

CHALMERS



Nitrogen removal in process water from the Gerum tunnel

A lab analysis using nitrification and denitrification as biological treatment

Master of Science Thesis in the Master's Programme Infrastructure and Environmental Engineering

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Department of Civil and Environmental Engineering
Division of *Water Environment Technology*
CHALMERS UNIVERSITY OF TECHNOLOGY
Göteborg, Sweden 2014
Master's Thesis 2014:113

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Examensarbete/Institutionen för bygg- och miljöteknik,
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Cover:

Left side figure: *The Gerum tunnel*. Upper right figure: *Pulsebäcken*. Lower right figure: *Gerumsälven*. (Photographs by Kjell Bengtsson)

Chalmers Reproservice / Department of Civil and Environmental Engineering
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ABSTRACT

High nitrogen concentrations in process water from tunnel constructions have become an issue due to residues from explosive, transported to recipients in the construction area. Biological treatment in terms of nitrification and denitrification are necessary to remove the nitrogen compounds in the process water. Skanska Sweden AB is currently constructing a tunnel in Tanumshede called the Gerum tunnel, for which the biological treatment of the process water has not worked during the project why this study was initiated. The aim of this master thesis was to assess which factors and parameters affect the biological treatment of process water in tunnel construction with regard to nitrification and denitrification. Lab tests on process water from the tunnel project were performed to achieve a complete nitrification and denitrification.

Three nitrification tests and one denitrification test were performed in the lab where the first nitrification test used process water from two separate days and sludge that was collected from a wastewater treatment plant. The nitrification process was incomplete since nitrate did not increase even though there was a slight decrease in ammonium and a slight increase in nitrite. The second test comprised of the same water and sludge materials as Test 1 with the exception of adding sodium dihydrogen phosphate, thereby increasing the phosphate concentrations which were thought to limit the reactions. This test also failed to show any nitrifying activity. The alkalinity measured in the process water was very low and for the third test, alkalinity and phosphate were therefore increased. This test gave the best results of the nitrification tests and resulted in ammonium being converted to nitrite, however the process failed to transform nitrite to nitrate. The results from the denitrification test did show a denitrifying activity when nitrate was transformed to nitrite at the same time as total nitrogen (TN) decreased which indicates that nitrogen gas has been formed, though the test should have been carried out for a longer period of time. Further lab analysis on process water in tunnels with high nitrogen content is crucial which should focus on metal concentration and the proportions of sludge and process water since those parameters were not thoroughly analyzed in this study.

Key words: nitrogen, nitrification, denitrification, process water, tunnels, inhibiting factors, water assessment

Kväverening av processvatten från Gerumstunneln

En labbanalys där nitrifikation och denitrifikation används som biologisk rening

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SAMMANFATTNING

Höga kvävehalter i processvatten från tunnelkonstruktion har blivit ett problem på grund av rester från sprängmedel som transporteras till vattenrecipenter inom byggområdet. Biologisk behandling såsom nitrifikation och denitrifikation är nödvändigt för att avlägsna kväveföreningar i processvattnet. Skanska Sverige AB bygger för närvarande en tunnel i Tanumshede, Gerumstunneln, där den biologiska reningen av processvattnet inte har fungerat under projektet varför denna studie påbörjades. Syftet med detta examensarbete var att bedöma vilka faktorer och parametrar som påverkar den biologiska reningen av processvatten under tunnelkonstruktion med avseende på nitrifikation och denitrifikation. Laborietetester med processvatten från tunnelprojektet utfördes för att uppnå fullständig nitrifikation och denitrifikation.

Tre nitrifikationstester och ett denitrifikationstest utfördes i labb där det första nitrifikationstestet använde processvatten från två olika dagar och slam som samlats in från ett avloppsreningsverk. Nitrifikationsprocessen var ofullständig då nitrat inte ökade trots att det fanns en liten minskning av ammonium och en liten ökning av nitrit. Det andra testet bestod av samma vatten - och slammaterial som Test 1 med undantaget att natriumdivätefosfat tillsattes, vilket ökade fosfatkoncentrationerna so ansågs begränsa reaktionerna. Detta test misslyckades också med att visa någon nitrifierande aktivitet. Alkaliniteten som analyserades i processvattnet var mycket låg och till det tredje testet ökades både alkalinitet och fosfat. Detta test gav de bästa resultaten av nitrifikationstesterna och resulterade i att ammonium omvandlades till nitrit, dock så misslyckades testet med att omvandla nitrit till nitrat. Resultatet från denitrifikationstestet visade en denitrifierande aktivitet när nitrat omvandlades till nitrit samtidigt som totalkväve minskade, vilket indikerar att kvävgas har bildats, dock borde testet ha utförts under en längre tidsperiod. Ytterligare labbanalys av processvatten från tunnlar med hög kvävehalt är viktigt vilket borde fokusera på metallkoncentrationen och proportionerna av slam och processvatten eftersom dessa parametrar inte grundligt analyserats i denna studie.

Nyckelord: kväve, nitrifikation, denitrifikation, processvatten, tunnlar, inhibiterande faktorer, vattenanalys

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PREFACE

This master thesis has been conducted in the master's program *Infrastructure and Environmental Engineering* for Skanska Sweden AB - Infrastructure with the aim of assessing nitrogen treatment during tunnel construction. The thesis was carried out from January to May 2014 with guidance from both Skanska Sweden AB and the department *Water Environment Technology* at Chalmers University of Technology.

I would like to thank my supervisor Britt-Marie Wilén at Chalmers University of Technology for taking the time to help and guide me through the laboratory work. I would like to acknowledge my supervisors at Skanska Sweden AB, Mikael Andersson and Jenny Karlsson. Thank you for giving me the opportunity to conduct my thesis at Skanska Sweden AB and for helping me to select this topic for my thesis. I would also like to acknowledge John Nyberg at Skanska Sweden AB for your optimism, guidance and interest in this topic, thank you. This thesis would not have been completed without the help of Magnus Hallberg at Halfor AB, thank you for your ebullient commitment, guidance and competence throughout this time. Further, I would like to thank the “water boy” Niclas Johansson at Skanska Sweden AB in the project E6 Pålen - Tanumshede. Thank you for all the help with water collection, water samples and analysis at site. I would also like to acknowledge Mats Bäckman and Charlotte Bougart at Hammargårdens treatment plant. A final thank to my opponents Erik Eidmar and Johan Hultman for your valuable input and feedback on my work.

This project has been very interesting and it has contributed to increase my knowledge about the complexity of water contaminants and treatment processes which is a worldwide issue. It is important to keep our waters clean, today and tomorrow, for us and our next generations.

Göteborg, May 2014

Victoria Liljedahl

ABBREVIATIONS

ANFO	ammonium nitrate fuel oil
BOD	biochemical oxygen demand
DO	dissolved oxygen
MLVSS	mixed-liquid volatile suspended solids
NFA	national food agency
NOD	nitrogen oxygen demand
SP	suspended particles
SS	suspended solids
SSE	site sensitive emulsion
STA	the Swedish Traffic Administration
TAN	total ammonia nitrogen
TIN	total inorganic nitrogen
TKN	total Kjeldahl nitrogen
TN	total nitrogen
TOC	total organic carbon
TP	total phosphorus
US EPA	environmental protection agency of the United States
VSS	volatile suspended solids

CHEMICAL COMPOUNDS

CH_3CO_2^-	acetate
$\text{C}_5\text{H}_7\text{NO}_2$	nutrients, biomass
CH_3OH	methanol
$\text{C}_{18}\text{H}_{19}\text{O}_9\text{N}$	heterotrophic organism
CO_2	carbon dioxide
HCO_3^-	bicarbonate
$\text{NH}_4\text{-N}$	ammonium nitrogen
$\text{NH}_3\text{-N}$	ammonia nitrogen
$\text{NO}_3\text{-N}$	nitrate nitrogen
$\text{NO}_2\text{-N}$	nitrite nitrogen

NOTATIONS

A	alkalinity
A_{req}	calculated required alkalinity
c	concentration
e	base of Napierian logarithms
ΔN	increase or decrease of the nitrogen compounds
r	nitrification or denitrification rate
t	time
T	temperature
V	volume
μ_N	maximum specific growth rate of <i>Nitrosomonas</i> d^{-1}

1 INTRODUCTION

Water is an essential part of life and only 2.5 percent of water resources around the world are available freshwater. Sweden is one of few countries in the world where freshwater availability currently is not a big problem. Sweden is covered by approximately 10 percent of inland waters in terms of rivers, streams and lakes (SCB, 2012). There are 119 main watercourses with catchment areas bigger than 200 km² that end up by the coast. There are in total 27 663 watercourses and the total length of all watercourses is 192 000 km (SMHI, 2010). Hence, surface water is not a rarity in the Swedish landscape. But even with sufficient groundwater and surface water, a sustainable management of the freshwater resources is still very important. Especially for future generations as the aim for Swedish waterways is to be ecologically sustainable (Naturvårdsverket, 2014).

Due to this high areal coverage of surface water, the development of infrastructure can be challenging and complex. Infrastructural construction in or near water in Sweden is therefore inevitable. Tunnels are one construction that enables easy access to and from different locations for commute users in the Swedish landscape, both in terms of crossing waterways but also through rock passes. There is, for example, the Tingstad tunnel in Gothenburg that crosses Göta Älv, the Northern Link in Stockholm and the controversial railway tunnel through Hallandsås all of which are developed to increase the infrastructure efficiency. Tunnel construction is increasing in Sweden and there are currently around 20 road tunnels (Vägverket, 2001), which are mostly concentrated to Stockholm, Gothenburg and the West coast and there are also 168 railway tunnels around the nation (Trafikverket, 2014).

There are many types of tunnel constructions in Sweden but the most common and least expensive is a rock tunnel executed by blasting (Vägverket, 2001) where most explosives used today contain ammonium nitrate, sodium nitrate and calcium nitrate as the oxidizing agent (Forsyth, et al., 1995). One issue with the explosives is the significant required concentrations of nitrogen (N) which is transferred or leached through the runoff to recipients near the tunnel construction (Forsyth, et al., 1995). The anthropological impact on the tunnel construction area, which leads to excessive nitrogen concentrations in the runoff water, could if untreated have fatal impacts on the aquatic life as well as declined quality of freshwater sources. With the large areal coverage of waterways in Sweden, the risk of contaminating the water sources during construction work is high and need cautious handling, such as efficient water treatment at the construction site. The water treatment technologies in operation today do not always meet the requirements of different parameters, especially treatment of nitrogen compounds during colder temperatures.

1.1 BACKGROUND

Skanska Sweden AB is currently running a highway project in western parts of Sweden called *E6 Pålen-Tanumshede*. This project started in 2013 and aims to expand and relocate the last phase of the highway E6 which runs from Pålen to Tanumshede. The new stretch of E6 in that area requires a tunnel consisting of two tubes which is performed by blasting. Blasting processes requires explosives which contain nitrogen, and the contaminants in particular are ammonium and nitrate. Nitrogen has become an issue due to its high concentrations in the water runoff from the tunnel construction which mainly leads to water contamination in two ways. The first contamination source is in the front of the tunnel where water is used to wash the blasted rock

masses in order to settle dust and remove blasting residues. The other contamination source is at the disposal site where crystalline nitrogen residues are kept which is infiltrated to the groundwater or transported to the nearby recipients during precipitation. The area has suffered from quite heavy precipitation which has increased the concentration of nitrogen in the runoffs, thus putting higher pressure on the water treatment facility. The biological treatment for nitrogen removal during the project has not worked, which was presumed to be related to low water temperatures. However, other inhibiting parameters could cause the nitrogen removal failure.

1.2 AIM

The aim of this master thesis is to assess which factors and parameters that affect the biological treatment of process water in tunnel construction with regard to nitrification and denitrification. A tunnel project, the Gerum tunnel in Tanumshede, Sweden, is used as a case study due to arisen difficulties of meeting acceptable nitrogen concentration in the recipients. The wastewater is a combination of process water from the tunneling construction, stormwater runoff and leachate water, which becomes contaminated by excessive nitrogen concentrations from explosive residues during blasting and storage. One issue is the water temperature which is below 10°C at which nitrification and denitrification processes are significantly decelerated, which limits the nitrogen removal rate. Thus, the aim is to achieve a complete nitrification and denitrification process in the lab by using the process water from the project.

1.3 SCOPE

This master thesis is carried out for Skanska Sweden AB due to heightened restrictions of nitrogen concentration in process water discharge to recipients during tunnel constructions. The thesis first presents background information regarding nitrogen removal, conventional wastewater treatment and the case study. This is followed by lab analysis descriptions and its results, which in turn is used for calculations of which scale the water treatment facility need to have for different temperatures. The thesis finalizes with a discussion and a conclusion which provides necessary information to achieve sufficient nitrogen removal.

1.4 METHOD

This master thesis comprises of two parts. The first part consists of a literature study with information about the current wastewater treatment method in Tanumshede and other technologies that could potentially be of relevance for tunnel constructions. The theory behind nitrogen is also presented in order to fully understand the different toxic compounds. The second part is a lab analysis where water samples from Tanumshede will be used to characterize the process water's composition and conduct some smaller experiments to assess if the process water can be nitrified and denitrified with activated sludge from a nutrient removal wastewater treatment plant.

1.5 LIMITATIONS

The primary focus will be on nitrogen compounds even though other parameters, such as heavy metals or phosphorus, could be factors that could inhibit the nitrification and denitrification in process water from other tunnel constructions. The nitrogen compounds in focus are ammonium nitrogen, nitrite nitrogen and nitrate nitrogen. Throughout the report when discussing ammonium, nitrite and nitrate, they are always referred to the forms of ammonium nitrogen $NH_4 - N$, nitrite nitrogen $NO_2 - N$ and

nitrate nitrogen $NO_3 - N$, not to be mixed with NH_4 , NO_2 or NO_3 . Thus, if ammonium is mentioned, it is in the form of ammonium nitrogen, otherwise it will be clearly stated.

The lab analysis will be conducted in a smaller scale and cannot fully represent the actual treatment process in full-scale, which is more complex. The lab is also performed during a limited time which could give different results compared to studies under longer periods. The sludge needs to be collected outside of Gothenburg which also limits the time and day for when the lab can be carried out. Due to this geographical limitation, the sludge will be kept for a few days between some experiments and will not always be used the same day as it was collected from the treatment plant. There are geographical limitations for the process water as well since the tunnel project is located approximately 135 km from Gothenburg. The process water will most likely not contain much biological material why the same water will be used throughout all experiments, but it will be stored in a cool place in between.

The focus in the lab will only be on nitrification and denitrification with some alterations between the different tests in order to obtain different results. There are many factors that could contribute to inhibition of nitrification and denitrification, however, since the time period for this thesis is limited, only a few factors will be analyzed. The water temperature could affect the nitrogen treatment and in reality, the temperature is around $10^{\circ}C$ in the tunnel water. However, the tests in the lab are carried out in room temperature to observe if nitrification and denitrification could be achieved at all and to assess other possible factors that could decline the nitrifying and denitrifying activity.

2 NITROGEN THEORY

The largest source of nitrogen on the planet is the atmosphere which contains around 78 percent of nitrogen. Plants are dependent on nitrogen for growth and development and the main source in soils are organic matter, which in turn originates from animal and plant residues. However, plants need inorganic forms of nitrogen, thus bacteria in the soils convert organic nitrogen to inorganic forms before being taken up by the plant roots. Humans and animals eat the plants which are later returned to the soil in residues (Killpack & Buchholz, 1993). There is a deficit of nitrogen in soil which slows down the decomposition process of dead organic substances, which in turn decrease the emissions of greenhouse gases (Golubyatnikov, et al., 2013). The nitrogen cycle is presented in Figure 2.1. This shows that atmospheric nitrogen is transported to the soils and plants. Animal residues decompose to the nitrogen compound ammonium which is nitrified to nitrites and thereafter nitrates before denitrified back to the atmosphere (U.S. EPA, 2012). The nitrification and denitrification process are further described in Section 2.2.1 and 2.2.2. Rain contributes atmospheric nitrogen when it reaches the soil and could reach the groundwater aquifers. Nitrogen from factories producing commercial nitrogen fertilizers could also enter the nitrogen cycle through farming and gardening.

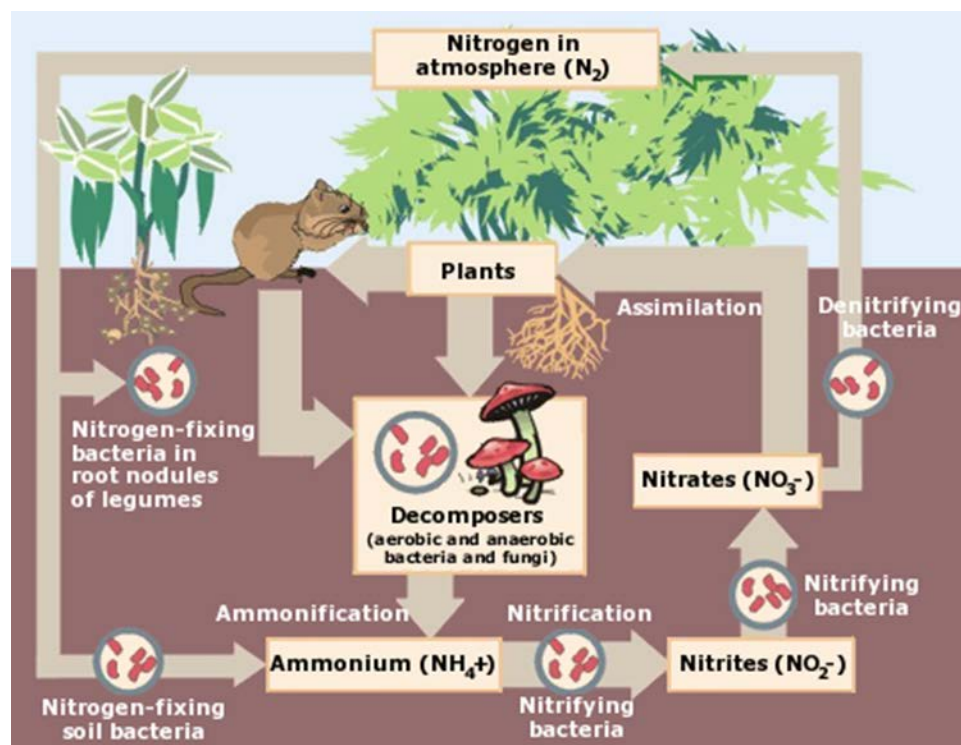


Figure 2.1. The nitrogen cycle (U.S. EPA, 2012)

In tunnel construction it is important to monitor the discharge water from the construction process. There are many parameters that should be carefully observed such as total nitrogen, pH, alkalinity, sulphite, nitrate, nitrite, ammonium, suspended particles and conductivity (Trafikverket & Ramböll, 2012). However, many parameters are treated to designated limits and do often not constitute a problem. The exception is nitrogen contaminations which is the primary nutrient of focus in this master thesis due to high concentrations in the runoffs during tunnel construction. Nitrogen is essential for growth of microorganisms, plants and animals and also in the

synthesis of protein. The principle sources of nitrogen compounds in wastewater are plant and animal origins, sodium nitrate and atmospheric nitrogen. Most nitrogen compounds found in soil and groundwater are biologically originated (Tchobanoglous, et al., 2004) but during tunnel construction, excessive nitrogen compounds originate from the explosives.

