47 g/g-d at 14°C, which is within the range of reported SNDR values (0.04 to 0.42 g/g-d). The amount of NO₃-N that can be reduced was found:.

N_{ox}= (0.47 g/g-D)(187,148 m³)(235 mg/L) = 21,180 kg/d

• Compared to 16,057 kg/d, there is about 32% excess nitrate removal capacity. Thus t=6.8 h is acceptable but a lower residence time can be used.

Oxygen Credit:

• The amount of oxygen supplied by nitrate reduction (oxygen credit) is:

$$Oxygen\ Credit = O_C = \left(\frac{2.86\ g\ O_2}{gNO_3 - N}\right)(N_{OX} - N_e)Q = 1,900\ \text{kg/hr}.$$

• The net actual oxygen required is:

Net O₂ = Oxygen Demand – Oxygen Credit Net O₂ = 9,668-1,900=**7,768 kg/hr**.

• The oxygen required can be reduced by 20% by the addition of extra volume to accommodate an anoxic zone for denitrification.

Mixing Energy:

Based on 5 W/m³ for mechanical mixing, the anoxic zone mixing energy is expected to be 936 kW.

Chemical Demand:

- Influent alkalinity is 225 mg/L as CaCO₃. Alkalinity required for nitrification was 194.3 mg/L as CaCO₃. Alkalinity produced by denitrification = $3.57 (NO_x N_e) = 86.8 \text{ mg/L}$ as CaCO₃. Alkalinity balance = (225-194.3+86.8) mg/L = 117.5 mg/L as CaCO₃ remaining. Therefore this reaction is no longer constrained by alkalinity demands and no chemical is required.
- No methanol is required for pre-anoxic denitrification since there enough scBOD entering the reactor to ensure complete denitrification.

Item	Unit	Value
Effluent NO3-N	mg/L	2.9
Internal recycle ratio	N/A	7.86
RAS recycle ratio	N/A	0.52
Anoxic Volume	m3	187,148
Overall SDNR	g NO3-N/g MLSS-d	0.06
Retention Time	h	6.8
Mixing Power	kW	936

Table 11: Summary of the pre-anoxic design

6.5.2 Suspended Growth Aerobic/Anoxic Denitrification Process

As an alternative design for comparison, a postanoxic suspended growth system was designed as a posttreatment step for the nitrification reactor previously described, with an effluent target of 2.9 mg/L as NO₃-N. For the postanoxic system shown in Figure 40, the scBOD substrate has been consumed in the aerobic process, leaving little available carbon for the heterotrophic denitrifiers. Therefore, methanol must be added to increase the rate of nitrification, where 1.5 g bCOD/g CH₃OH. The intention of this section is to demonstrate the required methanol dose, the modified anoxic volume and detention time, and waste sludge production for a post-anoxic suspended sludge system.

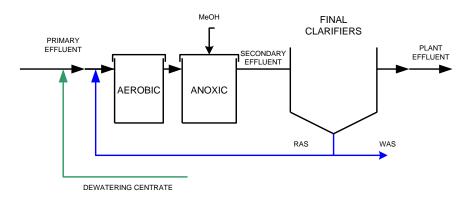


Figure 40: PFD for Postanoxic Nitrification/Denitrification with no side-stream treatment

Anoxic Basin Volume and HRT:

• To find the production from the growth of heterotrophic denitrifiers, the residual bCOD in the effluent from the anoxic zone was found using growth kinetics for denitrifiers given in Table 10 and the mass balance approach given in Equation 12 (p.35):

$$S = \frac{K_S[1+(k_d)SRT]}{SRT(\mu_m - k_d) - 1} = \frac{10.9[1+(0.05)7]}{7(0.18 \times 6.7 - 0.05) - 1} = 10.8 \text{ mg bCOD/L}$$

• The target NO₃-N reduction is from 27.2 mg/L to 2.9 mg/L (24.3 mg/L).

• Solids production in the anoxic volume was found using Equation 14 (p.36), assuming that the TSS entering the anoxic denitrification reactor after the aerobic nitrification reactor is 20 mg/L,:

$$P_{X,TSS} = \frac{QY(S_o - S)}{[1 + (k_d)\theta_c]^{0.70}} + \frac{f_d(k_d)QY(S_o - S)\theta_c}{[1 + (k_d)\theta_c]^{0.70}} + Q(TSS_o) = 12,919 + 632 + 13,210$$

$$P_{X,TSS} = 26,034 \text{ kg/day}$$

• The volume of the anoxic reactor can now be found at MLSS=1,750 mg/L:

$$(X_{TSS})(V) = (P_{X,TSS}) \theta_c$$

 $V_{nox} = \frac{(26,167)*(7)}{1,750} = 102,869 \text{ m}^3$

• The residence time for the anoxic reactor was found to be **3.7 hours** at the design flowrate for the reactor.

Exogenous Carbon Requirements:

Recall that the purpose of biological denitrification is to reduce nitrate biologically. Thus, there
must be sufficient bCOD to provide an electron donor for nitrate removal. The general rule is 4 g
of bCOD per g NO₃-N reduced. The required amount of bCOD per NO₃-N reduced is impacted by
the system denitrifier biomass yield, as described in Equation 22 (p.44):

$$\frac{bsCOD}{NO_3 - N} = \frac{2.86}{1 - 1.42Y_n} = 3.5 \text{ g bCOD used/g NO}_3 - \text{N reduced}$$

 It is assumed that no bCOD is available subsequent to the upstream aerobic nitrification process. Therefore, the required bCOD to reduce the NO₃-N must be supplied from an external carbon source, such as methanol. Thus, the total methanol requirements was calculated:

> bCOD Dose = (3.5 bCOD/NO₃-N)x(24.3 mg/L NO₃-N)+10.8 mg/L bCOD = 96.6 mg/L as bCOD x 1.5 mg COD/mg MeOH Methanol Dose = **64.4 mg MeOH/L** Daily Methanol Consumption = 64.4 mg MeOH/L x 660,523 m³/day = **42,536 kg/d**

Oxygen & Alkalinity Credit:

• The oxygen credit and alkalinity credit are identical to that developed for the preanoxic process design in Section 6.5.1.

Mixing Energy:

• The anoxic zone mixing energy is expected to be 505 kW.