2.1 NITROGEN COMPOUNDS

There are several nitrogen compounds of importance but the most common in general are ammonia (NH_3), nitrite (NO_2^-) and nitrate (NO_3^-) (Shrimali & Singh, 2001). The most common compounds in wastewater are ammonia, ammonium, nitrogen gas, nitrite ions and nitrate ions (Tchobanoglous, et al., 2004). Table 2.1 summarizes some of the most common nitrogen compounds where the primary compounds in tunnel construction are ammonium, nitrite and nitrate.

Table 2.1. Different forms of nitrogen and their definitions, modified from (Tchobanoglous, et al., 2004)

Forms of nitrogen	Abbreviation	Definition
Ammonia gas	NH_3	NH_3
Ammonium ion	NH_4^+	NH_4^+
Total ammonia nitrogen	TAN	$NH_3 + NH_4^+$
Nitrite	NO_2^-	NO_2^-
Nitrate	NO_3^-	NO_3^-
Total inorganic nitrogen	TIN	$NH_3 + NH_4^+ + NO_2^- + NO_3^-$
Organic nitrogen	Organic N	$TKN - (NH_3 + NH_4^+)$
Total Kjeldahl nitrogen	TKN	Organic N + $NH_3 + NH_4^+$
Total nitrogen	TN	Organic N + $NH_3 + NH_4^+ + NO_2^- + NO_3^-$

Nitrogen removal is correlated to temperature where the reaction rates increase with temperature. Chang, et al., (2013) showed how the nitrogen removal efficiency changed during the four seasons for nitrate (NO_3^-) and total nitrogen (TN) which turned out to be as expected, the highest removal efficiency during summer and the lowest during winter using the IVCW (integrated vertical constructed wetland) system. Vertical constructed wetlands can have up-flow or down-flow and IVCW combines these two wetlands to improve the water quality. Wetlands consist of plants and some sort of media, e.g. gravel, where the wastewater runs through the media (wetland) (Peng, et al., 2014).

It is important to separate ammonium with ammonium nitrogen for example in terms of concentrations. It is the same nitrogen compound, but when concentrations are analyzed, it differs between the two types. If a concentration is presented as ammonium, a factor is required to convert this concentration to ammonium nitrogen. The atomic mass for nitrogen is 14, hydrogen 1 and oxygen 16. This is used to calculate the factors for each nitrogen compound. These factors are presented in Equations 2.1-2.4.

$$NH_3 \rightarrow NH_3 - N: 1 \times \frac{14}{14+(3 \times 1)} = \frac{14}{17} = 0.82 \quad (\text{Eq 2.1})$$

$$NH_4 \rightarrow NH_4 - N: 1 \times \frac{14}{14+(4 \times 1)} = \frac{14}{18} = 0.78 \quad (\text{Eq 2.2})$$

$$NO_3 \rightarrow NO_3 - N: 1 \times \frac{14}{14+(3 \times 16)} = \frac{14}{62} = 0.23 \quad (\text{Eq 2.3})$$

$$NO_2 \rightarrow NO_2 - N: 1 \times \frac{14}{14+(2 \times 16)} = \frac{14}{46} = 0.3 \quad (\text{Eq 2.4})$$

2.1.1 NITRITE

Nitrite NO_2^- is considered to be quite unstable and is easily oxidized to nitrate. Nitrite is measured colorimetrically and it is an indicator of past pollution. Nitrite is often found in small concentrations, rarely above 1 mg/l in wastewater, but it is none the less very toxic to fish and other aquatic animals. If nitrites are found in wastewater, they are oxidized with chlorine in wastewater treatment plants in order to be reduced (Tchobanoglous, et al., 2004). If nitrite is present in high concentrations in water and if the water is ingested by human or animals, then nitrite can turn haemoglobin to methemoglobin which can cause anoxia and death since methemoglobin cannot transport oxygen to cells (Alonso & Camargo, 2003), also known as the blue baby syndrome.

2.1.2 NITRATE

The most oxidized form of nitrogen in wastewater is nitrate NO_3^- of which the concentrations could be measured using specific-ion electrodes, or as nitrite with colorimetric methods. The U.S. EPA limits nitrate in drinking water to 45 mg/l and the wastewater effluent concentration often varies between 0 and 20 mg/l. High concentrations of nitrates in water can have serious and even fatal effects for infants (Tchobanoglous, et al., 2004).

Camargo et al. (2005) shows that nitrate can be removed by the use of for instance aquatic plants, algae and bacteria but also by using ion exchange, reverse osmosis (RO) and electro-dialysis since the conventional processes such as chlorination, UV and filtration are not viable for nitrate ions (Shrimali & Singh, 2001). However, the treatment processes are quite expensive and some of them rather cause new problems in terms of nitrate disposal.

2.1.3 AMMONIA AND AMMONIUM

Ammonia gas NH_3 is the oxidation state in most organic compounds. Ammonia can be present as either ammonia gas NH_3 or ammonium ion NH_4^+ in aqueous solutions, which depends on the pH. The concentrations of ammonia can be measured colometrically, titrimetrically or with specific-ion electrodes (Tchobanoglous, et al., 2004). The un-ionized molecular ammonia exists in equilibrium with the ionized ammonium dissolved in water which depends on both pH and temperature. The existence of ammonia in environments with a pH below 7 is very low, hence the ammonia ions increases with increased pH (U.S. EPA, 1993). If the concentration of ammonia in waterways is in the range of 0.2-0.5 mg/l, the toxicity to fishes can be fatal (Miladinovic & Weatherley, 2008).

2.1.4 ADVERSE EFFECTS FROM NITROGEN COMPOUNDS

Three major environmental problems have been identified by Camargo & Alonso (2006) of which all include inorganic nitrogen. Eutrophication is one problem,

acidification of ecological ecosystem the second problem and the third is direct toxicity which affects aquatic life's survival, growth and reproduction.

EUTROPHICATION

Increased concentrations of various nitrogen forms in freshwater could generate in eutrophic conditions. Eutrophication presents itself with excessive plant growth and algae blooming which is the result of over-fertilization in water bodies. With increased plant growth, reduced DO and non-clear waters, the organisms in the water body receives less sunlight and less oxygen since it is being depleted by the growing plants and algae, hence the survival of the aquatic life could be critical (U.S. EPA, 1993). If wastewater from tunneling shows increased nitrogen values, the recipient could potentially indicate increased nutrient growth, hence degrading the water quality. Eutrophication in water resources is a worldwide problem in both freshwater and marine environment. In Sweden, the problem is concentrated in the southern parts but some mountain areas indicate eutrophication as well (Naturvårdsverket, 2013). There are many adverse effects on freshwater, some includes increased productivity and biomass of phytoplankton and suspended algae, shifts in phytoplankton composition to toxic species, threats to aquatic species, problems in drinking water supply, depletion of oxygen and decreased recreational values (Smith, 2003). It is not only the aquatic life and environmental surroundings that are affected, humans could also be threatened. When blooming algae dies, algae's toxin can be produced which is harmful to humans. Inorganic nitrogen (ammonia, ammonium, nitrite and nitrate), in particular nitrate, is used in the eutrophication reaction but total nitrogen (TN) is also considered when assessing eutrophication. Nitrite concentration in eutrophic water is also a threat to humans since the product of nitrite nitrification is carcinogenic (Yang, et al., 2008). Nitrate could especially reduce eutrophication if it is decreased (Deng, et al., 2012).

ACIDIFICATION

Nitrogen dioxide NO_2 and nitrogen oxide NO are the main nitrogen acidifying pollutants (sulphur dioxide SO_2 is also a major pollutant) in lakes and streams. These pollutants emit into the atmosphere and can transform by complex reactions into sulfuric acid H_2SO_4 and nitric acid HNO_3 (Baker, et al., 1991). Anthropogenic acidification of streams and lakes could cause many adverse effects e.g. biotic impoverishment of salmonids and invertebrates. Other adverse affects in freshwater are increased accumulation and toxicity of aluminum (Sprenger & McIntosh, 1989), hatching delay of fish and amphibian eggs, increased migration of aquatic insect from their nests, declined zooplankton diversity and reduced growth rates in fish (Camargo & Alonso, 2006).

TOXICITY

Ammonia concentrations of 0.1-1.0 mg/l are often seen as lethal for the wildlife. Biological membranes are permeable to unionized ammonia which makes the toxicity of ammonia a function of pH (Abeliovich, 1992). Ammonia is very toxic to the aquatic wildlife, compared to the non- or less toxic ammonium (Camargo & Alonso, 2006).

Nitrite concentrations on the other hand should not exceed 0.1 mg/l in closed water systems. However, lethal concentrations are much higher, around 250-350 mg/l proved to be lethal to many fish, hence nitrite is less toxic than ammonia but should still be kept under observations (Abeliovich, 1992). Though, other studies show that

nitrite concentrations above 45 mg/l could result in anemia in infants and pregnant women (Odjadjare & Okoh, 2010). The main toxic effect of nitrite on fish is hypoxia which later results in death. This is due to a conversion of pigments former carrying oxygen shifting to a state where they are incapable of carrying oxygen. Other effects of nitrite on fish are cause of electrolyte imbalance, effects on membrane potentials, skeletal muscle contractions and heart function, tissue O_2 shortage and repression of the immune system (Jensen, 2003).

Nitrate toxicity has been considered irrelevant since it must be converted to nitrite before it becomes toxic. The nitrate uptake is much more limited than nitrite uptake which contributes to low nitrate toxicity. However, studies have shown that small concentrations of nitrate can have adverse effects on sensitive aquatic animals during long-term exposure. Further, freshwater animals tend to be more sensitive to nitrate toxicity compared to marine animals (Camargo, et al., 2005).

When assessing the effects of increased nitrogen concentration in drinking water of humans, methemoglobin could be the result from conversion of nitrates to nitrites under anaerobic conditions in the digestive tract (Greer & Shannon, 2005). Symptoms are headache, fatigue, stupor, convulsions and could lead to death. Ingested nitrates and nitrites could be contributing factor of developing cancer of the digestive tract (Fewtrell, 2004). Adverse affects from blooming toxic cyanobacteria on humans have been reported around the world, including Sweden, with effects such as eye irritation, fever, diarrhea, skin rash and muscular cramps. Some cases of human exposure led to lethal outcomes where children are the most sensitive group ((Hitzfeld, et al., 2000) and (Chorus, 2001)).

As mentioned earlier, algae can cause toxicity to aquatic animals and these toxins can either remain in the algal cells or be released into the water. Thus, animals can be directly exposed to toxins through absorption, drinking or ingesting by feeding activities. Cyanobacteria, diatoms and dinoflagellates are the predominate groups that contribute to toxic algae (Camargo & Alonso, 2006).

2.2 BIOLOGICAL TREATMENT FOR NITROGEN

Water in tunnel constructions differ in composition compared to regular household wastewater (e.g. the lack of faecal contaminations). A conventional wastewater treatment plant consists of mechanical, chemical and biological treatment and sometimes disinfection as a final step. Conventional wastewater treatment is necessary as a pre-treatment step for nitrogen removal. The biological treatment step use microorganisms such as bacteria in order to feed on the organic left-over matter from the mechanical treatment step (The World Bank, n.d). Microorganisms feed on nitrogen and around 20 percent is removed by flocculation and separation (Naturvårdsverket, 2009). However, since all nitrogen is not fully removed in biological treatment, a specific nitrogen removal step is added which is used in bigger wastewater treatment plants serving more than 10,000 people and sensitive recipients (Naturvårdsverket, 2009). Ammonia is converted into nitrate by nitrifying bacteria in an aerobic condition after which denitrifying bacteria convert nitrate into nitrogen gas in anoxic conditions (Gryaab, n.d). These nitrifying and denitrifying bacteria can be obtained from sludge in a wastewater treatment plant. The anticipated nitrogen removal efficiency is around 50 to 75 percent (Naturvårdsverket, 2009).

Nitrification and denitrification processes are the basis of all biological nitrogen removal methods at the moment used for wastewater treatment. This has proved to be

both economically and technically feasible for centralized and decentralized systems. Aerobic processes (oxygen rich) nitrify ammonium, i.e. transform ammonium into nitrate where after anoxic processes (oxygen absent) reduce nitrate to nitrogen gas (Oakley, et al., 2010). Ammonium and organic nitrogen are the most common compounds of nitrogen in a modern wastewater treatment plants and is removed by biological nitrification/denitrification (Van Hulle, et al., 2010). Figure 2.2 presents three different nitrification and denitrification processes where a) is a pre-anoxic process starting with denitrification in an anoxic tank with wastewater as the carbon source and is followed by nitrification in the aeration tank. The nitrified effluent is

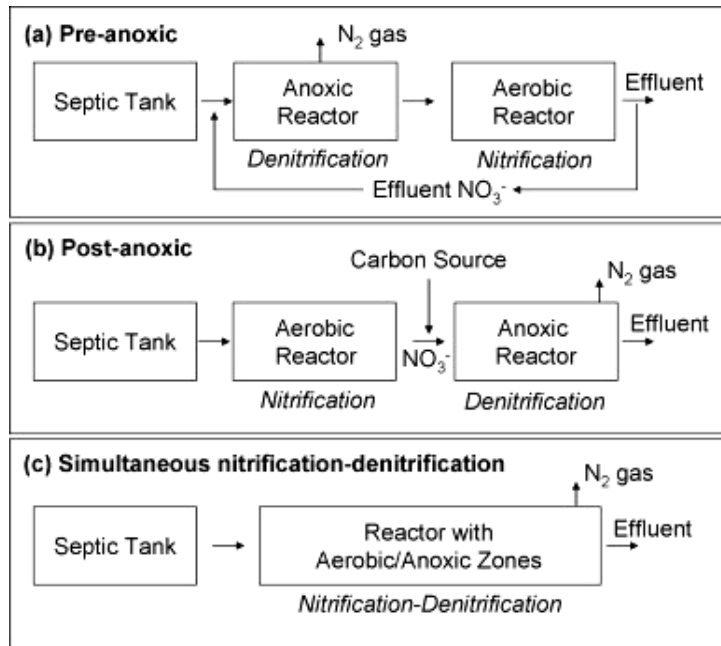


Figure 2.2. Nitrification and denitrification processes, retrieved from Oakley

later recycled back to the anoxic tank. In process b), which is a post-anoxic process, the order is reversed from the pre-anoxic system and the carbon source can be either external sources such as methanol or endogenous respiration of bacterial cells. In c) the nitrification and denitrification occurs simultaneously in the same reactor (Oakley, et al., 2010).

2.2.1 NITRIFICATION

The nitrification process is presented in Reaction 2.1 and 2.2 which describe the two-step biological process of ammonia oxidized to nitrite followed by oxidation of nitrite into nitrate (Tchobanoglous, et al., 2004). Reaction 2.1 shows how ammonia (or ammonium) is converted into nitrite with ammonium-oxidizing bacteria (AOB) such as Nitrosomonas as the most common group of bacteria while Reaction 2.2 converts nitrite into nitrate by nitrite-oxidizing bacteria (NOB) such as Nitrobacter (U.S. EPA, 2002).



Reactions using the AOB-bacteria and NOB-bacteria can also be presented as Reaction 2.3 and 2.4 in the nitrification process which nitrifies ammonium to nitrite and nitrate (Tchobanoglous, et al., 2004).

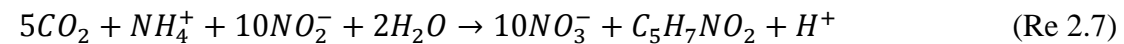
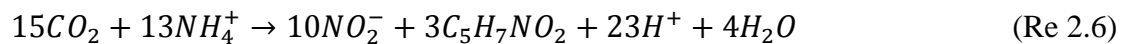


The total oxidation reaction is presented in Reaction 2.5 which shows how ammonium is oxidized into nitrate (Tchobanoglous, et al., 2004).



The nitrification process is not able to remove the toxic nitrogen compound ammonia, but the process converts ammonia into nitrate as stated in the reactions presented earlier. This reduces or eliminates the toxicity to fish as well as reducing the nitrogen oxygen demand (NOD) of the effluent. The rate of the nitrification process in wastewater is a function of time, and also, it is independent of the ammonia nitrogen concentration (Viessman, et al., 2009).

A low growth rate characterizes nitrifying bacteria which is due to the low energy yield, which in turn is connected to the oxidation of ammonia and nitrite. Most nitrifying bacteria are autotrophic and use carbon dioxide as the carbon source, which should be reduced before the carbon reacts with the cell mass. The reduction is performed through oxidation of ammonium and nitrite hence the reaction for growth is shown in Reaction 2.6 and 2.7 (Henze, et al., 1997).



Equation 2.5 is a suggested expression by the U.S. EPA (1993) of the maximum growth rate of *Nitrosomonas* as a function of temperature over the range 5-30°C.

$$\mu_N = 0.47e^{0.098(T-15)} \quad (\text{Eq 2.5})$$

μ_N = maximum specific growth rate of *Nitrosomonas* d^{-1}

e = base of Napierian logarithms, 2.718

T = temperature, °C

Since the temperature is affecting the nitrification, winter aeration need to be much longer than during summer. There are some ways to solve those seasonal issues, the mixed-liquor suspended solids (MLSS) could be increased and pH could be adjusted (Viessman, et al., 2009).

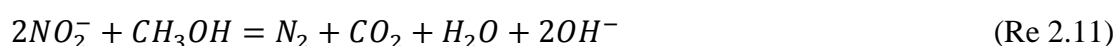
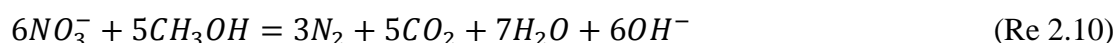
2.2.2 DENITRIFICATION

The denitrification process reduces the oxidized forms of nitrogen to nitrogen gas by heterotrophic bacteria and by using a carbon source of biodegradable organic matter (Mees, et al., 2014). First, there is a biological reduction of nitrate to nitric oxide NO, then nitrous oxide and finally nitrogen gas N₂ (Tchobanoglous, et al., 2004). The end product, nitrogen gas, has no significant effect on the environment (U.S. EPA, 1993). Denitrification occurs when oxygen is depleted, hence the process only occurs in anaerobic or anoxic environments with enough quantities of nitrate and where the oxygen demand surpasses the oxygen supply (Deng, et al., 2012). The nitrification process has limited options for types of bacteria whilst denitrification has a broad range of useful bacteria, which can be both heterotrophic and autotrophic. Some heterotrophic organism genera are *Achromobacter*, *Bacillus*, *Flavobacterium* and *Pseudomonas*, where the latter is the most common denitrifier. Most of the heterotrophic bacteria have the ability to use oxygen, nitrite or nitrate

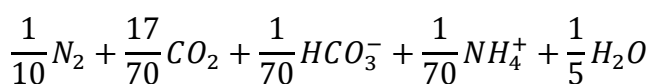
(Tchobanoglous, et al., 2004) though nitrite or nitrate is the wanted forms for the denitrification process. However, if oxygen is present it will be used naturally instead of nitrate and vice versa in anoxic environments. Nitrite and nitrate are considered to be the electron acceptors in the respiratory electron chain, which is the primary mechanism for energy generation in cells. Consequently, nitrite and nitrate replaces oxygen in the transport chain which generates to some extent less energy (U.S. EPA, 1993). A simplified denitrification reaction is seen in Reaction 2.8 (Henze, et al., 2002).



The denitrification process needs an organic carbon source to act as a hydrogen donor and to supply the biological synthesis. Some organic carbon sources could be acetic acid, ethanol, methanol or organic matter. Methanol is preferred common source since it is the least expensive synthetic compound that can be used due to its quality of not leaving residual BOD in the effluent. Methanol first reduces DO in order for the biological reduction of nitrate and nitrite to take place (Viessman, et al., 2009). These different reactions, using methanol (CH_3OH) are presented in Reactions 2.9-2.11.



The energy yielding process for denitrifying bacteria in Reaction 2.12 is a combination of two half-expressions. The process uses organic matter in wastewater as the energy and carbon source (Henze, et al., 1997).



2.2.3 TREATMENT TIME SPANS

The trial spans are quite long for many of the researches in the literature with examples of observation and analyze time of 180, 225 and 550 days for nitrification and denitrification processes (Hoilijoki et al. 2000, Jokela et al. 2002 and Koren et al. 2000). Isaka et al. (2007) had a start-up period of 3 months before the nitrification process was fully working whilst Jokela et al. 2002 had less than 3 weeks of start up for nitrification and around 2 weeks for denitrification, though the entire analysis of nitrification and denitrification proceeded over more than 225 days. Thus the time for nitrification and denitrification can vary significantly.

2.2.4 NITRIFICATION AND DENITRIFICATION RATES

There are different nitrification and denitrification rates presented in several studies, summarized in this section and in Table 2.2 and 2.3. One research using cold mine water had nitrification rates of 29 and 38.3 g N/m³ at 5°C and 10°C respectively. The nitrification rate at 5°C increased to 43 g N/m³ if the salinity was increased. The denitrification rates were 605 and 522 g N/m³ for 5°C and 10°C (Karkman, et al., 2011). Zaitsev et al. (2008) had a nitrification removal rate of 0.54 g NH₄-N/m²-d as the highest at 5°C. The denitrification rate was 4.1 g NO₃-N/m²,d at 5°C. There were also presented other denitrification rates at 3°C and 7°C of 1.5 g NO₃-N/m²-d and 1.6 g NO₃-N/m²-d respectively.

One study performed by Rostron et al. (2001) achieved different nitrification rates for CSTRs (continuously stirred tank reactors) which varied from 2.28 to 4.24 g N/m²-d, media with the synthetic ammonia feed with a concentration of ammonia nitrogen 500 mg/l. There are some other nitrification rates found in the literature using immobilized biomass. With the use of domestic sewage for example, a rate of 0.29 kg N/m³-d was obtained with the ammonia nitrogen concentration of 50 mg/l. Several studies have used synthetic feed water where the rates have been 0.58, 0.2, 0.25, 0.73 and 1.5 kg N/m³-d with respectively 27, 35, 50, 200 and 50 mg/l of ammonia nitrogen (Rostron, et al., 2001). Hoilijoki et al. (2000) reported maximum nitrification rates of 0.13, 0.27±0.06 and 1.78 mg NO₃-N g/VSS.h.at 7, 10 and 24°C in December, August and July respectively. This study was conducted using an aerobically pretreated leachate.

Some in-situ nitrification rates during cold seasons (average of 10°C) varied for different sites from 204 to 548 mg N/m²,d while the denitrification rates in the same season varied from 180 to 491 mg N/m²,d. These rates are based on influent values of 62 mg/l for TN and 60 mg/l for NH₄-N. Sewage with low median strength was used in the study for colder temperatures (Zimmo, et al., 2004). The nitrification rate in sediment in lakes have been assessed which showed to be much higher than in the water columns. The ammonia nitrogen concentration in the sample was 2-10 mg/l and the nitrification rates in the sediments of two different lakes resulted in 0.32 and 0.37 g N/m²-d (Pauer & Auer, 2000).

The nitrification rate was analyzed in leachate from a landfill in Japan which had the characteristics of 400-610 mg/l NH₄-N and 420-650 mg/l TN in the influent water. The effluent water had NH₄-N concentrations of 16-35 mg/l which is a large decrease. The highest nitrification rate was 0.71 kg N/m³-d at 10°C which took up to 3 months to achieve (Isaka, et al., 2007) while Jokela et al. (2002) achieved nitrification rates of 0.05 kg N/m³-d at 5 and 10°C.

Table 2.2. Nitrification rates. 1) (Karkman, et al., 2011), 2) (Rostron, et al., 2001), 3) (Isaka, et al., 2007), 4) (Jokela, et al., 2002), 5) (Zimmo, et al., 2004) and 6) (Pauer & Auer, 2000).

Nitrification							
Tem p °C	Rate	Unit	Type of water	Tem p °C	Rate	Unit	Type of water
5	29 ¹	g N/m ³	Mine water	5/10	50 ⁴	g N/m ³ -d	Leachate water
5	38 ¹	g N/m ³	Mine water	-	290 ²	g N/m ³ -d	Syn. wastewater
10	43 ¹	g N/m ³	Mine water	10	0,204 ⁵	g N/m ² -d	Wastewater
-	580 ²	g N/m ³ ,d	Syn. wastewater	10	0,548 ⁵	g N/m ² -d	Wastewater
-	200 ²	g N/m ³ ,d	Syn. wastewater	-	0,32 ⁶	g N/m ² -d	Lake sediment
-	250 ²	g N/m ³ ,d	Syn. wastewater	-	0,37 ⁶	g N/m ² -d	Lake sediment
-	730 ²	g N/m ³ ,d	Syn. wastewater	-	2,28 ²	g N/m ² -d,med	Syn. wastewater
-	1500 ²	g N/m ³ ,d	Syn. wastewater	-	4,24 ²	g N/m ² -d,med	Syn. wastewater
10	710 ³	g N/m ³ ,d	Leachate water				

Table 2.3. Denitrification rates. 1) (Karkman, et al., 2011), 2) (Zimmo, et al., 2004) and 3) (Zaitsev, et al., 2008)

Denitrification			
Temp °C	Rate	Unit	Type of water
5	522 ¹	g N/m ³	Mine water
10	605 ¹	g N/m ³	Mine water
10	0,18 ²	g N/m ² ,d	Wastewater
10	0,491 ²	g N/m ² ,d	Wastewater
3	1,5 ³	g NO ₃ -N/m ² ,d	Mine water
5	4,1 ³	g NO ₃ -N/m ² ,d	Mine water
7	1,6 ³	g NO ₃ -N/m ² ,d	Mine water

2.2.5 SLUDGE FOR BIOLOGICAL TREATMENT

The sludge concentration varies throughout the literatures in different nitrification and denitrification studies. The study performed by Koren et al. (2000) used 750 ml liquid from activated sludge reactors which was mixed with 16 liters of water, thus around 4.5 percent of the total sample. The aeration flow was 2.4 l/min and the agitation rate was 1 000 rpm. Rostron et al (2001) used nitrifying biomass of 2.5 g VSS/l using CTSR (continuously stirred tank reactors) of 4 liters. Both Jokela et al. (2002) and Hoilijoki et al. (2000) used nitrifying activated sludge of 4 g VSS/l when studying leachate. Another study on wastewater in Hong Kong used a sludge concentration of 2 g MLVSS/l (mixed-liquid volatile suspended solids) in combination with aeration of 8 l/min (Li, et al., 2013).

2.2.6 INHIBITING FACTORS

Nitrification and denitrification can be inhibited for many reasons. Both occurrence and rate of the nitrification process are controlled by environmental and operating conditions, e.g. temperature and sludge age (Viessman, et al., 2009) and other parameters such as phosphate concentration and pH have also been proved to limit the nitrification and denitrification processes. Some of these factors will be considered during the lab analysis.

TEMPERATURE

Nitrification and denitrification processes are strongly dependent on the temperature where the rates of nitrification could double with a temperature increase of 10°C while the denitrification process could double for every increase of 4°C (Zaitsev, et al., 2008). Koren et al. (2000) states that nitrification is affected in temperatures below 10°C since the bacterial metabolism decreases and at temperatures below 4°C, the bacterial metabolism stops all together. However, nitrogen removal is achievable at temperatures below 10°C. The reduction rate is though decreased compared to room temperature of 20°C. One study showed a 30 percent nitrification reduction at 10°C compared to that of 20°C (Alawi, et al., 2009). Jokela et al. (2002), Hoilijoki et al. (2000) and Zaitsev et al. (2007) showed that a nitrification efficiency of higher than 95 percent could be achieved with different loadings. However, other literature found a 100 percent reduction in nitrification at 5°C and pointed out that high nitrification

efficiency in low temperatures can only be achieved with long retention times or a combination of increased nitrifier concentration and elevated temperature if it is possible to heat the water (Ducey, et al., 2010).

The bacteria genera *Nitrospira* is favoured during cooler temperatures and increased dissolved oxygen (Wells, et al., 2009). Adapting the nitrification process to lower temperatures has been an issue in colder northern regions such as Sweden. Some techniques have been tried and proved to be successful at 14°C and even at 7°C. They include extension of the aeration period and supplemental additions of nitrifying biomass in an aeration tank (Ward, et al., 2011). The nitrification rate decreases with about 50 percent for every 10-12°C drop in temperatures above 10°C and when the wastewater is even colder, e.g. decreasing the temperature to 5°C from 10°C, the ammonia oxidation is halved (Viessman, et al., 2009).

NITROGEN COMPOUNDS

Nitrite-nitrogen can inhibit the biological process if the concentration is too high. Nitrite-nitrogen concentration of 150 mg/l proved to inhibit the denitrification process while 1 350 mg/l of nitrate-nitrogen inhibited the denitrification process. However, inhibition of denitrification is not only dependant on nitrite concentration but the biomass concentration. In an experiment using the same nitrite-nitrogen concentration of 20-25 mg/l and a pH of 7-8, the denitrification was only completed with a biomass concentration of 500-1 000 mg/l whilst using a biomass concentration of 100-150 mg/l, the denitrification was inhibited. It was also concluded in this study that a decreased pH generates increased inhibition (Glass, et al., 1997). A critical nitrite-nitrogen dose for nitrification is 50-100 mg/l at a pH 7 which caused inhibition. Moreover, the inhibition lasted more than 10 days after the nitrite disappeared from the solution (Philips & Verstraete, 2000).

Ammonia, i.e. molecular free ammonia, is the major compound responsible for toxicity effects on aquatic life (U.S. EPA, 1993) and it can suppress the nitrifying process. Toxicity can have great impacts on nitrifiers and can kill the nitrifying bacteria. Free ammonia can halt the nitrification process in terms of being inhibiting on the bacteria *Nitrosomonas* at free ammonia concentrations of 10-150 mg/l and at 0.1-1 mg/l for *Nitrobacter* (U.S. EPA, 1993). Other studies show that concentrations of free ammonia higher than 0.2-1 mg/l inhibit the nitrite oxidation (He, et al., 2012).

PHOSPHATE

Phosphorus is most often present in the environment as organic or inorganic phosphate. Phosphate has been reported to limit nitrification, especially during low temperatures. In some nitrifying systems using filters, the phosphate concentrations were dosed 5-50 $\mu\text{g PO}_4^{3-} - \text{P}$ (de Vet, et al., 2012). de Vet et al. (2012) performed a study using filters. The two different tests using filters with 80 $\mu\text{g PO}_4 - \text{P}$ had complete nitrification processes whilst the two other tests with 20 and 30 $\mu\text{g PO}_4 - \text{P}$ resulted in incomplete nitrification. However, with concentration around 100 μg , the nitrification was inhibited as well.

One pilot study by Kors et al. (1998) showed that by dosing with phosphate, the ammonium removal increased in the nitrification process. With phosphate concentrations below 15 $\mu\text{g PO}_4/\text{l}$ in combination with very low temperatures, the nitrification process failed to convert ammonium to nitrate. It was then assumed that the phosphate concentration was a limiting factor for ammonium removal besides the temperature. When the influents in that study were dosed with 100-500 $\mu\text{g PO}_4/\text{l}$,

ammonium removal rates increased despite temperatures around 1°C (Kors, et al., 1998). Another pilot study also proved that phosphate was limiting the nitrification process. When dosing 10 µg $PO_4^{3-}P$, the ammonium removal rate increased and the nitrification process could be completed (van der Aa, et al., 2002).

METALS

Metals could also be inhibiting for the nitrification process. Some metals that have been identified as inhibitors are zinc, copper, chromium, nickel and cobalt (U.S. EPA, 1993). Some inhibiting concentration ranges for certain metals have been identified such as 0.08-0.5 mg/l for zinc and 0.005-0.5 mg/l for copper, thus copper inhibits the nitrite and nitrate production greater than zinc (Juliastuti, et al., 2003). Juliastuti et al. (2003) showed that when the concentrations of heavy metals increased, the inhibition also increased on nitrification. For example, they concluded that a 97 percent inhibition was reached when zinc had a concentration of 1.2 mg/l. Tchobanoglous et al. (2004) also presented some concentrations at which the metals could be inhibiting; 0.25 mg/l nickel, 0.25 mg/l chromium and 0.10 mg/l copper have for example proved to completely inhibit the ammonia oxidation. When it comes to nickel and cadmium, a study showed that both nickel and cadmium had higher inhibiting effects on the ammonium oxidation rate than on the nitrite oxidation rate (Hu, et al., 2002).

pH

The pH can inhibit the nitrification rate. Nitrification should be performed in conditions when the pH is around 6.5-8 but for optimal nitrification rates the pH should be 7.5 to 8.0 according to the U.S. EPA (1993). The percent of the maximum nitrification rates increases as pH increases from 5.0 to 8.0 before the maximum rates starts to decline as the pH continues to increase from 8.0. With pH-values below 6.8 the nitrification rate decline quite notably, the rates could be as low as 10 to 20 percent of the rate at pH 7 when pH decreases to around 5.8 and 6. 90 percent of the maximum rate is occurring at a pH of 7.5 and 8.5, and less than 50 percent of the rate occurs below a pH of 6.4 and above 9.6 (Viessman, et al., 2009). However, values of 7.0 to 7.2 are often used to maintain a reasonable nitrification rate.

An optimal value of pH for denitrification ranges between 7.0 and 9.0 though local conditions can cause variations. If the pH decreases below 7.0 it will affect the end product since nitric oxides, such as N_2O , are produced when pH is declining (Henze, et al., 2002). However, Viessman et al. (2009) claims the optimum pH to be between 6.5 and 7.5 (in accordance with most heterotrophic bacteria) and that the rate is decreased with up to 80 percent of the maximum if pH is reduced to 6.1 or raised to 7.9.

SLUDGE AGE

A research on nitrogen removal in wastewater using biological treatment showed that the sludge age is a factor that affects the biological treatment. The nitrification process was unstable in temperatures lower than 15°C and sludge age lower than 20 days. Though, when the sludge age was higher than 20 days, the colder temperatures had lower influence on the process which was considered to be stable. The sludge age effect on the nitrification process was analyzed during winter (<15°C) and summer (>15°C) conditions which resulted in that the sludge age had no influence on the nitrification process during summer while a sludge age above 20 days eliminated the negative effects of low temperatures on nitrification (Komorowska-Kaufman, et al., 2006).

3 NITROGEN IN TUNNEL CONSTRUCTION

The drill and blast technique is used in the Gerum tunnel which is a conventional tunnel construction method used as the primary technique in Sweden (Vägverket, 2008). The drill and blast technique is carried out in cycles where each cycle contains several steps that are needed to blast a few meters of tunnel. The first step in the drill and blast technique is most often pre-grouting. Groundwater leakages are often expected during tunnel construction why pre-grouting will be conducted regularly. The next step is drilling of boreholes in the required parts of the rock which are loaded with explosives. The boreholes could be approximately 2-6 meters deep but vary depending on the project (Vägverket, 2009). The third step is the actual blasting followed by transportation of the blasted and crushed material. The last step is mucking and scaling under which loose rock is removed at the blasted surface in order to avoid any large blocks of rock to fall down. At times, extra reinforcement is required such as bolting, shotcrete and post-grouting (Grinder, 2003). When one cycle is performed, another cycle begins which is followed by more cycles until the requested length of the tunnel is blasted. The cycles can vary in time due to variations of the rock quality along the tunnel stretch.

3.1 EXPLOSIVES

Both drilling and blasting contribute to dust which is managed by flushing water on the working face in order to settle the dust and reduce health risk for the workers. Dust is thus transported via the water in the tunnel and could be a potential threat for the recipient. The explosive itself during tunnel blasting is responsible for producing several bi-products which also will be mixed with water, hence polluting the wastewater further (Houfeng, et al., 2013). An explosive material can be defined as a compound that reacts very rapidly and generates large quantities of gases in conjunction with liberation of heat (Sudweeks, 1985).

The nitrogen residue from blasting is easily transferred into the water from the working face. Sometimes, for practical reasons, not all explosives are fully detonated and nitrogen in the chemical forms of ammonium NH_4^+ and ammonia NH_3 contaminates water from remains of undetonated explosives (Vikan & Meland, 2012) and nitrates can leach from the explosives in wet blast holes or during charging (Forsyth, et al., 1995). There are different types of explosives, but ammonium nitrate is a common component. Nitrogen dioxide NO_2 is produced with an ammonium nitrate explosive which can be seen in Reaction 3.1 whilst Reaction 3.2 shows how the gas NO_2 , which is freely soluble, is transformed into nitric acid. Ammonium nitrate is an emulsion explosive and the main pollutants are ammonia NH_3 and nitrate NO_3 (Houfeng, et al., 2013).