A summary of the anoxic design is given in Table 12.

Item	Unit	Metcalf-Eddy
Effluent NO3-N	mg/L	2.9
Anoxic Volume	m3	102,870
Methanol dose	Kg/d	42,536
Retention Time	h	3.74
Mixing Power	kW	514

Table 12: Summary of the post-anoxic design

6.6 Side-stream Treatment Facilities Sizing

The side-stream to be treated for the example facility is the centrate or sludge water stream that is recycled to the headhouse of the plant. The process is to be sized for a flow of 4,500 m³/day from the dewatering centrifuges. The influent Total Nitrogen concentration exceeds 1,100 mg/L, and a low COD/N ratio of 1.1. Other side-stream characteristics are given in Table 8 (p.73).

Because the side-stream treatment process has a relatively low COD/N ratio compared to the main stream process, exogenous carbon will be necessary to increase the denitrification rate, similar to a postanoxic reactor for the main stream process. Therefore, methanol cost will be one of the additional considerations in the treatment of the side-stream load.

In order to assess the impact of the side-stream treatment on the overall treatment plant operating costs, sizing calculations were developed for three side-stream treatment options, depicted in Figure 41:

- Nitrogen removal via nitrification/denitrification;
- Nitrogen removal via nitritation/denitritation; and
- Nitrogen removal via nitritation/Anammox.

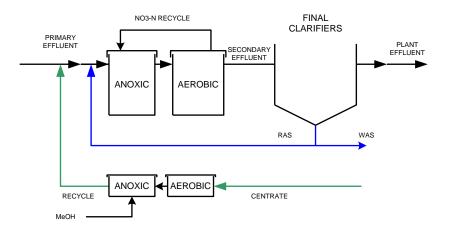


Figure 41: PFD for Side-stream options (a) nitrification/denitrification (b) nitritation /denitritation and (c) partial nitritation/ Anammox Assumptions:

- The mixed liquor temperature is 14°C because the centrate is mixed with the plant influent;
- All centrate TKN is biodegradable;
- The side-stream reactor is required to remove 90% of the centrate TKN prior to recycling to the head of the plant;
- The blower efficiency is 1.1 kg O₂/kWh;
- The price for methanol is \$446⁴² per metric tonne; and
- The price for electricity is \$0.10 per kWh.

The incremental annual oxygen and methanol requirement to treat the side-stream nitrogen load if it were routed to the main plant was found:

Oxygen requirements:

- The 90% removal efficiency of the side-stream reactor means that 1011 mg/L of TKN are reduced. For nitrification, 4.33 g O₂ is consumed per g TKN removed. The incremental aeration requirements for nitrification of the side-stream load is **825 kg O₂/hr** (AOTR).
- The addition of a denitrification step provides an oxygen credit of 545 kg/hr, for net O₂ consumption of 280 kg O₂/hr (AOTR) to treat the side-stream load in the main-stream reactor (a 66% reduction in oxygen used).

Methanol requirements:

• If the side-stream load were routed to the main plant, there would be sufficient bCOD available for denitrification in the mainstream anoxic reactor, therefore there would be no need to add methanol.

For the side-stream reactor configurations, the kinetic parameters for nitrifiers and methanol-degraders at 30°C are used as the basis for the design, with nitrifier growth at its maximum rate given in Table 13. The calculations are based on the design outlined in Chapter 17 (Side-stream Treatment) of "Design of Municipal Wastewater Treatment Plants", by Water Environment Federation and the American Society of Civil Engineers.

⁴² Current price as of March 2012, <u>http://www.methanex.com/products/methanolprice.html</u>

	Coefficient	Unit	Value	θ values 44	Adjusted Value
	μ _N	g VSS/g VSS-d	1.94	1.08	1.94
Nitrifiers	K _N	g NH4-N/m ³	0.7	1.053	0.513
Numers	K _{d,N}	g VSS/g VSS-d	0.17	1.029	0.23
	SRT	d	1.28	-	-
Methanol	μ _M	g VSS/g VSS-d	1.3	1.072	2.61
degrader	K _{d,M}	g VSS/g VSS-d	0.04	1.029	0.05
growth	SRT	D	0.98	-	-

Table 13: Kinetic parameters for nitrifiers and heterotrophic methanol degraders at 30°C⁴³

Design goals for this stage are to determine:

- Aerobic and Anoxic reactor volume
- Oxygen requirements, blower air usage, power consumption
- Methanol consumption
- Effluent concentration of TN, Nitrate removal, total nitrogen removal
- Mixing energy requirements

6.6.1 Side-stream Nitrification/Denitrification

In this example, the side-stream reactor is made up of two complete-mix compartments in series, as seen in Figure 41. The first compartment is aerobic, the second is anoxic (post-anoxic configuration). There is no sludge residence so there is no requirement for a clarifier. A side-stream reactor was designed following a simplified approach to the calculation given in Sections 6.4 & 6.5 for the main-stream nitrification/denitrification reactor, with adequate mean cell residence time of nitrifiers and denitrifiers.

Aerobic and Anoxic Reactor Volume:

- The design approach selected was for two separate suspended-growth reactors. The reactor was sized for the side-stream temperature of 30°C, as the mixed liquor for the reactor will already be in the mesophilic range, and further heated by mechanical dewatering (i.e. centrifugation) and nitrification & denitrification reactions.
- The mean cell residence time required for the biomass residence of nitrifiers was found from Equation 10 (p.33) for design MLSS of 1,750 mg/L. Nitrifier growth and decay rate at 30°C is at an optimal temperature to maximize growth, as shown in Table 13. An operating factor of 2.5 was selected for the side-stream reactor:

$$\mu_n = \mu_{n,max} \left(\frac{N}{K_n + N}\right) \left(\frac{O_D}{K_o + O_D}\right) - k_{dn} = 1.94 \left(\frac{112}{0.513 + 112}\right) \left(\frac{2.0}{0.50 + 2.0}\right) - 0.23 = 1.32 \text{ g/g-d}$$

⁴⁴ Correction for temperature $\mu_{m,T} = \mu_m \theta^{T-20}$; $k_{d,T} = k_2 \theta^{T-20}$