Spillage of explosives during charging and undetonated explosives accounts for most nitrogen that leaks to recipients near the construction. Blasted rock masses contain the largest part of nitrogen compounds, as much as 60-70 percent of the nitrogen leaked to water comes from these masses (Tilly, et al., 2006). Commercial and modern explosives usually contain a fuel and oxidizer and examples of oxidizing agents are

ammonium nitrate NH_4NO_3 , calcium nitrate $CaNO_3$ and sodium nitrate $NaNO_3$. Explosives of common use can be divided into three groups; ANFO (ammonium nitrate and fuel oil), watergels/slurries and emulsions. All groups of explosives contain nitrogen compounds but with different water resistance, i.e. different solubility in water, hence varying degrees of introducing nitrogen to water systems (Forsyth, et al., 1995). Blasting work in tunnels generally requires more explosives per m^3 rock compared to open pits. Around 1.5-2 kg/m^3 explosives per solid m^3 rock is used during tunnel construction in Sweden and around 50 000 ton of explosives are used yearly in Sweden for mining, tunneling and other activities (Tilly, et al., 2006).

ANFO

The explosive ANFO is constituted by a relationship of around 95 percent ammonium nitrate and 5 percent diesel oil. The nitrogen content is around 34 percent in ANFO (Grinder, 2003) and the two nitrogen compounds ammonia and nitrate ions are very water soluble forms of nitrogen. ANFO has no water resistance why nitrogen is soluble when or if exposed to water. ANFO is also hygroscopic, i.e. absorbs any available water and if ANFO absorbs too much water it becomes de-sensitized and will not detonate (Forsyth, et al., 1995). This in turns leads to explosives being left undetonated in boreholes, hence nitrogen could potentially leach to the construction water.

SLURRY MIXTURES

Slurry mixtures, also known as water gels or dense blasting agents, consist of sensitizer, an oxidizer, water and a thickener (Nichols Jr., 2005). Slurries contain the same soluble nitrogen compounds as ANFO but with the difference that the water resistance is good once the cross-linker has activated the gum. The gelled gum creates an impermeable barrier between oxidizing agents and external water. The nitrogen content is between 20 to 30 percent in the mixture according to Forsyth et al. (1995).

EMULSION

The third group of explosives is emulsion explosives (EMX) which as the two latter groups also is constituted by the water soluble compounds ammonia and nitrate. The water in oil emulsion is very resistant and there is a thin film of oil surrounding a salt solution, which limits the contact with external water. An emulsion mixture contains around 20 to 30 percent of nitrogen (Forsyth, et al., 1995). Emulsion explosives containing no secondary explosives represent most commercial explosives produced today. There are many advantages with emulsion explosives compared to other explosives such as high safety in use, low cost, lower environmental impacts and the possibility of production at the construction site thus reducing transports. The amount of toxic nitrogen from emulsion explosives are 5-15 times less compared to commercial explosives with ammonium nitrate with the secondary explosive TNT (Yunoshev, et al., 2013). Emulsion explosives contain less pure nitrogen compared to ANFO, around 33-75 percent of ANFO's nitrogen content (Tilly, et al., 2006).

The explosive used in the Gerum tunnel is an emulsion explosive called *site sensitised emulsion* (SSE) which become more popular in recent years due to developments of using it underground. The explosive was first introduced by Dyno Nobel in 1995 for underground blasting which is today known as the Titan SSE-system. This emulsion matrix is classified as UN 5.1 oxidizing agent, meaning that it has other restrictions regarding storage and transport compared to conventional explosives (Fauske, 2003). SSE is an emulsion, hence the name, with an aqueous nitrate solution and an oil phase

which is mixed with a chemical sensitizer (Bakke, et al., 2001). The matrix only becomes explosive after a couple of minutes when it has been charged in the borehole together with additive of gas. This system often include a charging truck, two container tanks and pumps for the materials; emulsion matrix and sensitizing agent (Fauske, 2003).

3.2 WATER SOURCES IN TUNNELING

There are several types of water sources during tunnel constructions that in different ways are affected by the tunnel construction in terms of excessive nitrogen concentrations. Some effects such as groundwater contamination can be crucial during the project, but also long-term if the water management is not handled properly. This section presents some different water sources in tunnel construction in order to clarify how they differ amongst each other as well as their relevance in a tunnel construction.

BILGE WATER

During the construction process, inflowing groundwater and stormwater will be collected in excavation shafts which can be contaminated by various pollutants from the construction site (Trafikverket, 2011). This water accumulation is called bilge water and need to be treated before reaching the recipient.

DRAINAGE WATER

Drainage water is groundwater that infiltrates in to the tunnel and needs to be removed in separate drainage pipes (Trafikverket, 2011). The groundwater drops from the sealed rock and are led to drainage pipes by a waterproof membrane. It can also be led to the drainage pipes through the layer of macadam in the bottom of the tunnel (Trafikverket, 2012).

LEACHATE WATER

Leachate is wastewater created by percolation of rainwater and moisture in landfills through different types of waste. Leachate at tunnelling sites contains high concentrations of ammonium nitrogen and organic and inorganic compounds are transported from the waste to recipients (Hasar, et al., 2009). The rock material from blasting has to be stored before it is crushed and used, thus storage areas at the project site are exposed to explosive residues which contains nitrogen compounds. Precipitation infiltrates the landfills and cause leachate water with high nitrogen concentrations.

PROCESS WATER

During tunnel construction in rock, large volumes of process water are generated. The process water is used during drilling of boreholes for blasting, probing and grouting, as well as in cooling processes of the augers (drills). Large volumes of water are also needed to moist blasted rock masses and to wash rock surfaces prior to using shotcrete (Trafikverket & Ramböll, 2012). The process water is often contaminated due to residues in the construction process, such as oil and explosives (Trafikverket, 2011). Thus process water needs proper managing before discharged to recipients in order to reduce the contamination risk to the environment. The process water from the Gerum tunnel will be used in the lab analysis in Chapter 4 and consist of groundwater from the area.

3.3 WATER TREATMENT IN THE GERUM TUNNEL

The first proposed water treatment process was introduced in August 2013 which consisted of pH adjustment, a flocculation chamber, a lamella separator, DynaSand Oxo (Figure 3.1) for nitrification, a carbon source and finally DynaSand filters (Figure 3.2) with mechanical and contact filtration as well as biologically activated filtration. But this treatment process failed to meet the demands from the Swedish Traffic Administration, in terms of nitrogen concentration. Many efforts were done trying to activate the biological treatment, but there were no successful outcomes, which is assumed to be related to low temperatures ($<10^{\circ}\text{C}$). The sludge was collected



Figure 3.1. Dyna Sand Filters (Nordic Water, 2014)



Figure 3.2. Lamella Filters (Nordic Water, 2014)

from a local wastewater treatment plant which could be, in combination with cold water temperatures, one reason for why the nitrogen removal process decelerated. Low water temperatures will be a reoccurring problem for tunnelling construction in Sweden since the tunnel water will at most times be below 10°C (Hallberg, 2014). Since the original water treatment design could not meet the demands of nitrogen concentrations, especially the biological treatment, the treatment process was altered. As from November 2013, the process water is now treated in fewer steps without the biological treatment, hence the current treatment focuses on suspended particles, pH and oil contaminants. This treatment is functioning as a pre-treatment for nitrogen removal, which is considered to be efficient. The process water from the tunnel construction is first led to a container with oil skimmer treatment and then via a 1 m^3 container to a flocculation chamber. After the 1 m^3 container, a manometer is used which is followed by addition of a carbon source if pH needs adjustment. The carbon source is carbonic acid which needs proper mixing before continuing to the next step which is additive of flocculants before the coagulation of particles in the flocculation chamber. After flocculation, the water goes through a lamella separator to remove

sludge. From the lamella separator, the water is transported to a new container before ending up in an artificial dam and the sludge is collected by sludge trucks. The process water is currently recycled back into the tunnel construction due to limited water extraction from the water supply in the area which is around 1.8 m³ per hour. Thus the artificial dam is used, which protects the recipient to some extent. However, construction water outside the tunnel, e.g. rainwater mixed with blasting materials in storage areas, is led to a natural dam before reaching the recipient. The natural dam shows values precisely above allowed nitrogen concentrations. The main idea is to let all water go back to the recipient but since the nitrogen levels have been too high during tunnelling processes, it has to undergo recycling and is currently prohibited from entering natural water sources.

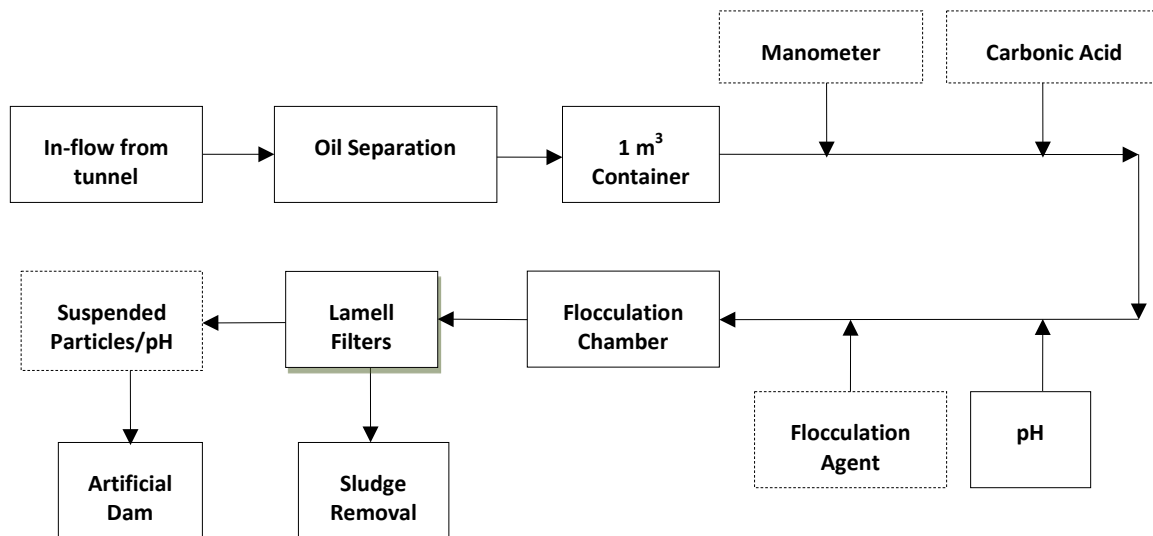


Figure 3.3. Schematic figure of the current water treatment plant for process water.

The current water treatment plant visualized in Figure 3.3 is nonetheless performing efficiently and is not an issue since it keeps the previous mentioned parameters under control. The problem is the absence of biological treatment to reduce nitrogen compounds.

3.4 NITROGEN CONCENTRATIONS IN EFFLUENTS

There is limited information about nitrogen removal and nitrogen concentrations in tunneling operations. However, the contaminated water i.e. the process water can be compared to effluents in mining activities which is also polluted by excessive nitrogen from explosives. There is more literature on mining, which will be used in order to identify a typical range of nitrogen concentrations in effluents.

The Swedish mine company LKAB is used for reference values in terms of outgoing process water. The values have been collected from 2011-2013 at two sites, Kiruna and Vitåfors. The temperature is similar to that in tunnel water with a range of 0-19°C and 1-21°C in Kiruna and Vitåfors with a median value of 3.7 and 7.5 respectively. The range of ammonium nitrogen is 10 to 1 000 µg/l and the median value is 430 µg/l at the site in Kiruna. The range in Vitåfors for ammonium nitrogen is 40 to 2 600 µg/l and the median value is 964 µg/l. Thus, ammonium nitrogen is higher in Vitåfors. The range for nitrate nitrogen is 2 100 to 28 900 µg/l and 2 000 to 49 800 µg/l in Kiruna and Vitåfors respectively and the median for the two sites are 18 400 and 29 550 µg/l. The range for total nitrogen varies between 25 700 to 43 900 µg/l with a median value

of 35 450 µg/l at the site in Kiruna. The same parameter in Vitåfors has a range of 30 300 to 69 300 µg/l and a median value of 54 300 µg/l (Suup, 2014).

One research on biological removal of ammonia and nitrate in Ontario, Canada on mine water had an inlet range of nitrate concentration between of 51 000-405 000 µg/l (which converted to nitrate-nitrogen is 11 730-93 150 µg/l). This was considered to be in the typical range of inlet nitrate concentrations. The general ammonia and nitrate concentrations in mine effluents can vary quite considerably with 10 000-40 000 µg/l for total ammonia and 25 000-300 000 µg/l for nitrate in the effluent water (Koren, et al., 2000). These ranges converted to total ammonia-nitrogen and nitrate-nitrogen is 8 200 to 32 800 µg/l and 5 750-69 000 µg/l respectively. Poláková et al. (2013) used mine water with ammonia-nitrogen concentrations of 10 000-11 000 µg/l and achieved 86 percent removal efficiency. Ammonia is not monitored to the same extent as ammonium, nitrate and nitrite, though it is interesting to know in which range ammonia is usually present in mine- and tunnel water which can be used for comparison. Other ammonia levels in mine water vary in the range of 500-26 800 µg/l (410-21 976 µg/l ammonia-nitrogen) which is based on a project in Finland where cold temperatures were analyzed in relation to the bacterial community. The nitrate concentrations in this project varied between 7 200-52 500 µg/l (1 656-12 075 µg/l nitrate-nitrogen) (Karkman, et al., 2011).

Another research on cold mine water was carried out in Finland in 2007 with water from one gold mine and one chromite mine. This research showed that fixed-bed biofilm reactors could be used to remove ammonium and nitrate in low temperatures. The ammonium-nitrogen concentrations of the two dewatering systems were 24 200 and 17 700 µg/l respectively and 17 500 and 4 300 µg/l for nitrate-nitrogen concentrations. However, ammonium rich water was presented as 50 000-100 000 µg/l why the ammonium concentrations in the two mines could be considered as moderate or low (Zaitsev, et al., 2008).

According to Tilly et al. (2006), bilge water in tunnelling usually has a nitrogen concentration of 100 000 to 1 000 000 µg/l compared to untreated wastewater with 20 000 to 40 000 µg/l. Landfill leachate could also be used as reference values for the process water, however, this water has much higher nitrogen concentrations. The composition of a landfill leachate, Tveta, in Södertälje, Sweden, had the range of 1 100 to 200 000 µg/l ammonium nitrogen between 1994 and 2002. The total nitrogen concentrations during the same period had a range of 900 to 230 000 µg/l (Kietlinska, 2004).

The explosives SSE and ANFO were used in two different tunnels in the project Botniabanan, performed by Skanska Sweden AB, in order to analyze the nitrogen contaminants from different explosives. The Björnböle tunnel used SSE and had an average nitrogen discharge in the process water of 170 000 µg/l but the levels varied from 25 000 to 275 000 µg/l. The total amount of SSE was 573 kg with a specific charge of 2.82 kg/m³ in the first analysis. The second analysis had a total charge of 620 kg SSE which gives a specific charge of 3.05 kg/m³. The Namntall tunnel used ANFO as the primary explosive and the average nitrogen discharge in the process water was 40 000 µg/l but it varied from around 10 000-50 000 µg/l. When the handling of ANFO was improved, i.e. less ANFO outside the boreholes, the average level decreased to 25 000 µg/l. The total charge was 290 kg of ANFO, DynoRex and Dynocord which gives a specific charge of 2.1 kg/m³. Thus, the total use of explosives is higher with SSE compared to ANFO. However, the nitrogen amount discharge using SSE in relation to charged SSE in kilograms is half of that using

ANFO per charged kilogram (Werneman, et al., 2005). Table 3.1 summarizes nitrogen concentrations in mine water, leachate and process water from different literatures and research experiences.

Table 3.1. Summary of nitrogen concentrations from different water samples. 1: (Koren, et al., 2000) 2: (Poláková, et al., 2013) 3: (Karkman, et al., 2011) 4: (Zaitsev, et al., 2008) 5: (Tilly, et al., 2006) 6: (Werneman, et al., 2005) 7: (Kietlinska, 2004) 8: (Jokela, et al., 2002) 9: (Hoilijoki, et al., 2000) 10: (Suup, 2014)

Type	Unit	NH ₃ -N	NH ₄ -N	NO ₃ -N	TN
Mine Water	mg/l			11.73-93.15 ¹	
General Mine Effluents	mg/l	8.2-32.8 ¹		5.75-69 ¹	
Mine Water	mg/l	10-11 ²			
Mine Water	mg/l	0.41-21.98 ³		1.7-12.1 ³	
Mine Water	mg/l		17.7/24.2 ⁴	4.3/17.5 ⁴	
Bilge Water Tunnels	mg/l				100-1 000 ⁵
Untreated Wastewater	mg/l				20-40 ⁵
Process Water SSE	mg/l				170 ⁶
Process Water SSE	mg/l				25-275 ⁶
Process Water ANFO	mg/l				10-50 ⁶
Landfill Leachate	mg/l		1.1-200 ⁷		0.9-230 ⁷
Landfill Leachate	mg/l		160-270 ⁸		
Pretreated Landfill Leachate	mg/l		53-270 ⁹	0.069-0.782 ⁹	53-290 ⁹
Mine Water Kiruna LKAB	mg/l		0.01-1 ¹⁰	2.1-28.9 ¹⁰	25.7-43.9 ¹⁰
Mine Water Vitåfors LKAB	mg/l		0.04-2.6 ¹⁰	2-49.8 ¹⁰	30.3-69.3 ¹⁰

NITROGEN LEVELS IN THE GERUM TUNNEL

Concentrations in the process water from the Gerum tunnel are seen presented in Table 3.2 in order to get an overview of different concentrations. Ammonium rich water for example was set to 50 000-100 000 µg/l (Zaitsev, et al., 2008) but the levels in the Gerum tunnel are higher with its 119 800 µg/l, which must indicate a very ammonium rich water. There are similar results for nitrate, which based on the literature varies from around 5 000-93 000 µg/l though the actual values in the process water from the Gerum tunnel is more than three times higher.

Table 3.2. Nitrogen concentration in the process water

The Gerum tunnel	Unit	NH ₄ ⁺ – N	NO ₂ ⁻ – N	NO ₃ ⁻ – N
2014-03-13	mg/l	119.8	5.98	304
2014-03-31	mg/l	104	5.58	346
2014-04-03	mg/l	111	6.00	386

3.5 GENERAL DISCHARGE DEMANDS

Many municipalities in Sweden have similar restrictions to the most common measured parameters in wastewater. Stockholm, Trollhättan and Malmö vicinity are some examples of areas using guidelines based on Table 3.3 from Svenskt Vatten. However, there could be local variances depending on external environmental factors. Table 3.3 presents parameters that, according to Svenskt Vatten, could affect the treatment processes and the water quality and are applied to industrial process water (Svenskt Vatten, 2012).

Table 3.3. Discharge guidelines for some parameters for industrial process water (Svenskt Vatten, 2012)

Parameter	Unit	Level
pH	-	6.5-10
Lead, Pb	µg/l	50
Cadmium, Cd	µg/l	0
Chromium, Cr	µg/l	50
Nickel, Ni	µg/l	50
Silver, Ag	µg/l	50
Zinc, Zn	mg/l	0.2
Ammonium NH_4^+	mg/l	60
Oil index	mg/l	5-50

The Swedish Food Administration has set out some guidelines for many parameters in drinking water. Table 3.4 presents the guidelines for when the water is unsuitable for drinking water and the guidelines for when the water is potable.

Table 3.4. Guidelines from the Swedish Food Administration (SLV, 2001)

Parameter	Unit	Limit unsuitable	Limit potable
Cadmium	µg/l	5.0	
Copper	mg/l	2.0	0.2
Chromium	µg/l	50	
Nickel	µg/l	20	
Nitrate	mg/l	50	20
Nitrite	mg/l	0.50	0.10
Ammonium	mg/l		0.50

3.5.1 MUNICIPAL VARIATIONS

The municipality of Norrköping has provided guidelines for discharge of stormwater to recipients of different sensitivities. These guidelines apply for stormwater discharge

and are presented in Table 3.5. However, guidelines for process water from different activities could have different and more restricted limits.

Table 3.5. Discharge guidelines from Norrköping municipality (Dagvattengruppen, 2009).

Parameter	Unit	Discharge source		
		From operational activity	Subarea to recipient without protective values	To recipient with protective values
Phosphorus, P	mg/l	0.250	0.175	0.160
Nitrogen, N	mg/l	3.5	2.5	2.0
Lead, Pb	µg/l	15	10	8
Copper, Cu	µg/l	40	30	18
Zinc, Zn	µg/l	150	90	75
Cadmium, Cd	µg/l	0.5	0.5	0.4
Chromium, Cr	µg/l	25	15	10
Nickel, Ni	µg/l	30	30	15
Suspended solids, SS	mg/l	100	60	40
Oil index	mg/l	1.0	0.7	0.4

The guidelines for discharge demands in Gothenburg are referred to the concentration in the discharge point, i.e. the effluents. These guidelines have been developed using several Swedish legislations and environmental goals and are seen in Table 3.6.

Table 3.6. Guidelines for effluent concentration (Göteborgs Stad Miljöförvaltningen, 2013)

Parameter	Unit	Concentration in effluents
Chromium, Cr	µg/l	15
Cadmium, Cd	µg/l	0.4
Lead, Pb	µg/l	14
Copper, Cu	µg/l	10
Zinc, Zn	µg/l	30
Nickel, Ni	µg/l	40
Oil index	mg/l	1.0
pH	-	6-9
Total phosphorus, TP	µg/l	50
Total nitrogen, TN	mg/l	1.25
TOC	mg/l	12
Suspended solids, SS	mg/l	25

3.5.2 DEMANDS IN THE GERUM TUNNEL

Nitrogen concentrations are not always incorporated in the guidelines unless there is a sensitive recipient. There are mainly two surface water sources near the Gerum tunnel which are Pulsebäcken and Gerumsälven. These two recipients are considered as sensitive since the environmental assessment by the Swedish Nature Centre showed presence of many different fish as well as beavers near Gerumsälven (Trafikverket & Ramböll, 2012). An internal analysis of the two recipients showed increased nitrogen concentrations but also increased metal concentration during the construction which is related to the construction work. The Swedish Transport Administration set out maximum concentration values for ammonium, nitrate and nitrite in Pulsebäcken and Gerumsälven in Tanumshede prior to the tunnel project start-up. Table 3.7 presents maximum concentration values for the three parameters which show that Gerumsälven has higher nitrogen limits due to a higher flow.

Table 3.7. Acceptable parameter limits according to the STA (Trafikverket & Ramböll, 2012)

Parameter	Pulsebäcken	Gerumsälven
Ammonium NH ₄ – N	<78 µg/l	<312 µg/l
Nitrate NO ₃ – N	<230 µg/l	<1150 µg/l
Nitrite NO ₂ – N	<15 µg/l	<30 µg/l
pH	6-8	6-8

4 BIOLOGICAL NITROGEN REMOVAL TESTS

The aim with tests in the lab is to try to get the biological treatment to work in terms of nitrification and denitrification of the process water. Nitrification and denitrification tests using water with temperatures below 10°C will not be analyzed, thus the water is at room temperature (19°). The reason for this is that the aim was to first see if it is possible to conduct a complete nitrification test at all at room temperature. The first experiment (Test 1) used only the process water and the nitrifying and denitrifying sludge while the second test (Test 2) used, besides water and sludge, sodium dihydrogen phosphate to compensate any potentially low phosphate concentrations. The third nitrification test (Test 3) increased the phosphate concentration as in Test 2 but the alkalinity was also increased. The reagents needed for the test were process water, sludge and compressed air. Process water was collected from two different days, 2014-03-31 and 2014-04-03. The water was stored in a cool place and then brought to Chalmers. A denitrification test was also carried out which used the process water from 2014-04-03 and a carbon source.

4.1 PREPARATIONS

Before starting the nitrification process, some preparations were needed. The sludge concentration needed to be analyzed, the pH, the alkalinity and the nitrogen concentrations of the process water and sludge liquor had to be known in order to compare the results from the nitrification process, i.e. if there have been any activity during the test. The pH was measured to 6.5 in the process water from 2014-03-31 and 6.4 in the water from 2014-04-03. Process water from the two different sampling days was poured into three 50 ml test tubes each. These samples were frozen until they could be analyzed in the ion chromatograph which will give the concentrations of ammonium, nitrite and nitrate.

The alkalinity was decided for the two different samples of process water from 2014-03-31 and 2014-04-03 by the use of titration. 50 ml water each from the two days was used. Three drops of the indicator solution were added which turned the solution turquoise (at a pH of 5.4). The aim of this method (Standard Methods, EN ISO 9963-1:1995) is to titrate the turquoise solution until it becomes transparent. The titration solution used in this test was 0.02 M hydrochloric acid. The required volume of hydrochloric acid in the water to obtain a transparent color was 5.4 ml for the process water from 2014-03-31 and 5.3 ml for the water from 2014-04-03. When these volumes have been determined, the alkalinity can be calculated using Equation 4.1. The alkalinity for the two different water samples is calculated in Equation 4.2 and 4.3.

$$A = \frac{c(HCl) \times V_5 \times 1000}{V_4} \quad (\text{Eq 4.1})$$

A = Alkalinity, mM/l

$c(HCl)$ = Concentration of the hydrochloric acid, M/l

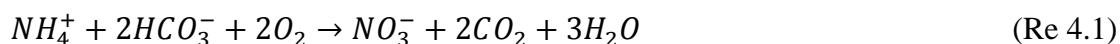
V_4 = Volume of the water sample, ml

V_5 = Volume of the hydrochloric acid consumed by the sample, ml

$$A(2014/03/31) = \frac{0.02 \times 5.4 \times 1000}{50} = 2.16 \frac{\text{mM}}{\text{l}} \times 61 = 131.76 \text{ mg/l} \quad (\text{Eq 4.2})$$

$$A(2014/04/03) = \frac{0.02 \times 5.3 \times 1000}{50} = 2.12 \frac{\text{mM}}{\text{l}} \times 61 = 129.32 \text{ mg/l} \quad (\text{Eq 4.3})$$

The stoichiometry for nitrification and alkalinity is seen in Reaction 4.1 (Tchobanoglous, et al., 2004). For every mole of ammonium nitrogen to be transformed to nitrate nitrogen, two moles of HCO_3^- are required.



However, for this test it is assumed that 2.5 mole of alkalinity is required per one mole ammonium nitrogen to make sure that the nitrification process is not limited by alkalinity. This relation was used at Ryaverket in Gothenburg and was used to see if the alkalinity was high enough for the ammonium nitrogen concentrations.

PROCESS WATER 2014-03-31

$$A = 2.16 \text{ mM}$$

$$\text{NH}_4 - \text{N} = 104 \frac{\text{mg}}{\text{l}} \Rightarrow \frac{104}{14} = 7.43 \text{ mM} \quad (\text{Eq 4.4})$$

$$A_{\text{req}} = 7.43 \times 2.5 = 18.58 \text{ mM} \quad (\text{Eq 4.5})$$

The required alkalinity for a $\text{NH}_4 - \text{N}$ concentration of 104 mg/l is 18.58 mM according to calculations in Equation 4.4 and 4.5, but the real alkalinity is only 2.16 mM. Thus, the alkalinity is too low and could inhibit the nitrification process.

PROCESS WATER 2014-04-03

$$A = 2.12 \text{ mM}$$

$$\text{NH}_4 - \text{N} = 111 \frac{\text{mg}}{\text{l}} \Rightarrow \frac{111}{14} = 7.93 \text{ mM} \quad (\text{Eq 4.6})$$

$$A_{\text{req}} = 7.93 \times 2.5 = 19.83 \text{ mM} \quad (\text{Eq 4.7})$$

The alkalinity was calculated for this process water in Equation 4.6 which was 7.93 mM. The calculated required alkalinity in Equation 4.7 was 19.83 mM, thus the alkalinity in the water is too low in this sample which could limit the nitrification activity.

SUPERNATANT 2014-05-02

The supernatant is the top layer liquid produced when the bigger particles of the sludge is allowed to settle. The alkalinity for the supernatant from May 2 was determined using the same method as for the process waters with a 50 ml sample and a 0.02 M acid. The required volume of the hydrochloric acid to make the sample go from turquoise to transparent was 5.8 ml. The alkalinity of the sludge is seen in Equation 4.8:

$$A(2014/05/02) = \frac{0.02 \times 5.8 \times 1000}{50} = 2.32 \frac{\text{mM}}{\text{l}} \times 61 = 141.52 \text{ mg/l} \quad (\text{Eq 4.8})$$

4.2 NITRIFICATION TEST 1

The first nitrification test was conducted on April 7 2014 where 1 500 ml samples of process water from the two days were mixed with 500 ml sludge using a magnetic stirrer. The solution was continuously aerated throughout the experiment. A total number of 26 samples, each 20 ml in size, were collected from the two solutions according to the schedule in Table 4.1. Each 20 ml sample was first centrifuged 4 000 RCF for 5 min. Thereafter the samples were filtered through 0.45 μm and stored in the refrigerator. These samples were later frozen for analysis due to maintenance problems of the ion chromatograph. The temperature was around 19°C in the two containers during the nitrification test.

Table 4.1. Sampling scheme for nitrification
Test 1

Time	Process water 2014-03-31	Process water 2014-04-03
Min	Samples	Samples
0	0	0
15	1	1
30	2	2
45	3	3
60	4	4
90	5	5
120	6	6
150	7	7
180	8	8
240	9	9
300	10	10
360	11	11
1260	12	12

The nitrification test and equipments are seen in Figure 4.1. In order to determine the concentration of ammonium, nitrate and nitrite, an ion chromatograph is used which measures all samples taken during the nitrification test. Each sample takes 20 minutes



Figure 4.1. Nitrification test in the lab.

and with 31 samples, including two tubes of MQ water, the analysis was run for approximately 10 hours. However, there were certain preparations required before using the ion chromatograph. The 26 samples from the nitrification test were diluted 5 times with 2 ml of the sample and 8 ml MQ water. Three more samples were run in the chromatograph, the filtered samples of the process water and the sludge which gives a total of 29 samples. The first and last sample in the ion chromatograph is always filled with only MQ water which is used as a calibration to see that the instrument is working properly.

4.2.1 SLUDGE CONCENTRATION TEST 1

The sludge was collected from Hammargårdens treatment plant in Kungsbacka on April 7 (for Test 1 and 2) and May 2 (for Test 3 and the denitrification). This sludge consists of both nitrifying and denitrifying bacteria which are required for a nitrification and denitrification processes. The sludge concentration was approximately 4.6-5 g/m³ or 4 600-5 000 mg/l on April 7 and 2.3 mg/l on May 2 according to the instrument at site, however, the concentration was measured during the lab at Chalmers as well to obtain a more accurate concentration.

First, the sludge was allowed to settle for a few minutes in order to remove most of the supernatant. 5 ml samples were collected of the sludge before weighing 1.6 µm filters which was required for the calculations of sludge concentrations. The 5 ml samples were then filtered through the 1.6 µm filters after which the filters were heated in the oven in 105°C for 2.5 hours for filter 1 and 2 (the minimum time for heating is 2 hours). The filters were cooled before being weighed a second time. The sludge sample from Test 2 was diluted with the process water, thus lower concentration. The filters were then heated in the oven again, but this time in 550°C for 30 min in order to measure the organic material content. Suspended solids could be calculated if the weight of the filter before and after heating in 105°C is known. The following equation is used:

$$\frac{X_{dry}-X_{wet}}{v} \quad \text{(Eq 4.9)}$$

X_{dry} = the weight of the filter after 2 hours heating in 105°

X_{wet} = the weight of the filter before heating

v = volume of the sample, l

The weight of the filters and the suspended solids are presented accordingly:

Filter 1

- Before usage 0.0897 g
- After heating in 105°C 0.1241 g
- After heating in 550°C 0.0946 g

$$\text{Suspended solids} \frac{0.1241-0.0897}{0.005} = 6.88 \frac{g}{l} \Rightarrow \frac{6.88}{4} = 1.72 \text{ g/l}$$

Filter 2

- Before usage 0.0888 g
- After heating in 105°C 0.1249 g
- After heating in 550°C 0.0941 g

$$\text{Suspended solids} \frac{0.1249-0.0888}{0.005} = 7.22 \frac{g}{l} \Rightarrow \frac{7.22}{4} = 1.81 \text{ g/l}$$

The nitrification rates can now be calculated for the two filters since the sludge concentrations is known. However, since this test failed to achieve a nitrification process, the rates will not be relevant.

A 50 ml sample of the supernatant was also filtered through 1.6 μm on April 7 which was frozen for further analysis of the sludge content. The pH of the sludge, measured in the supernatant on April 7, was 6.84. The sludge age is not known but was approximated to be between 15-20 days according to the staff at Hammargården.

4.2.2 RESULTS TEST 1

The results from the ion chromatograph fluctuated and did not present values for all samples. The ion chromatograph turned out to give invalid results, this was determined after the instrument was run with some standards solutions. These samples were however kept and frozen until a new analysis was carried out when the instrument was repaired. A second analysis with the samples from April 7 was run on April 15 under correct conditions of the instrument. The concentration of phosphate was insignificant and below the detection limit, which could be the reason for why the biological treatment failed at site in Tanumshede.

The theoretical sample 0 (start concentration after mixing process water with activated sludge suspension) is calculated to compare with the results from the ion chromatograph using Equation 4.10.

$$\text{Sample } 0_x = \frac{1.5 \times P + 0.5 \times S}{2} \quad (\text{Eq 4.10})$$

Sample 0_x = the first sample in the nitrification test

P = concentration in the process water, mg/l

S = concentration in the supernatant, mg/l

The theoretical samples for ammonium, nitrite and nitrate are calculated for the process waters from 2014-03-31.

$$\text{Sample } 0_1 (\text{NH}_4 - \text{N}) = \frac{1.5 \times 104 + 0.5 \times 5}{2} = 79.3 \text{ mg/l}$$

$$\text{Sample } 0_1 (\text{NO}_2 - \text{N}) = \frac{1.5 \times 5.6 + 0.5 \times 1.6}{2} = 5.4 \text{ mg/l}$$

$$\text{Sample } 0_1 (\text{NO}_3 - \text{N}) = \frac{1.5 \times 346 + 0.5 \times 2.9}{2} = 261.7 \text{ mg/l}$$

When comparing the theoretical values with the analyzed values, 97.2, 5 and 305.6 mg/l respectively, it is clear that there have been some dilution faults. The theoretical first values of the samples using process water from 2014-04-03 are also calculated.

$$\text{Sample } 0_1 (\text{NH}_4 - \text{N}) = \frac{1.5 \times 111 + 0.5 \times 5}{2} = 84.5 \text{ mg/l}$$

$$\text{Sample } 0_1 (\text{NO}_2 - \text{N}) = \frac{1.5 \times 6 + 0.5 \times 1.6}{2} = 4.9 \text{ mg/l}$$

$$\text{Sample } 0_1 (\text{NO}_3 - \text{N}) = \frac{1.5 \times 386 + 0.5 \times 2.9}{2} = 290.2 \text{ mg/l}$$

The values from the ion chromatograph show 71.7, 4.6 and 191.2 mg/l respectively which also indicate that there are some errors with the dilution. The results from the ion chromatograph were therefore normalized against sodium (anion) and chloride (cation) to get more accurate results. These compounds are expected to be constant during the experiment and by normalizing the data with these concentrations the errors in the measurements should be eliminated. Normalization using sodium had the best correlation and is presented in Figure 4.2.