⁴³ Adapted from (Water Environment Federation and the American Society of Civil Engineers/Environmental and Water Resources Institute , 2010)

$$\theta_c = rac{2.5}{\mu_n} = rac{2.5}{1.32} =$$
 1.89 days

• Therefore the minimum SRT required for nitrifier biomass residence is lower than the **6.72 days** if at ambient temperature. Similary, the SRT required for denitrifier (methanol-degrader) growth was found:

$$\theta_c = \frac{2.5}{\mu_{max} - k_d} = \frac{2.5}{2.61 - 0.05} = 0.98 \text{ days}$$

- The design flow rate for the side-stream was 4,523 m³/d. For the minimum solids residence times calculated above with a design MLSS of 1,750 mg/L, the required aerobic reactor volume for nitrifer growth was **5,638 m**³, and for **4,431 m**³ for the anoxic reactor.
- For a reactor designed for 90% removal of TKN, with effluent nitrate of no more than 150 mg/L as NO3-N, and effluent ammonia concentration of no more than 112 mg/L as NH₃-N, to determine the internal recirculation (R) ratio:

$$R = \left[\frac{N_{TKN} - N_{Nitrate,eff}}{N_{Nitrate,eff}}\right] - 1 = \left[\frac{1,124 - 112}{150}\right] - 1 = 7.67$$

Therefore a 767% internal recycle from the end of the anoxic zone to the head to the reactor is required. This may present technical challenges due to dissolved oxygen being recycled to the anoxic zone with the nitrates and is recommended for further study.

Oxygen Requirements:

• The aeration energy requirements are identical to that developed for the main stream reactor. The advantage to the side-stream configuration is the flexibility to optimize the oxygen transfer efficiency (OTE) by increasing basin depth, requiring a smaller footprint in an existing plant layout, and less energy required for aeration.

Methanol Requirements:

Denitrification will require the addition of a carbon source like methane to provide the bCOD necessary to remove nitrate from the side-stream. Assuming that 2.47 g of methanol are required for every gram of TKN removed via nitrification and denitrification. Higher dosages may be needed to overcome methanol losses to the effluent and scavenging by oxygen from the internal recycle. For the 1, 011 mg/L of TKN which are removed through the side-stream treatment, the required methanol dose will be nearly 2,500 mg/L. The daily methanol demand would be 11,300 kg/day.

Other Considerations:

- The excess nitrate that is produced by the side-stream reactor that is fed to the main plant BNR process was approximately 680 kg/day. This would reduce the excess denitrification capacity of the main plant from 135% to 129% (about 6%).
- Mixing energy would entail approximately 22 kW for the anoxic zone.
- The exothermic nitrification & denitrification reactions may result in the increase of the reactor temperature in excess of 35°C, after which point the kinetics of nitrifiers become inhibited. A detailed heat balance should be performed to assess the need for cooling.

6.6.2 Side-stream Nitritation/Denitritation

The side-stream reactor configuration used in this example is aerobic nitritation followed by anoxic denitritation. Assume 90% removal of nitrogen using a reactor composed of two complete mix compartments in series. An internal recirculation stream provides nitrates to the head of the side-stream treatment train. The reactor size will be similar to that already presented for the side-stream nitrification/denitrification reactors. The difference in this case was that the aeration requirements were reduced because the TKN is hydrolyzed to ammonia, then the ammonia oxidation is stopped at nitrite:

Oxygen requirements:

- Assume that 3.43 g O₂ is consumed per g TKN removed by nitritation/denitritation. For 1,011 mg/L as TKN converted to NO₂-N, the required oxygen is 614 kg O₂/hr (actual). Using nitritation instead of full nitrification saves approximately 212 kg O₂/hr.
- The addition of a denitritation step provides an oxygen credit of 316 kg/hr, for net O_2 consumption of **298 kg O_2/hr** to treat the side-stream load in the main-stream reactor (a 50% reduction in oxygen used over nitritation).

Methanol requirements:

 Methanol requirements will be reduced since less carbon is needed to convert nitrite than for nitrate. Assuming that 1.53 g of methanol is required for every gram of TKN removed via nitritation and denitritation, for the 1, 011 mg/L of TKN which are removed through the sidestream treatment, the required methanol dose will be about 1,550 mg/L. The daily methanol demand would be 7,000 kg/day. Using denitritation from nitrite instead of full denitrification from nitrate will save 4,300 kg methanol per day.

6.6.3 Side-stream Partial Nitritation/Anammox

Oxygen requirements:

1. Recall that the goal of partial nitritation is to achieve an effluent concentration with $50\% \text{ NO}_2\text{-N}$ and $50\% \text{ NH}_3\text{-N}$ to provide optimal feed for the Anammox process. This entails the conversion of

500 mg/L as NH_3 -N to NO_2 -N by the aerobic nitritation process. This would require **341 kg O₂/hr** (actual). Compared to a nitrification/denitrification at 280 kg O_2 /hr, the Partial Nitritation/Anammox process would use more oxygen.

Methanol requirements:

2. Using the Partial Nitritation-Anammox process for side-stream treatment does not require any methanol addition.

Other Considerations:

3. Cooling of the side-stream reactor contents may not be necessary since the Anammox process is not as strongly exothermic as denitrification. Detailed heat balance calculations are needed to assess the need for cooling.

6.7 Summary of Reactor Design Concepts

A comparative evaluation of the reactor designs presented in the above sections is discussed in this section. The main plant aerobic reactor concepts are summarized in Table 14. The main plant anoxic reactor concepts follow in Table 15, and the side-stream reactors are summarized in Table 16.

For the main plant aerobic reactors:

- In order to support nitrifier growth, the required aeration basin volume more than doubled with the addition of nitrification to carbonaceous BOD removal. Likewise, the oxygen requirements to support the nitrification process increased by over 10,000 kg/h compared to BOD-removal only. It is notable that the oxygen requirement for the main stream aerobic reactor was reduced by about 5,000 kg/h, or 23% by adding the side-stream treatment process.
- 2. The nitrification process effectively reduced the ammonia to 10 mg/L in the effluent. However, nitrification produced 27.2 mg/L of nitrate, compared with the 20.3 mg/L of nitrate produced in the scenario which includes side-stream treatment. This allows a smaller denitrification reactor volume to be used than if the centrate were routed to the main plant for treatment.