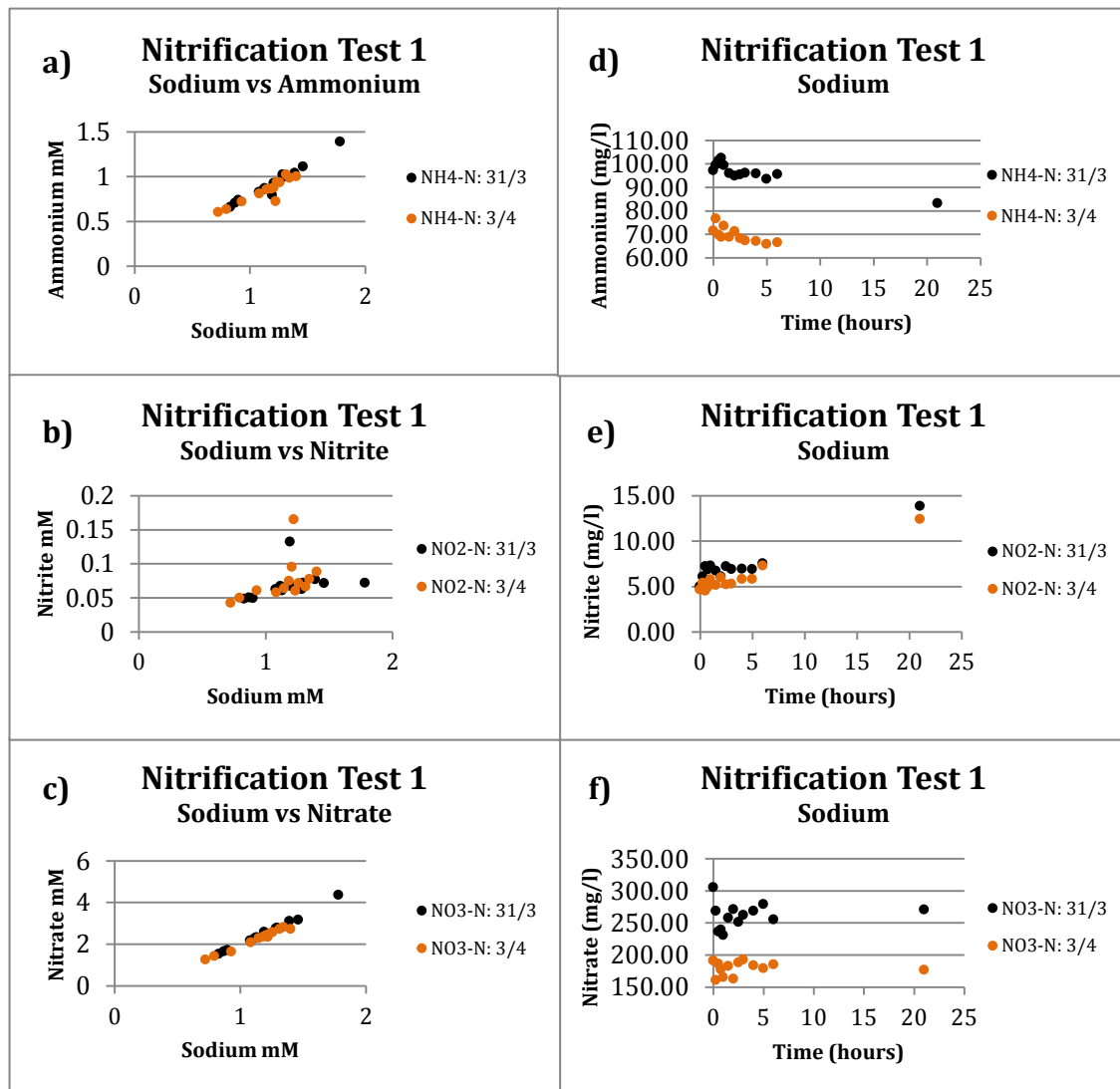


Figure 4.2. Results from Nitrification Test 1. Normalized values in figures a-c show how well sodium is correlating with the compounds. Figures d-f show the trend for ammonium, nitrite and nitrate.

The normalization was carried out by analyzing the ammonium, nitrite and nitrate concentrations in relation to sodium and chloride in order to consider the potential dilution errors. Thus the zero sample of the sodium concentration of the process water was used for samples when normalizing with sodium. The first (sample zero) was divided with the sodium concentration of the next sample X_n and then multiplied with the ammonium concentration of that same sample, seen in Equation 4.11. The first sodium sample is then divided with the third sodium sample X_{n+1} and multiplied with the third ammonium sample. The same procedure is performed with nitrite and nitrate and with chloride as the normalizing agent. Sodium and ammonium is thereafter

plotted against each other which shows if the two correlates well i.e. a linear line should be observed in the diagram.

$$\text{Ammonium conc} = \frac{\text{Sample 0 (sodium)}}{\text{Sample } X_n \text{ (sodium)}} \times \text{Sample } X_n \text{ (ammonium)} \quad (\text{Eq. 4.11})$$

All results for sodium and chloride are presented in Appendix 1 in larger scales figures. The first diagrams in the appendix show the time plotted against the concentration with the dilution errors. It shows a clear increasing trend for nitrite but not a decreasing trend for ammonium, which should be the case. However, when the values were normalized, ammonium showed clear decreasing trends when using sodium and chloride. Nitrite had increasing trends for the sodium and chloride. This indicates that nitrification is in progress since the goal with nitrification is to decrease

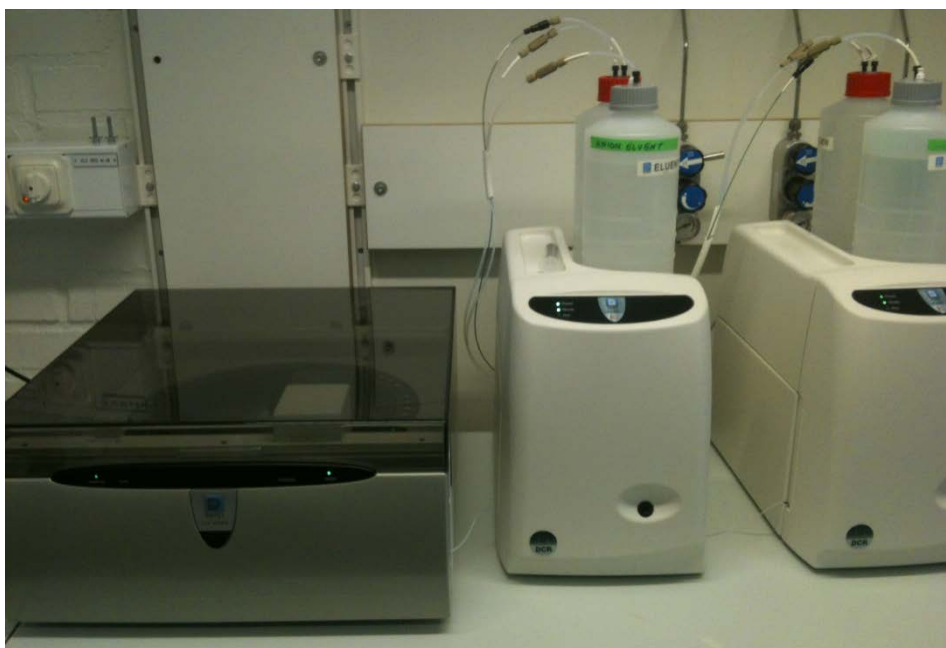


Figure 4.3. Ion chromatograph

ammonium to nitrite before being transformed to nitrate. When analyzing the nitrate concentration, there is no clear trend that shows an increasing concentration, which is the result of an incomplete nitrification. The diagrams that show the normalizing of the values from the ion chromatograph using sodium and chloride are added in Appendix 1. Both sodium and chloride show good correlation but the sodium diagrams tend to have more similar results between the two types of process water. Thus, the actual decrease or increase for ammonium, nitrite and nitrate should be based on the diagrams using sodium concentration for normalization. This is shown in Figure 5.3. Furthermore, a decrease in ammonium should correspond to an increase in nitrite. This is not applied for the normalized values with neither sodium nor chloride. This further indicates an incomplete nitrification. The ion chromatograph is seen in Figure 4.3.

TOC (total organic carbon) and TN (total nitrogen) were also analyzed. TOC was analyzed in order to know if there were high concentrations of organic material that could be of importance for the test and possibly affect the nitrogen concentrations in the process water. Figure 4.4 shows the TOC concentrations which are quite low. The increase in TOC concentration could possibly due to break-up of the biomass.

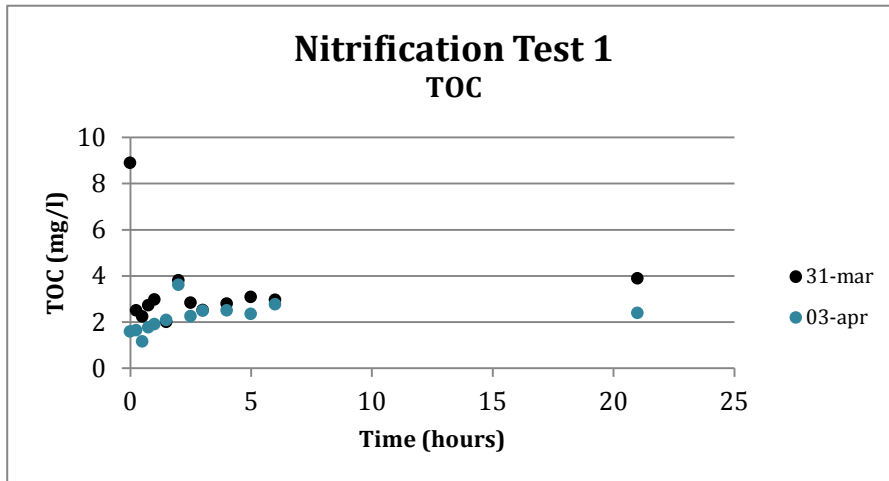


Figure 4.4. Results TOC Nitrification Test 1

The concentrations of TN are presented in Figure 4.5. The concentration of TN should be on a similar level throughout a nitrification test since ammonium should be transformed into nitrate. However, in Test 1 the concentration of TN varied with a slight trend of increasing which could be due to disintegration of sludge flocs during the experiment.

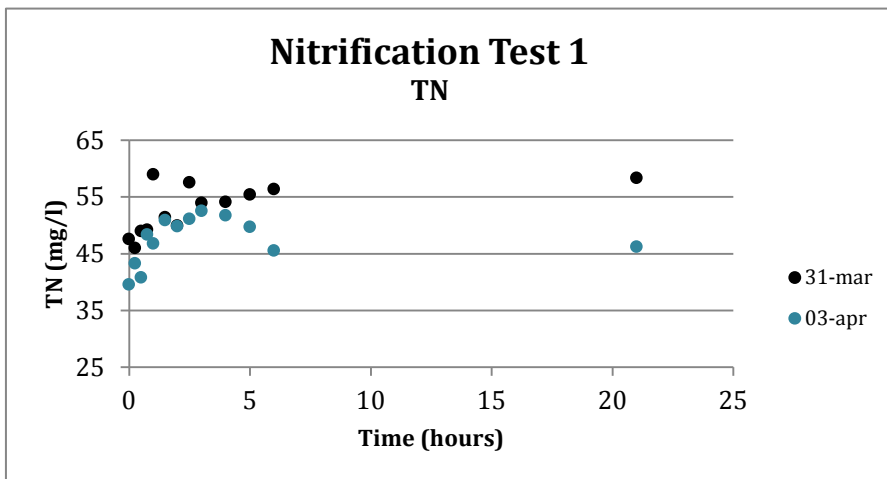


Figure 5.5. Results TN Nitrification test 1

4.3 NITRIFICATION TEST 2

This nitrification test was carried out on April 10 using the same method as Test 1. However in this test, 0.078 g sodium dihydrogen phosphate was added due to the low phosphate concentrations in Test 1 (which were below detection limit). This gives a phosphorus concentration of 10 mg/l and could help the nitrification since the required phosphate concentration could be too low. Samples were taken out according to the schedule in Table 4.2 and the pH was measured. All samples were frozen after Test 2 for further analysis. The temperature was around 16-17°C throughout the experiment.

Table 4.2. Sampling scheme for nitrification Test 2

Time	Process water 2014-03-31		Process water 2014-04-03	
	pH	Sample	pH	Sample
0	6.9	0	6.84	0
15	-	1	-	1
30	-	2	-	2
45	-	3	-	3
60	8.31	4	8.25	4
90	-	5	-	5
140	-	6	-	6
170	-	7	-	7
220	8.31	8	8.25	8
1030	8.01	9	6.56	9

4.3.1 SLUDGE CONCENTRATION TEST 2

The sludge concentration in Test 2 was determined by the same method as for Test 1 with the only difference that the 5 ml sample was taken from the mixed solution in this test (process water and sludge). Filters 3 and 4 were heated in the 105°C oven around 14 hours (they were kept in the oven overnight) and for 30 min in the 550°C oven.

Filter 3

- Before usage 0.0908 g
- After heating in 105°C 0.1020 g
- After heating in 550°C 0.0936 g

$$\text{Suspended solids} = \frac{0.1020 - 0.0908}{0.005} = 2.24 \text{ g/l}$$

Filter 4

- Before usage 0.0878 g
- After heating in 105°C 0.0962 g
- After heating in 550°C 0.0885 g

$$\text{Suspended solids} = \frac{0.0962 - 0.0878}{0.005} = 1.68 \text{ g/l}$$

4.3.2 RESULTS TEST 2

The ion chromatograph was used on April 22 for the samples from the Nitrification Test 2. The theoretical sample 0 for ammonium, nitrite and nitrate are calculated for the process waters from 2014-03-31 using the same equation as in Section 4.2.2. The

process water samples (not from the nitrification solution) were mixed with HNO₃ which is the reason for the high results below.

$$\text{Sample } 0_2 (\text{NH}_4 - \text{N}) = \frac{1.5 \times 132.9 + 0.5 \times 5}{2} = 100.9 \text{ mg/l}$$

$$\text{Sample } 0_2 (\text{NO}_2 - \text{N}) = \frac{1.5 \times 3.7 + 0.5 \times 1.6}{2} = 3.2 \text{ mg/l}$$

$$\text{Sample } 0_2 (\text{NO}_3 - \text{N}) = \frac{1.5 \times 26\,800 + 0.5 \times 2.9}{2} = 20\,100.7 \text{ mg/l}$$

When comparing the theoretical values with the analyzed values, 129.6, 6.3 and 473.3 mg/l respectively, it is clear that there have been some dilution faults. The theoretical first samples from 2014-04-03 are also calculated as previous.

$$\text{Sample } 0 (\text{NH}_4 - \text{N}) = \frac{1.5 \times 121.5 + 0.5 \times 5}{2} = 92.4 \text{ mg/l}$$

$$\text{Sample } 0 (\text{NO}_2 - \text{N}) = \frac{1.5 \times 4.1 + 0.5 \times 1.6}{2} = 3.5 \text{ mg/l}$$

$$\text{Sample } 0 (\text{NO}_3 - \text{N}) = \frac{1.5 \times 28\,508 + 0.5 \times 2.9}{2} = 21\,381.7 \text{ mg/l}$$

The values from the ion chromatograph show 109.6, 5.6 and 354.8 mg/l respectively which also indicate that there are some errors with the dilution. However, by normalizing these values, new accurate values could be retrieved. Figure 4.6 show how the three nitrogen compounds have been normalized against both sodium (anion) and chloride (cation) as well as the originally concentration, plotted against time, which showed no clear trends for any of the compounds. Ammonium shows a slight decreasing curve for both sodium and chloride, in particular for the process water from 2014-03-31 with lower concentrations. Nitrite is first increasing for the process water from 2014-03-31 but shows decreased values in the last point. The process water from April 3 shows a clearer increasing trend. Nitrate has no clear trend for either of the process waters. Figure 4.6 show how sodium and chloride are correlated to ammonium, nitrite and nitrate. Normalization with sodium results in a better correlation for all three compounds and should, as in Test 1, be used to analyze the activity of the nitrification process. Appendix 2 shows the diagrams in larger scale and the normalization with chloride as well as the original plots with concentration plotted against time. Since there were no clear trend for ammonium and nitrate, this also indicates that the nitrification in Test 2 was incomplete. As observed in Test 1, the decrease in ammonium should correspond to the increase in nitrite or nitrate. But when observing the diagrams in Appendix 2, it is clear that the amount of ammonium does not follow the increase in nitrite. This further proves that the nitrification was not completed.

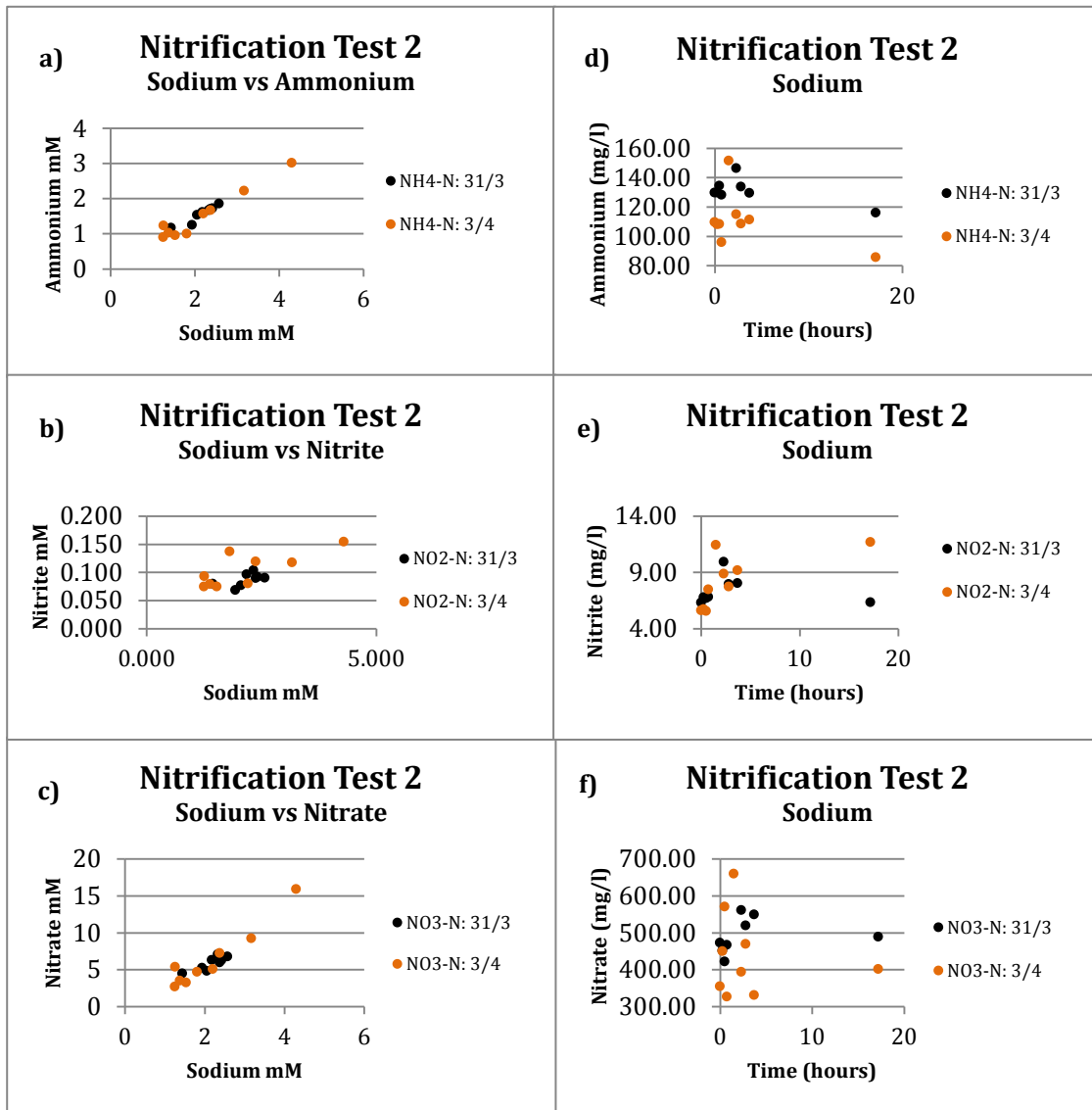


Figure 4.6. Results from nitrification test 2. Figures a-c show how ammonium, nitrite and nitrate correlate with sodium while figures d-f shows the results for the three compounds.