Design Parameter	Unit	BOD Removal Only	BOD removal and Nitrification	BOD Removal and Nitrification with Side-stream Treatment
Aerobic SRT	d	3	7	7
Aeration basin volume, total	m3	245,836	512,622	510,751
Hydraulic residence time	Hours	9	19	19
MLSS	mg/L	1,750	1,750	1,750
% VSS	-	70%	70%	70%
F/M	g/g-d	0.72	0.35	0.35

Table 14: Summary of Activated Sludge Treatment Reactors for BOD-Removal and Nitrification

Design Parameter	Unit	BOD Removal Only Nitrification		BOD Removal and Nitrification with Side-stream Treatment
BOD loading	Kg BOD/m3-d	0.80	0.38	0.38
Excess Sludge Production	Kg/d	143,420	129,732	129,259
Observed Yield	Kg TSS/kg bCOD	0.78	0.71	0.70
	Kg VSS/kg bCOD	0.55	0.49	0.49
Oxygen Required (AOTR)	Kg/h	5,830	9,668	8,870
Oxygen Required (SOTR)	Kg/h	11,339	21,153	16,305
RAS ratio	-	0.52	0.52	0.52
Alkalinity Consumption	mg/L as CaCO3	0	194.3	145.3
Effluent NH4-N concentration	mg/L	42	10	10
Effluent NO ₃ -N concentration	mg/L	0.0	27.2	20.3

A comparison of the denitrification system follows:

- 1. The anoxic reactor volume required for the post-anoxic configuration is about half of that required for the pre-anoxic system, with and without the side-stream treatment reactors included. Likewise, the HRT of the postanoxic configuration is about half.
- 2. The oxygen credit by adding an anoxic denitrification zone for the main plant reduced the total oxygen demand by 20% for the full-strength load, and by 16% for the nitrogen-reduced load.
- 3. The alkalinity credit produced enough alkalinity equivalents (as CaCO3) to ensure that excess alkalinity existed after nitrification had occurred such that adequate alkalinity remained for pH-neutrality, and also saving the associated costs for chemical addition.
- 4. There is a significant cost associated with the methanol demand for the post-anoxic configurations. The methanol is required as a carbon source for the heterotrophic denitrifiers since there is not enough remaining after the aerobic zone.
- 5. There appears to be little advantage to post-anoxic configuration over preanoxic configuration, due to increased chemical consumption of methanol as a carbon source for heterotrophic denitrification. At a cost of \$446/MT MeOH, this amounts to \$5-6 million in chemical usage per year. For this system, postanoxic configuration would result in unnecessary costs and associated greenhouse gas emissions resulting from the need to add methanol. This "avoided cost" is illustrative of the benefit of the pre-anoxic configuration for denitrification for the main plant. Therefore, only the preanoxic configuration was included in the mainstream treatment process comparison.

Item	Unit	Preanoxic	Postanoxic	Preanoxic w/Side- stream	Postanoxic w/side- stream
Anoxic Volume	m³	187,148	102,870	185,867	90,157
Retention Time	hrs	6.80	3.74	6.80	3.28
Oxygen Credit (AOTR)	kg/h	1,900	1,900	1,364	1,364
Alkalinity Credit	mg/L as $CaCO_3$	87	87	62	62
Methanol dose	kg/d	0.0	64.4	0.0	48.2
Methanol cost	\$MIL/year	\$6.92	\$6.92	\$5.19	\$5.19

Table 15: Summary of Denitrification Reactor

For the side-stream reactors:

- The side-stream reactors were designed to accommodate a centrate load of 4,523 m³/day @ 1,011 mg-N/L entering the reactor for approximately 90% removal of NH3 and 77% removal of TN;
- 2. Exogenous carbon would be needed for Nitrification/Denitrification, since the dewatering centrate has a COD/N of less than 1.5. The Partial Nitritation-Anammox side-stream, with no requirement for exogenous carbon source. Using the Anammox from nitrite and ammonia instead of full denitrification from nitrate will save 11,298 kg of methanol per day.
- 3. With the side-stream 90% removal rate of NH3 and ~77% of TN, the output of the reactor should be 262 kg-N/day, or with the 12,990 m³ nitrification-denitrification reactor described above, 0.067 kg-N/m³-day which is well within the range of typical PN-Anammox processes reported in the literature.
- 4. The deciding factor would then be the comparison of cost between the nitritation/denitritation process with methanol addition with the oxygen credit resulting from denitritation, or the partial nitritation/Anammox process with no methanol addition but higher oxygen demand.

		Nitrification/ Denitrification	Nitritation/ Denitritation	PN/Anammox
Aerobic reactor volume	m3	8,559	6,587	6,587
Anoxic reactor volume	m3	4,431	4,431	N/A
Oxygen Demand SOTR	kg/hr	545	580	663
Aeration Power Consumption=	kWh/day	1.70	1.81	2.07
Methanol Demand	kg/day	11,298	6,998	-
Energy Cost*	\$/year	\$ 61,999	\$ 65,964	\$ 75,448
Chemical Cost+	\$/year	\$ 1,839,188	\$ 1,139,254	\$ -
Total	\$/year	\$ 1,901,187	\$ 1,205,219	\$ 75,448

Table 16: Summary of side-stream reactor concepts

=Assume blower efficiency is 18 Wh/m³; Fine-bubble 25% diffusers * 2012 Electricity price assumed to be \$0.10/kWh

+ 2012 Methanol price assumed to be \$446/metric tonne

The costs for energy and chemical for main stream reactors was developed based on power demands for aeration and mixing, and for chemical costs attributed to alkalinity and methanol in Table 17. No cost comparison to other full-scale processes is possible at this time as the real cost data are not widely available.

Table 17: Cost calculation development for main stream reactors

Reactors	Aerobic Vol	Anoxic Vol	Footprint	Aeration Power	Mixing Power	MeOH	Energy Cost	Chemical Cost	Total Cost
	m³	m³	m²	kWh/day	kWh/day	kg/day	\$/day	\$/day	\$/day
BOD Removal - Coarse Bubble	245,836	0	61,459	268,552	0	0	26,855	0	26,855
BOD Removal - Fine Bubble	245,836	0	61,459	68,749	0	0	6,874	0	6,874
BOD and N Removal - Nitrification	512,622	0	128,156	128,252	0	0	12,825	0	12,825
AST and Preanoxic Denitirification	512,622	187,148	174,943	103,043	22,458	0	12,550	0	12,550
AST and Postanoxic Denitrification	512,622	102,870	153,873	103,043	12,344	42,536	11,538	18,971	30,509
AST and Preanoxic Denitirification with Side-stream Pretreatment	510,751	185,867	174,154	88,506	22,304	0	11,081	0	11,081

Similarly, the costs for energy and chemical for the side stream reactors is summarized in Table 17.

		Aerobic Vol	Anoxic Vol	Footprint	Aeration Power	Mixing Power	MeOH	Energy Cost	Chemical Cost	Total Cost
Reactor 1	Reactor 2	m³	m³	m²	kWh/day	kWh/day	kg/day	\$/day	\$/day	\$/day
Nitrification	N/A	8,559	0	2,140	5,003	0	0	500	0	500
Nitrification	De- nitrification	8,559	4,431	3,248	1,699	531	11,298	702	5,039	5,740
Nitritation	N/A	6,587	0	1,647	3,721	0	0	372	0	372
Nitritation	De- nitritation	6,587	4,431	2,754	1,807	531	6,998	712	3,121	3,834
Partial Nitritation	Anammox	6,587	49,755	14,085	2,067	0	0	207	0	207

Table 18: Cost calculation development for side stream reactors

The operating costs for the different strategies of biological removal of Total Nitrogen that were discussed are summarized in Table 19. It is clear that although the operating cost for the activated sludge process for BOD-removal only with fine bubble diffusers (Case 2) is the lowest, it does not meet the requirements for less than 10 mg/L as NH3 and 2.9 mg/L as NO3-N in the plant effluent. And although the advanced secondary treatment with preanoxic denitrification (Case 3) meets the requirement for ammonia removal, it does not meet the requirement for nitrate removal. Since the operating cost for the side-stream treatment using the Nitritation/Denitritation process (Case 4) was higher than for the conventional BNR process (Case 4), it was not considered feasible due to cost considerations.

The best choice was the BNR process coupled with side-stream Partial Nitritation-Anammox treatment (Case 5). In comparison to Case 4, it is projected that Case 6 (using an Anammox process) will result in savings of **\$460,739 per year**, while attaining a high level of Total Nitrogen removal (i.e to the performance target of 10 mg/L NH3-N and 2.9 mg/L NO3-N). The cost per megaliter treated for the process with Anammox side-stream treatment was **\$17.1/ML** with 70% TN removal, which provides more effective Total Nitrogen removal compared to either Case 3 at \$19.4/ML with only 12% TN removal, Case 4 with \$19.0/ML with 70% TN removal, or Case 5 with \$22.6/ML for equivalent treatment.

Case	Plant Configuration	kWh/day	m²	\$/year	NH3%	TN%	\$/ML
1	ASP with Coarse Bubble Diffusers	268,552	61,459	9,802,139	0%	0%	40.7
2	ASP with Fine Bubble Diffusers	68,749	61,459	2,509,348	0%	0%	10.4
3	AST with Fine Bubble Diffusers	128,252	128,156	4,681,196	76%	12%	19.4
4	AST and Preanoxic Denitirification	125,500	174,943	4,580,761	76%	70%	19.0
5	AST and Preanoxic Denitirification + Side- stream Treatment by Nitritation/Denitritation	112,617	177,402	5,443,850	76%	70%	22.6
6	AST and Preanoxic Denitirification + Side- stream Treatment by PN/Anammox	112,877	188,733	4,120,022	76%	70%	17.1

Table 19: Summary of operating costs for different plant configurations

6.8 Plant-Wide Simulations

6.8.1 Introduction to CHEApet

Activated sludge treatment involves complex mixture of bacteria and substrates which has led to the development of matrix models to describe reactions under aerobic and anoxic conditions. Various software programs are available to handle the complex set of equations developed.

The Activated Sludge Model No. 2 (ASM2) is an example of a generic model can be used study biological processes and/or applied to optimize full-scale processes. Modeling software such as BioWIN or GPS-X can be used to dynamically model processes using the ASM for novel treatments such as Nitritation/Denitritation and DEMON. Other modeling tools such as Biosolids Emissions Assessment Model (BEAM) to determine greenhouse gas emissions resulting from biosolids management practices, and the Life-Cycle Assessment Manager for Energy Recovery (LCAEMER) from anaerobically digested wastewater solids (Water Environment Research Foundation) can be used to generate estimates for greenhouse gas emissions.

CHEApet is a web⁴⁵-based tool that provides estimates for a plant-wide steady-state mass and heat balances that includes an option for side-stream Anammox treatment. CHEApet was selected for the simulation of total plant scenarios on the merits of its simplicity and wide availability. The ASM2d model is used as the basis for the aeration system modeling in CHEApet. Carbon Heat Energy Analysis Plant Evaluation Tool developed by CH2M Hill through WERF. CHEApet was used to compare scenarios to evaluate the impact of different treatment strategies on the criteria of nitrogen removal, operating cost, and green house gas emissions.

6.8.2 Treatment Plant Simulations for CHEApet

CHEApet was used to simulate five different treatment plant configurations (scenarios):

- 1. Activated sludge process (ASP) for BOD-removal using coarse-bubble diffusers
- 2. Activated sludge process (ASP) for BOD-removal using fine-bubble diffusers
- 3. Advanced Secondary Treatment (AST) for BOD and ammonia removal with extended SRT
- 4. Biological Nitrogen Removal (BNR). The Modified Ludzack-Ettinger preanoxic system was selected for the mainstream plant.
- 5. Biological Nitrogen Removal (BNR) with Side-stream Treatment. The Partial-Nitritation/Anammox system was selected for the side-stream process.

The simulations were carried out under two scenarios, (a) baseline and (b) optimized. Under the baseline treatment scenario, the effluent target was 10 mg/L as NH3-N and 2.9 mg/L as NO3-N. The

⁴⁵ The tool is freely available for use at <u>cheapet.werf.org</u>

simulations were repeated for optimized treatment scenarios, in which the target effluent concentration for ammonia and for nitrate was the limit of the technology (e.g. maximize the total nitrogen removal % that could be achieved using the simulation software).