The results from TOC and TN are presented in Figures 4.6 and 4.7.

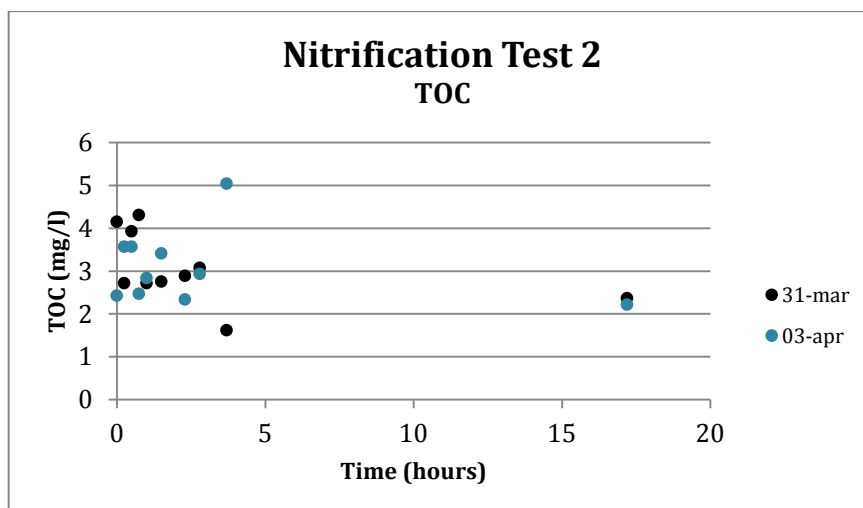


Figure 4.6. Results TOC Nitrification Test 2

TOC is still low in concentration and varies throughout the test. TN seems to first decrease before it increases again, thus there is no complete nitrification in this test as already declared.

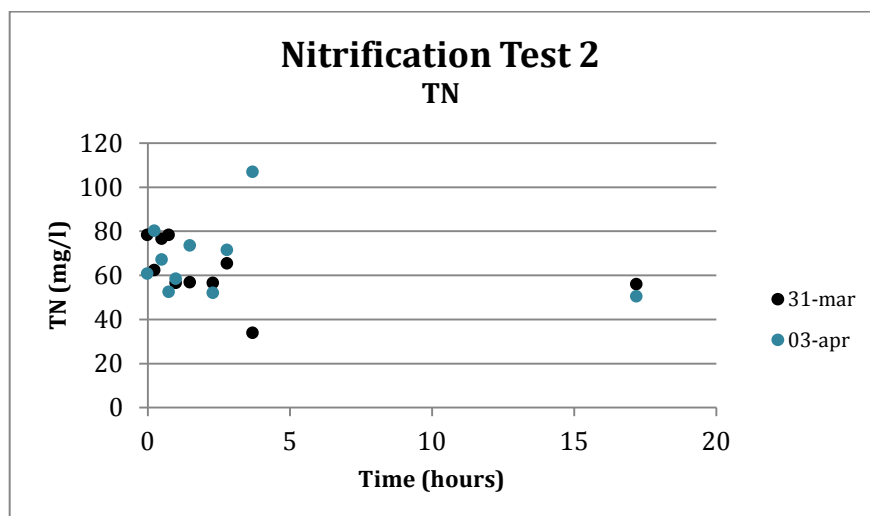


Figure 4.7 Results TN Nitrification Test 2

4.4 NITRIFICATION TEST 3

Since neither Test 1 nor Test 2 resulted in any evident nitrification activity, another test was carried out, Test 3. It was suspected from the alkalinity tests that the alkalinity could be too low which was considered in this test. The proportion of the process water and sludge was altered in this test to see if that could have been a factor. The solution for the nitrification consisted of 1 000 ml process water and 1 000 ml sludge. The phosphate was, as in Test 2, increased by adding 0.078 g sodium dihydrogen phosphate in each container. The alkalinity was increased in this test by adding 1.34 g sodium hydrogen carbonate which was determined by looking at the ammonium concentration of the process water and the stoichiometry of alkalinity (estimated that one mole of ammonium nitrified consumes two moles of alkalinity). The following calculations were made in order to determine the required volume of sodium hydrogen carbonate $NaHCO_3$ to increase the alkalinity to meet the ammonium nitrogen concentrations:

- Process water March 31

$$\frac{NH_4-N}{2} = \frac{104}{2} = \frac{52 \frac{mg}{l}}{14} = 3.7 \text{ mmole} \quad 3.7 \times 2 = 7.4 \text{ mmol}$$

- Process water April 3

$$\frac{NH_4-N}{2} = \frac{111}{2} = \frac{55.5 \frac{mg}{l}}{14} = 4 \text{ mmole} \quad 4 \times 2 = 8 \text{ mmol}$$

The test was run for 22 hours and 15 min with a total of 12 samples per container. Samples were collected at continuous intervals presented in Table 4.3 with the majority of the samples collected during the first 5 hours.

Table 4.3. Sampling scheme nitrification Test 3

Time	Process water 2014-03-31		Process water 2014-04-03	
	pH	Sample	pH	Sample
0	7.83	0	7.82	0
15	-	1	-	1
30	-	2	-	2
45	-	3	-	3
60	8.29	4	8.16	4
90	-	5	-	5
120	-	6	-	6
150	-	7	-	7
180	8.26	8	7.98	8
240	-	9	-	9
300	8.14	10	7.85	10
1335	7.79	11	7.97	11

4.4.1 SLUDGE CONCENTRATION TEST 3

The sludge concentration was determined with the same method that was used in Test 1. The 5 ml sample that was filtered was taken from the sludge solution after it had been thickened i.e. much of the supernatant had been removed. Filters 5 and 6 were heated for 2.5 hours in 105°C and 30 min in 550°C.

Filter 5

- Before usage 0.0949 g
- After heating in 105°C 0.1505 g
- After heating in 550°C 0.0885 g

$$\text{Suspended solids: } \frac{0.1505 - 0.0949}{0.005} = 11.12 \frac{g}{l} \Rightarrow \frac{11.12}{4} = 2.78 \text{ g/l}$$

Filter 6

- Before usage 0.0926 g
- After heating in 105°C 0.1424 g
- After heating in 550°C 0.1003 g

$$\text{Suspended solids: } \frac{0.1424 - 0.0926}{0.005} = 9.96 \frac{g}{l} \Rightarrow \frac{9.96}{4} = 2.49 \text{ g/l}$$

The nitrification rate can be calculated using the sludge concentration in Section 4.2.1 and Equation 4.12. However, since the nitrification only worked from ammonium to nitrite, which is the only nitrification rate that is possible to obtain. These calculations

are only carried out in Test 3 since both Test 1 and Test 2 showed no clear result of any nitrification.

$$r = \frac{\Delta N}{SS \times t} \quad (\text{Eq. 4.12})$$

$$r = \text{nitrification rate, } \frac{mg}{g \cdot h}$$

$$\Delta N = \text{increase or decrease for the nitrogen compound, } \frac{mg}{l}$$

$$SS = \text{suspended solids, } \frac{g}{l}$$

$$t = \text{time, } h$$

$$r(\text{filter 5, ammonium}) = \frac{\Delta N}{SS \times t} = \frac{60}{2.78 \times 5} = 4.32 \frac{mg}{g, h}$$

$$r(\text{filter 6, ammonium}) = \frac{\Delta N}{SS \times t} = \frac{60}{2.49 \times 5} = 4.82 \frac{mg}{g, h}$$

$$r(\text{filter 5, nitrite}) = \frac{\Delta N}{SS \times t} = \frac{50}{2.78 \times 5} = 3.60 \frac{mg}{g, h}$$

$$r(\text{filter 6, nitrite}) = \frac{\Delta N}{SS \times t} = \frac{50}{2.49 \times 5} = 4.02 \frac{mg}{g, h}$$

The ammonium transformation is quite similar to the nitrite formation as the nitrification rate for ammonium is 4.32 mg/g,h compared to the rate of nitrite of 3.60 mg/g,h for filter 5 and 4.82 compared to 4.02 mg/g,h for filter 6.

4.4.2 RESULTS TEST 3

The dilution process was performed with accuracy and the pipette had undergone maintenance, thus normalization should not be necessary. However, the results were normalized with sodium and chloride to compare with the other tests. These results did not correlate as good as in Test 1 and 2 for neither sodium nor chloride. The results without recalculating the values are therefore valid and will be used. Figure 4.9 show the trends for ammonium, nitrite and nitrite for Test 3.

The results from normalizing the values are presented in Appendix 3. This nitrification test shows a clear decrease in ammonium concentrations and a clear increase for nitrite. But similar to the other test, despite an increase in alkalinity the nitrification process stops at nitrite and will not transform to nitrate. It can be seen in the figure that the decrease of ammonium is similar to the increase in nitrite. This test show much clearer relations between the two compared to Test 1 and 2. There are however two distinctive samples for nitrite, the last samples at time 22.25 hours which show nitrite concentrations of 1.1 and 0.3 mg/l, thus a decrease, when the start of the test showed a strong increase.

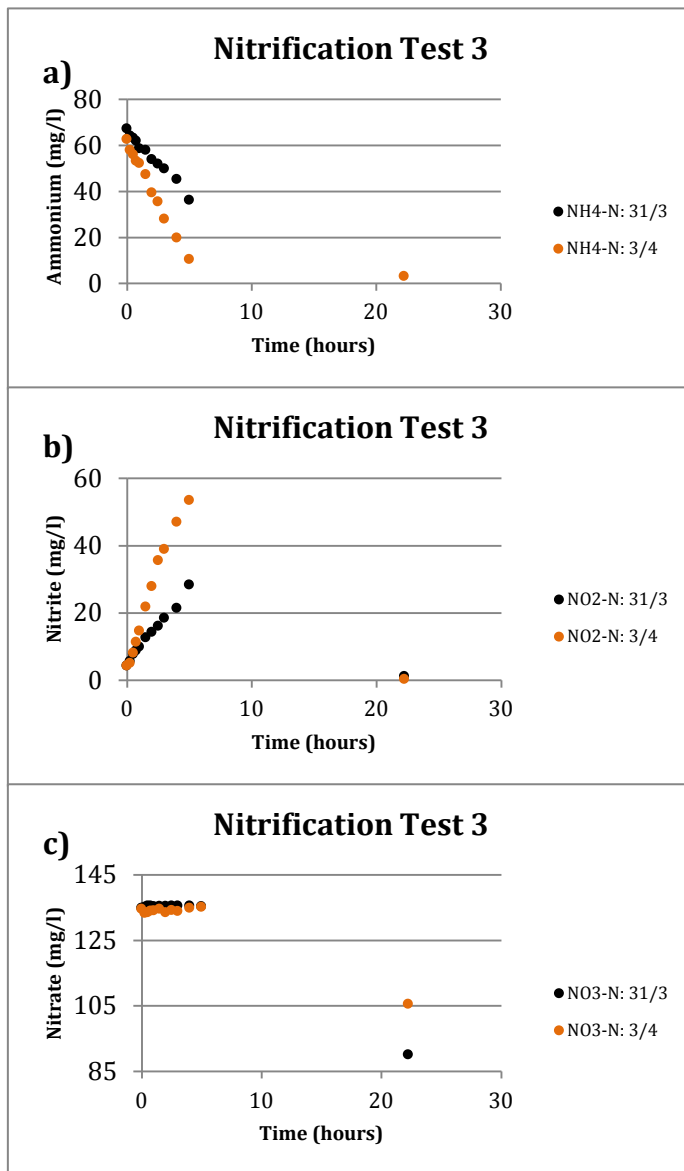


Figure 4.9. Results from nitrification test 3

The concentration of ammonium, nitrite and nitrate was run in the ion chromatograph to see if the levels had changed since Test 1. The results are presented in Table 4.4.

Table 4.4. Variation in nitrogen concentration in the process water

Compound	Test 1		Test 3	
	March 31	April 3	March 31	April 3
NH ₄ – N mg/l	104	111	139.7	129
NO ₂ – N mg/l	5.6	6	9.3	8.8
NO ₃ – N mg/l	346	386	-	-

Ammonium and nitrite have both increased since the process water was collected. The ion chromatograph showed no values for nitrate. The alkalinity was based on the nitrate concentrations measured in Test 1 and since this real concentration was

undetected during the analysis, the additive of sodium hydrogen carbonate could be incorrect in order to achieve the wanted alkalinity.

Test 3 was also analyzed for TOC and TN. The result for TOC is presented in Figure 4.10 for the two different types of process water. The figure shows an increasing curve of TOC concentration, though; there are some negative values for the process water from April 3. This could be a result of the concentration being very close to detection limit, thus limits that the machine is unable to discover. Therefore the machine could have resulted with some negative values in these measurements.

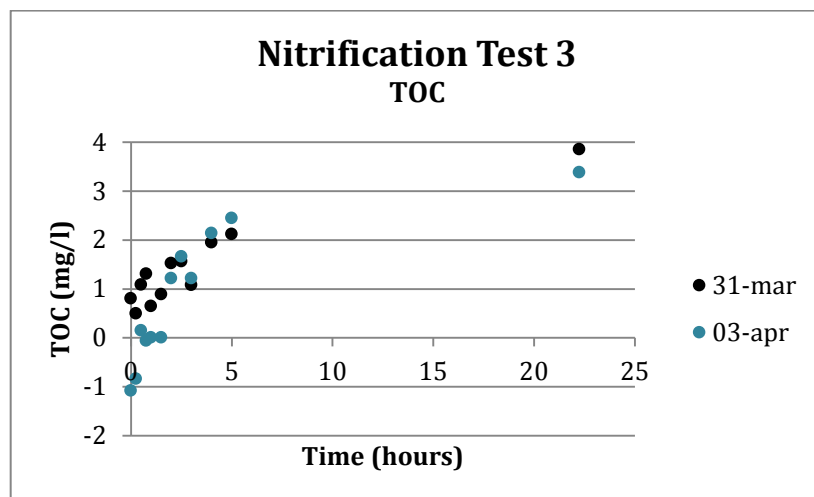


Figure 4.10. Results TOC nitrification test 3

Figure 4.11 shows the trend for TN for both process waters. The results for the two days are very similar and show that TN is staying at the same level, which indicates that some nitrification might be occurring since ammonium turned into nitrite.

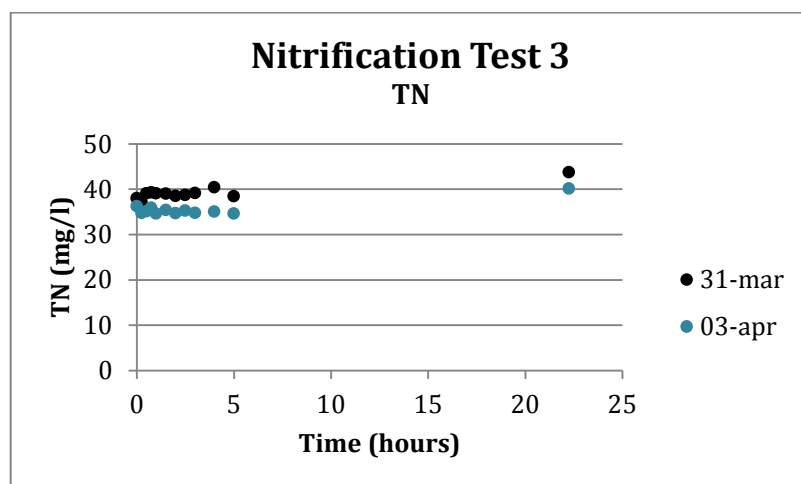


Figure 4.11. Results TN Nitrification Test 3

4.5 DENITRIFICATION TEST

A denitrification test was also carried out in the lab at Chalmers University on May 5 using sludge from May 2 collected at Hammargården wastewater treatment plant. The process water from April 3 was the only type used in this test due to the similarities in water compositions between March 31 and April 3. April 3 had slightly higher concentrations of nitrate why this sample was chosen. The sludge was first thickened by removing some of the supernatant. It was also aerated for 30 minutes before used

in the experiment since it had been stored cool for a few days. 1 100 ml of water and 900 ml sludge were first mixed by a magnetic stirrer. Nitrogen gas was then added for 30 min before the carbon source was added. The carbon source was acetate $CH_3CO_2^-$ and the required amount was calculated to 2.86 g/2 l using the stoichiometry for acetate and the concentration of nitrate which was determined to 386 mg/l in the nitrification test. The experiment was run for 3 hours (based on other standard denitrification tests) during regularly sampling, which is seen in Table 4.5. Sample 0_A is when acetate was added but due to measurement errors it was only 1 900 ml in total why another 100 ml of process water was added when sample 0_B was taken. The samples were centrifuged for 5 min as in the nitrification tests and then filtered directly through a 0.45 µm filter. The nitrogen samples were diluted 5 times using 2 ml filtered samples and 8 ml MQ water. These samples were then run in the ion chromatograph.

Table 4.5. Sampling scheme for denitrification test

Time	Process water	
	2014-04-03	
Min	pH	Sample
0	-	0 _A
8	-	0 _B
15	7.97	1
30	-	2
45	-	3
60	-	4
90	8.29	5
120	-	6
150	-	7
180	8.47	8

4.5.1 SLUDGE CONCENTRATION DENITRIFICATION

The sludge concentration in the denitrification test was also determined by using the same method as described in Test 1, Section 4.2.1. Filters 7 and 8 were heated for 2.5 hours in 105°C and 30 min in 550°C.