Evaluation criteria:

- (1) Total Nitrogen Removal (%) was defined as the mass of total sum of nitrogen compounds removed from the raw sewage influent to the final plant effluent. The percentage does not represent the level of nitrification because a portion of the TKN is taken by biomass.
- (2) Total Operating Cost (\$MIL) was defined as the cost for electrical power for aeration and for chemical costs such as methanol addition.
- (3) Total Greenhouse Gas Emissions were defined as the sum of the direct process emissions and the indirect emissions.

The block flow diagram of a generic wastewater treatment plant is provided on Figure 42 for CHEApet.

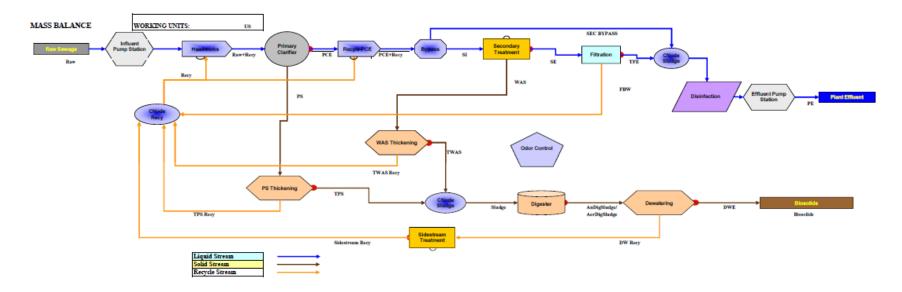


Figure 42: Schematic of Wastewater Plant from CHEApet⁴⁶

⁴⁶ Adapted from (Crawford, Johnson, Johnson, Krause, & Wilner, 2011)

6.8.3 Results of Baseline Treatment Simulations

For the baseline treatment scenarios, the following assumptions were made:

- 1. Target 10 mg/L as NH₃-N and 2.9 mg/L as NO₃-N (13 mg/L as NO₃ ion);
- 2. SRT = 4.7 was selected from Figure 13 for target ammonia removal;
- 3. A reactor volume of 201,682 m³ was used for the main process. For the BNR process, the anoxic volume was 25% of the reactor volume and the aerobic volume was 75%.

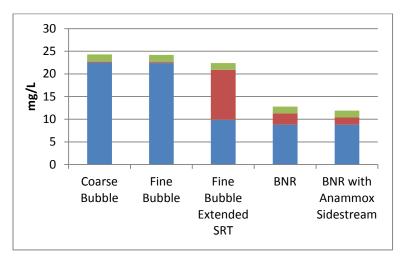
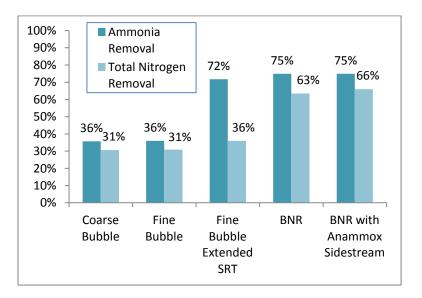


Figure 43: Comparison of Effluent Concentrations for baseline scenarios with influent load of 35 mg/L as Total Nitrogen





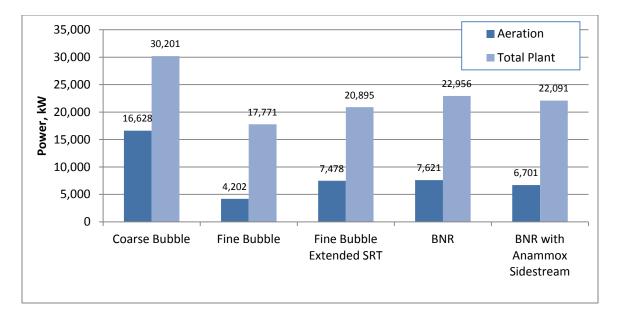


Figure 45: Comparison of Aeration Energy Demand Total Plant Energy Demand for baseline scenarios

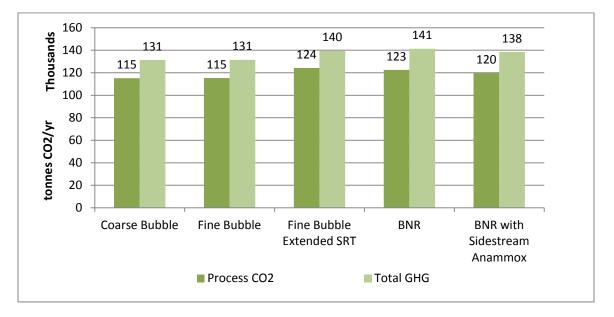


Figure 46: Comparison of Direct Process Emissions and Total Plant Greenhouse Gas Emissions

6.8.4 Results of Optimized Treatment Simulations

For the "optimized" treatment scenarios, the assumptions were:

- 1. Removal to "limits of technology", or as much as could be removed by the simulator software
- 2. SRT = 7 was selected from Figure 13 for target ammonia removal

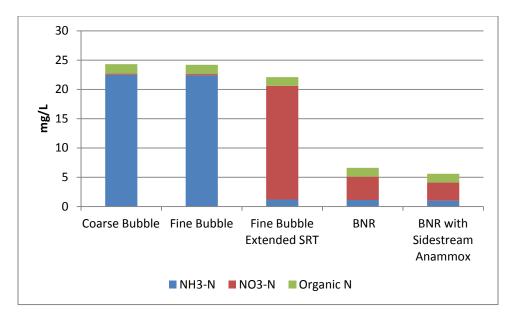


Figure 47: Comparison of Effluent Concentrations for "optimized" scenarios with influent load of 35 mg/L as Total Nitrogen

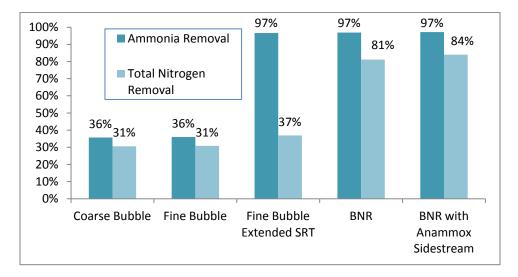


Figure 48: Comparison of percentage nitrogen removal for "optimized" scenarios

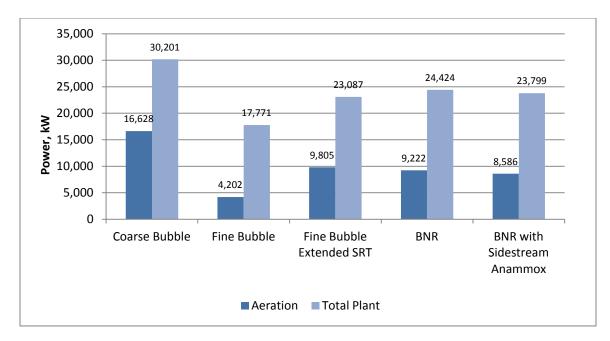


Figure 49: Comparison of Effluent Concentrations for "optimized" treatment scenarios

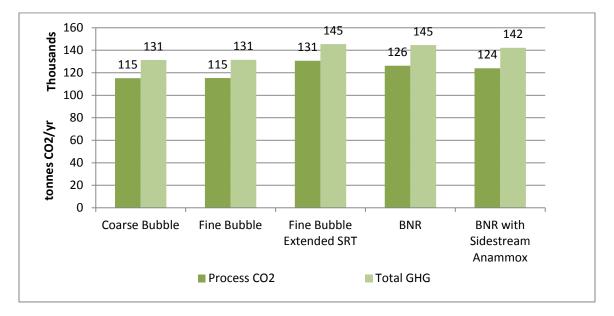


Figure 50: Comparison of Greenhouse Gas Emissions for "optimized" treatment scenarios

6.9 Discussion

Comparison of different treatment configurations, for both the baseline and optimized treatment scenarios, from Table 20:

- <u>Activated Sludge Process</u>: With low SRT, only BOD-removal occurred. There was no impact on nitrogen removal due to use of coarse or fine bubble diffusers (expected). However the improved oxygen transfer efficiency resulting from the use of fine bubble diffusers significantly reduced the airflow and aeration power consumption from \$22.22/ML to \$5.61/ML. The cost to run a BNR process with coarse bubble aeration is prohibitive and was not considered further than to demonstrate the energy requirements for nitrification with a coarse bubble system. Greenhouse gas emissions were the lowest for this treatment scenario. However, Ammonia removal was only 36%, and TN removal was only 31%.
- 2. <u>Advanced Secondary Treatment</u>: Extension of the SRT from 4.7 days to 7 days increased the removal of ammonia to above 70%, without effectively removing Total Nitrogen (i.e. 31%). The occurrence of nitrification drastically increased the oxygenation requirements, such that the operating cost increased from \$5.61/ML to \$9.99/ML for the baseline scenario. Also, the greenhouse gas emissions increased by 9 ktonne CO₂/year for the increased conversion of ammonia to nitrate. For the optimized scenario, the ammonia removal efficiency was increased to 37% for a cost of \$13.10/ML.
- 3. <u>Biological nitrogen removal</u>: The addition of the anoxic denitrification process for BNR without side-stream improved TN removal from 36% to 63% in the baseline scenario, and up to 81% in the optimized scenario. The operating cost compared to Case III was \$10.18/ML for the baseline scenario, or \$12.32/ML for the optimized scenario. This demonstrates a reduction in aeration demand for the mainstream reaction process resulting from the oxygen credit, compared to the process without denitrification. The addition of the denitrification reactor reduced direct process emissions by about 1 tonne CO₂/year.
- 4. <u>Biological nitrogen removal with side-stream treatment:</u> The addition of side-stream treatment with a Partial Nitritation-Anammox reactor further increased 66% TN removal, and up to 84% in the optimized scenario. Cost for the treatment was reduced compared to \$8.85/ML for the baseline scenario and \$11,47/ML, when compared to the cost of treatment if there were no side-stream treatment. As shown in Figure 46, direct process emissions were reduced by 3% compared with nitrification-only (from 124 to 120 tonnes CO₂ emitted/year).

From Table 20, it can be seen that the operating cost (based on power consumption) for a BNR process with side-stream Partial Nitritation-Anammox treatment (Case III) predicted by CHEApet is **\$680,652 per year** for the Case I scenario (or \$1.04/ML) compared to extended SRT (Case II). For the optimized treatment scenario, the cost avoidance for using side-stream Partial Nitritation-Anammox treatment (Case VIII) versus the extended SRT process (VI)is **\$1,067,844 per year** (or \$1.63/ML).

Case		Total	Operating Cost		Total Greenhouse Gas
		Nitrogen	(\$/year)	\$/ML	Emissions
		Removal (%)			(tonnes CO ₂ /year)
I	ASP w/ Coarse Bubble	31%	\$ 14,566,128	\$22.22	131
П	ASP w/ Fine Bubble	31%	\$ 3,680,952	\$5.61	131
Ш	Extended SRT (baseline)	36%	\$ 6,550,728	\$9.99	140
IV	BNR (baseline)	63%	\$ 6,675,996	\$10.18	141
V	BNR w/ Anammox (baseline)	66%	\$ 5,870,076	\$ 8.95	138
VI	Extended SRT (optimized)	37%	\$ 8,589,180	\$ 13.10	145
VII	BNR (optimized)	81%	\$ 8,078,472	\$ 12.32	145
VIII	BNR w/ Anammox (optimized)	84%	\$ 7,521,336	\$ 11.47	142

Table 20: Summary of Total Nitrogen Removal, Operating Cost, and Total Greenhouse Gas Emissions using CHEApet

7 Conclusions & Recommendations for Future Study

7.1 Side-stream vs. Mainstream Treatment

After the calculations presented in the previous sections, it is clear that the addition of the side-stream treatment with the Partial Nitritation-Anammox process results in overall improved Total Nitrogen removal, lower operating costs in terms of both oxygen and chemical requirements, and lower GHG emissions. It meets all three of the criteria imposed for the process that is the "best choice" in the removal of nitrogen from wastewater. The savings are defined in terms of comparison to a nitrifying plant (the extended SRT Advanced Secondary Treatment configuration, which removes ammonia removal but low total nitrogen removal).

- <u>Total Operating Cost Reduction</u>: is projected to result in savings of \$460,739 to \$680,652 per year. The avoided cost per megaliter treated for the process with Anammox side-stream treatment compared to advanced secondary treatment for nitrification is projected to be between \$1.04 to \$2.30/ML
- 2. <u>Total Nitrogen Removal</u>: is expected to increase Total Nitrogen removal to **81%-84%**, the highest level of Total Nitrogen removal for all configurations that were considered.
- <u>Total Greenhouse Gas:</u> is estimated to reduced Total Greenhouse Gas emissions by 7,000 tonnes of CO₂ emitted per year.

These results demonstrate that the side-stream Anammox treatment is effective for the removal of Total Nitrogen from wastewater effluent more cost-effectively and with lower carbon emissions than if treated through the main process by either (a) conventional biological removal of Total Nitrogen by nitrification/denitrification, or by (b) Advanced Secondary Treatment for ammonia removal. It should be stressed that the results presented in this project are the result of a preliminary modeling effort as proof-of-concept. It is recommended that detailed modeling and further investigation be carried out.

7.2 Next Steps for Anammox

The role of microbes in regulating global processes is gaining wider recognition in the public eye. For example, in 2011 the Netherlands postal service celebrated the centenary of the Dutch Society of Microbiology with a set of stamps depicting 10 different bacteria, of which Anammox was one, as seen in Figure 51, in which the arrow points to the Anammox bacterium:



Figure 51: Stamps issued to celebrate the 100th anniversary of the Dutch Society of Microbiology⁴⁷

The application of Anammox in engineered systems for efficient nitrogen removal represents a triumph of the scientific community that took less than five decades to move from postulate to discovery to fullscale implementation, and will undoubtedly continue to play an important role in the future of wastewater treatment:

- <u>Wide-scale revision of the nitrogen cycle:</u> Environmental science and engineering texts will start to print a revised version of the nitrogen cycle, showing the Anammox shortcut, and to emphasize novel metabolic routes such as nitritation/denitritation or nitritation/Anammox for engineering applications (Figure 5).
- <u>Wastewater Plants as Energy-producing Biorefineries</u>: As water resources come under more intensive usage through increased global industrialization and climate change, the reuse of wastewater will become an increasingly important topic in North America. Further attention may yet turn to nutrient recovery in addition to biogas utilization. For example, it has been

⁴⁷ Adapted from <u>http://www.exploremars.org/urine-to-fuel-your-rocket/</u>

suggested that the we will run out of phosphorus reserves than before we run out of petroleum. We will not call it "wastewater" then, but will be seen as an opportunity to extract resources such as fertilizers, chemical precursors, and fuels – like struvite, methanol, ammonia, nitrates, and biosolids, to name a few commercially viable products ((Chandran, 2011))

- <u>Seed Sludge as a Commodity</u>: What Anammox biomass was considered to be a desirable commodity for new plant start-ups? Each sludge is unique and may cultivate Anammox strains that are acclimated to certain types of treatment (e.g. Low temperature), enhancing the selection of sludges for other treatment facilities using the Anammox process.
- <u>Emissions Credits</u>: What if a cap-and-trade system became a reality? The upgrade of Advanced Secondary Treatment plants to include biological denitrification would be a step towards reducing N2O emissions from the bioreactor. In this scenario, greenhouse gas emission credits could provide an incentive to upgrade AST plants to include denitrification and side-stream removal. Under such a system, carbon-capped industries may trade credits from wastewater treatment facilities that were carbon-neutral or carbon-sink. Some analyses have suggested that the economic impact of emissions credits would be to leverage wastewater treatment plants to generate revenue based on the reduction of greenhouse gas emissions (Wang, Hamburg, Pryor, Chandran, & Daigger, 2011).
- <u>Hydrazine Biorefinery for Propulsion in Space:</u> October 2011 saw a profusion of articles in popular science magazines with sensational titles like "Urine to fuel your rocket-ship", and "Pee power: Urine-loving bug churns out space fuel." The naturally-occurring hydrazine yields are too low for this to be feasible. What processes are involved in the production of hydrazine, to improve the catalyst and boost production by the bacteria (Jetten 2011)?

7.3 Mainstream Redesign

From a technical standpoint, the next steps involve the challenge of holistic redesign of the wastewater treatment process in order to largely circumvent aeration. By increasing solids capture in the primary clarifiers, more organic compounds are converted to increase biogas production in anaerobic digesters.

The design concept for mainstream Anammox process for biological removal of Total Nitrogen involves a very-high-load activated sludge system where soluble matter is converted to biomass that can be flocculated and separated together with non-degraded suspended material in a settler. Most organic matter is removed and concentrated, and digested anaerobically to produce biogas and stabilized sludge. The effluent from the first stage and digester supernatant is treated in a granular-sludge Anammox reactor, as seen in Figure 52.

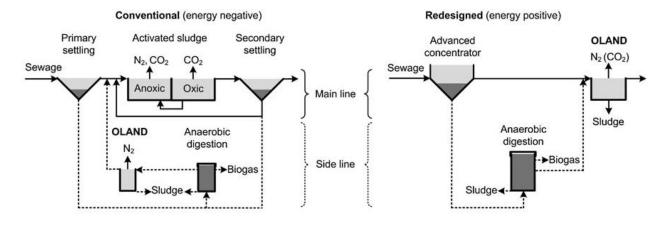


Figure 52: Conventional and redesigned sewage treatment schemes with OLAND in the side and mainstream line⁴⁸

Studies suggest that a wastewater treatment plant that removes nitrogen by using the Anammox process in the mainstream would yield energy (Kartal, Kuenen, & Van Loosdrecht, 2010). In the redesigned scheme, energy-positive sewage treatment can, in principal, be obtained by maximizing energy recovery through anaerobic digestion of concentrated organics in the side stream and by minimizing energy consumption for the biological conversion step and the residual nitrogen removal step (Clippeleir, Yan, Verstraete, & Vlaeminck, 2011). This would represent a major achievement in energy sustainability that could enable wastewater treatment plants of today to transform into the biorefineries of the tomorrow.

⁴⁸Adapted from (Clippeleir, Yan, Verstraete, & Vlaeminck, 2011).

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