Filter 7

- Before usage 0.0954 g
- After heating in 105°C 0.1431 g
- After heating in 550°C 0.1017 g

$$\text{Suspended solids: } \frac{0.1431 - 0.0954}{0.005} = 9.54 \frac{g}{l} \Rightarrow \frac{9.54}{4} = 2.39 \text{ g/l}$$

Filter 8

- Before usage 0.0934 g

- After heating in 105°C 0.1406 g
- After heating in 550°C 0.0999 g

$$\text{Suspended solids: } \frac{0.1406 - 0.0934}{0.005} = 9.44 \frac{g}{l} \Rightarrow \frac{9.44}{4} = 2.36 \text{ g/l}$$

4.5.2 RESULTS DENITRIFICATION TEST

Figure 4.12 presents the results from the denitrification test. The denitrification process should give decrease in nitrate before transformed to nitrite which in turn should be transformed to nitrogen gas. The results show increasing nitrite and decreasing nitrate which indicates that the denitrification process was working to some extent. However, the test was only conducted during 3 hours at which the nitrate concentration was measured to around 110 mg/l, which is still a high concentration since the goal was to decrease nitrate close to zero mg/l. Thus the nitrate concentration was decreased around 20 mg/l while nitrite was increased with 40 mg/l. This shows that the denitrification was not fully complete since not all nitrate was transformed into nitrite.

The pipette was working when the samples were diluted which would make the results valid. But the results were normalized as in Test 3 with sodium and chloride.

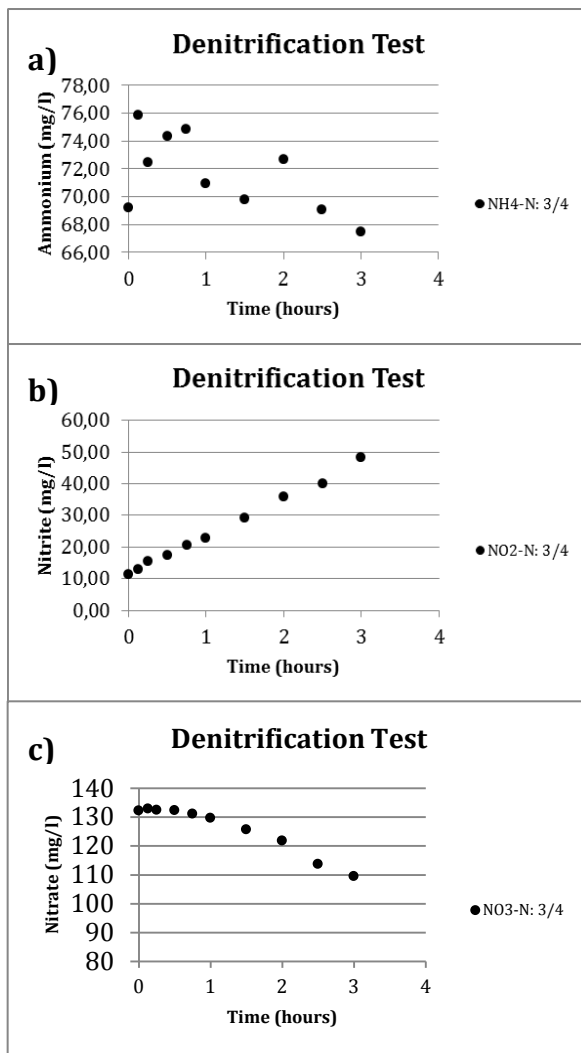


Figure 4.12. Results denitrification test

The results correlated better in this test than in Test 2, however the values fluctuated for all three nitrogen compounds why the final results for the denitrification process are analyzed using Figure 4.12. The normalized results are presented in Appendix 4 as well as Figure 4.12 in larger scale.

The result for the TOC test is presented in Figure 4.13 for the denitrification test. TOC is decreasing over time. TOC decreased from 85 mg/l to around 73 mg/l which is a decrease of 12 mg/l during 3 hours. The result for TN in the denitrification test is presented in Figure 4.14 which also shows a decline in concentration over time. This should be the result when nitrate is turned into nitrogen gas. It is therefore likely that some of the nitrogen during the denitrification test is transformed into nitrogen gas, thus evaporates from the solution. TN decreased from around 36 mg/l to 32-33 mg/l, which is a very small decrease and not similar to the decrease in TOC and in particular not equal to the nitrate decrease. This further proves that the denitrification was only partial.

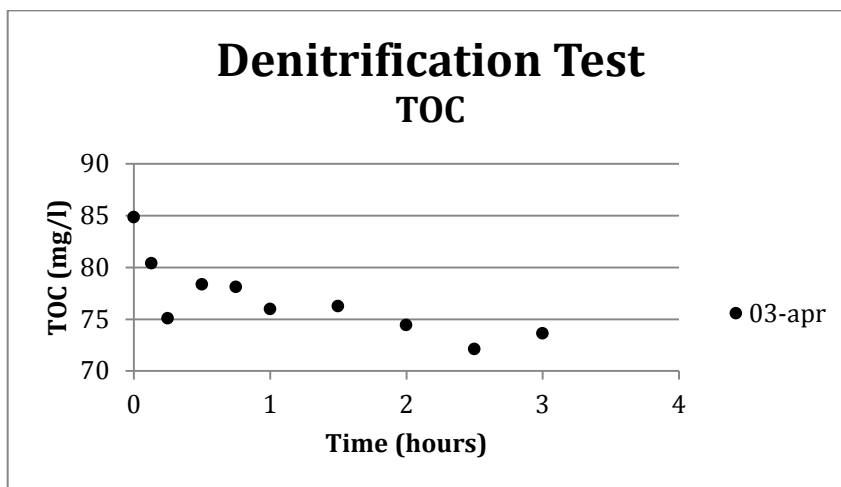


Figure 4.13. Results TOC Denitrification Test

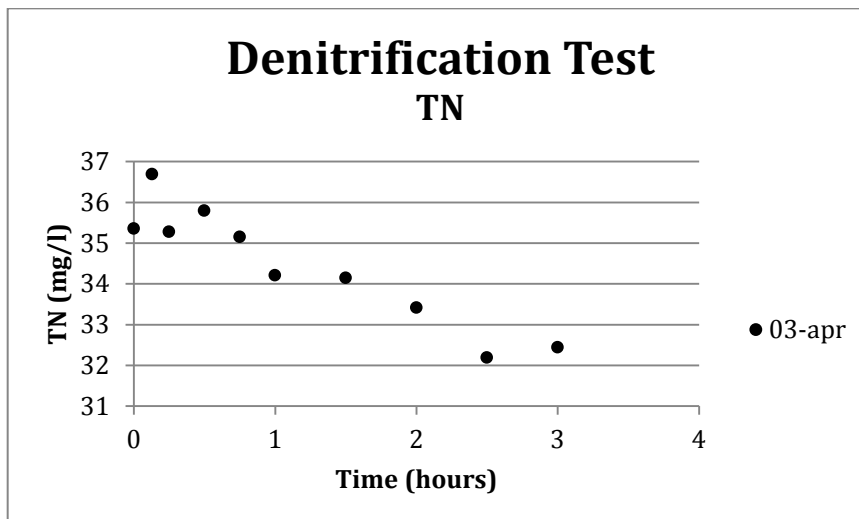


Figure 4.14. Results TN Denitrification Test

5 DISCUSSION

The main purpose of this master thesis was to conduct nitrification and denitrification tests in the lab with process water from the Gerum tunnel. The concentrations of ammonium, nitrite and nitrate were measured in the first test and the other tests were based on these concentrations. Though, when the process water was analyzed in the third nitrification test presented in Section 4.4.2, the nitrogen concentrations had increased. This could have affected the results in the test, in particular the calculations used to increase alkalinity in Test 3 since it was based on the nitrate concentrations from Test 1.

5.1 NITRIFICATION TESTS

The results showed a small decrease in ammonium (which should occur), a very small increase in nitrite and finally fluctuating values for nitrate. This shows that there was no complete nitrification, nor a very good result for the start of the nitrification process. It was suspected that the phosphate concentration was too low in the process water why it was decided to increase the concentration in the next nitrification test. It was also believed that the dilution had not been performed with great accuracy due to the pipette used. Thus, the results were normalized using sodium (cation) and chloride (anion) and the theoretical first value was calculated. The theoretical values were not consistent with the measured values why the conclusion was that the dilution had been compromised during the use of the pipette, where the volume in the test tubes varied from around 9.5-10.7 ml, in which there should have been exactly 10 ml. The correlation between sodium and ammonium, nitrite and nitrate were very good. The normalization with chloride also showed good correlation but sodium proved to be the best one why the diagrams and results are based on sodium. Since the results showed no clear sign of nitrification, a new test needed to be carried out. The concentration of TN varied during Test 1, which should have been at the same level throughout the entire test if the nitrification had been completed since ammonium is transformed into nitrate, thus the amount of TN should still be the same. But with the result presented in Section 4.2.2, it shows that the process was not working properly. The reason for both increases and decreases of TN during the test could be the lack of phosphate, low alkalinity and possibly lack of aeration during certain periods due to irregularly pressure in the flow. It could also have been simultaneous denitrification due to the irregular flow if the aeration was insufficient.

The second nitrification test used the same process water as in Test 1. Since the results in Test 1 did not show nitrification activity as hoped, it was decided to increase the phosphate concentration to see if it would help the process. These samples were diluted at the same time as Test 1 (the samples had been frozen) which gave Test 2 the same dilution errors as Test 1 since the same pipette was used. The results were therefore normalized with sodium and chloride to see if any of the two had better correlation. Sodium proved to have better correlation again and these values were used when analyzing the curves and ammonium, nitrite and nitrate were plotted against the time. Ammonium should decrease during nitrification, and since phosphate had increased the hope was to achieve better results. However, the trend for ammonium or nitrite was not very distinctive. This shows that nitrification was not working in this test either, despite increase of phosphate. The pH was measured in the beginning of the test to around 6.9 for both process water solutions, but the process water from March 31 increased during the test to 8.3 after 220 min but stopped at 8

after the entire test. The process water from April 3 also reached around 8.3 during the test but when the pH was measured after 17 hours, it had decreased to 6.6. The pH should be between 6.5 and 8.0 for nitrification to occur, which the pH was during the test, thus the pH is most likely not the parameter causing nitrification failure. The concentration of TN during Test 2 fluctuated, as in Test 1, which further proves that this test had an incomplete nitrification.

The third nitrification test used sludge collected the same day from the wastewater treatment plant. From the previous two tests, and since something in the project was decreasing the alkalinity in the recipients, it was suspected that the alkalinity was too low. The alkalinity was determined to 2.16 mM and 2.12 mM for the process waters which were below the required alkalinity for the high concentration of ammonium in the waters. The alkalinity was therefore increased by adding sodium hydrogen carbonate. The phosphate concentrations were increased to the same levels as in Test 2. The pH was monitored and it was around 7.8 in the beginning of the test for both process waters. The process water from March 31 increased to 8.3 but decreased to 7.8 at the end of the test. The process water from April 3 also increased in the middle of the test to 8.2 and then decreased to 8 at the end. The pH has been in the range of what is preferable for nitrification (6.5-8.0) which should not have impacted the nitrifying activity. If pH increases in the solution, thus becoming more basic, ammonium can transform into free ammonia. However, since pH has been kept at a stable level, this should not be the reason for nitrification failure. The pipette had undergone maintenance and gave good results without normalizing with sodium and chloride. Nonetheless, the final step in the nitrification process i.e. nitrite to nitrate, did not work in this test either. The nitrate concentrations were kept at a similar level, thus an incomplete nitrification. The ammonium levels were close to zero mg/l at the end of the nitrification test but if the test would have been kept running longer, it is a possibility that all ammonium would have transformed into nitrite which maybe would have started to transform into nitrate. The concentrations of TN during Test 3 remained at the same levels which indicate that some nitrification is occurring since the amount of nitrogen compounds is similar during the entire test with the exception of a small increase at the end of the test. Thus, nitrification was occurring at the beginning of the test but was inhibited between the nitrite-nitrate transformations.

5.2 DENITRIFICATION TEST

There were only one denitrification test performed in this study. Denitrification requires a carbon source and acetate was used for which the amount was determined based on the nitrate concentrations from Test 1. The phosphate and the alkalinity concentrations were not increased in this test in order to observe the performance of only process water, sludge and acetate. The aim with denitrification is to change nitrate into nitrite which turns into nitrogen gas. The dilution for this test was also done accurately with the pipette and should not need normalization. The results show that nitrate is decreasing while nitrite is increasing. Ammonium is also decreasing but with some fluctuating values. The results are quite good for the denitrification, even though not all ammonium is removed. The test was only run for 3 hours, which decreased nitrate from 130 mg/l to 110 mg/l (Δ 20 mg/l) while nitrite increased from 10 mg/l to 50 mg/l (Δ 40 mg/l). If the test would have been run for longer time, e.g. around 20 hours as in the nitrification tests, the nitrate could have been further decreased and nitrite further increased. Denitrification aims to decrease nitrogen levels and the concentration of TN shows a decrease during the test. This proves that some of the nitrate and nitrite are transformed into nitrogen gas which has left the

solution, thus denitrification is partially in progress. However, when assessing the results it is seen in Figure 4.12 that all nitrate is not transformed into nitrite and all nitrite is not transformed into nitrogen gas. This test had other sludge-process water proportions compared to the nitrification tests, i.e. more sludge was used in relation to the volume of water. This could have contributed to better results and it would have been interesting to run a test with 50/50 proportions for all three nitrification tests as well to see if the results would be different. If phosphate and alkalinity concentrations would have been increased in this test it might have resulted in an even better outcome since it already showed some denitrifying activity.

5.3 SLUDGE

The sludge used was collected from a treatment plant in Kungsbacka which had both nitrifying and denitrifying bacteria. Due to logistics, the sludge was only used fresh i.e. the day it was collected from the plant, for Test 1 and Test 3. For Test 2 and the denitrification test, the sludge had been kept cooled in containers for maximum 3 days. This could affect the processes even though the sludge was aerated before usage. It would have been preferable and probably better for the tests to have new daily sludge for all tests, but due to logistic difficulties of collecting the sludge, the same sludge was used in more than one test. However, another aspect of failed nitrogen removal could be related to the sludge in terms of acclimation to the environments for the bacteria. The bacteria in the sludge are not used to process water from a tunnel that was used in this study, i.e. those bacteria are used to regular wastewater. Bacteria sometimes need time to acclimate to new environments and time for acclimatization was not provided for the bacteria prior the tests. This could be a factor that should be assessed further, whether or not the bacteria in the sludge need to be familiar with the process water before running a nitrification and denitrification test. For further analysis, it would be interesting to run nitrification tests using the same process water but different sludge from different wastewater treatment plants to see if the processes differ or if the problem with nitrification only relates to the composition of the water. The sludge age could also be a factor inhibiting nitrification. The exact sludge age was never known, but an approximated guess at the wastewater treatment plant was 15-20 days. It would have been interesting to use sludge with higher age since sludge older than 20 days has proved to help the nitrification process in cold temperatures.

5.4 SUMMARY

No tests in this analysis showed any distinct results on nitrification. They were all incomplete and the nitrification process only went from ammonium to nitrite, but it failed to change into nitrate which was the common factor for all three nitrification tests. Thus, the second step in the nitrification test is causing the failure and the reason for this is unclear. The denitrification process was quite successful since nitrate decreased, nitrite increased and TN decreased. However, since the total time was not sufficient enough to remove larger quantities of nitrogen, the test should have been performed much longer. The reason for why nitrification failed is still undetermined. Some thoughts and assumptions have been developed along the analysis. First, the concentrations of nitrogen in the process water are much higher than in other activities, such as in mining industries or in regular wastewater, which increases the pressure on the biological treatment. Some metals have inhibiting effects on nitrification such as zinc, copper and nickel, and some of these metals had increased in concentration in both Pulsebäcken and Gerumsälven during the tunnel project. The

metal increase comes most probably from the process water in the tunnel. The impacts from metals have not been taken into account during the lab. This could therefore be a reason, in combination with other factors such as phosphate and alkalinity, of the nitrification failure. It is a possibility that the phosphate concentrations should have been increased even further in Test 2 and Test 3 in order to achieve a complete nitrification since the same concentration was used for the two. Same connection applies for alkalinity which could have been increased even more in Test 3. Other inhibiting parameters are pH and nitrite itself. However, pH has been quite stable during the tests and has almost at all times kept within the range of 6.5-8.0 and should not be the primary affecting parameter. The concentration of nitrite could also inhibit the process if it is too high but nitrite was below the critical levels. Another problem could also be a shortage of micronutrients that are necessary for microbial growth.

The problem for projects such as the Gerum tunnel is the cold water in the biological treatment which inhibits the nitrification process in terms of decreasing the nitrification rate. However, all tests in the lab were performed in room temperature and the nitrification process was still not completed. This indicates that the low temperature is not the major issue, even though it could be a problem as well. It was assumed prior to this study that nitrification would be possible in at least room temperature and that the problem would be to manage the cold water. The analysis in the lab does however present other complex results which brings more questions than just those about low temperatures. There is no clear answer to why the nitrification tests were uncompleted, but it is not only cold water that contributes to the failure, the proportions of sludge and process water and the concentrations of metals could also have been a strong factor to the results.

6 CONCLUSION

The conclusions of this master thesis are stated in this section.

- The first nitrification test resulted in an incomplete nitrification which was assumed to be correlated with low phosphate concentrations in the process water.
- The second nitrification test also showed incomplete nitrification despite a phosphate concentration increase. The phosphate increase could however have been too low in order to achieve a complete nitrification.
- The third nitrification test showed the best results from the nitrification tests assumed to be correlated with an alkalinity increase in combination with an increase of the phosphate concentrations. Hence, the ammonium-nitrite transformation was working for all three tests but the second step of transforming nitrite to nitrate failed. This is most likely due to the phosphate and alkalinity concentrations, but also to the metal concentrations of the process water which was not assessed.
- The denitrification test showed partial denitrification which could be related to a proportion alteration between process water and sludge compared to the nitrification tests and that denitrification is less sensitive.

The issue prior this study was the low water temperatures in tunnels and in the biological treatment. All tests in the lab were performed in room temperature and the nitrification process still failed. Thus, the low water temperature at site is not the major concern. Other inhibiting parameters on nitrification are metals where the assessment on the recipients showed increases in several metals that could affect the nitrification process. The pH could also affect nitrification, though it should be between 6.5-8.0 which it was for most part of the tests. The proportions in the solutions of sludge and process water was 500/1 500 ml in the nitrification tests and 900/1 100 ml in denitrification test, which also had the best results. This could be a factor that affects the nitrification process in combination with increased metal concentrations, low alkalinity and low phosphate concentrations.

Further studies are required to provide substantial answers and a solution to why the process water from tunnels is difficult to treat using biological treatment of nitrification and denitrification. Water is complex and cannot always be analyzed in a universal perspective, the local conditions differ and a tunnel in other parts of the world could have other water compositions and surrounding environments. Hence, further lab analysis on process water with high nitrogen content is crucial which should focus on phosphate and metal concentration, sludge age and the proportions of sludge and process water since those parameters were not thoroughly analyzed in this study.

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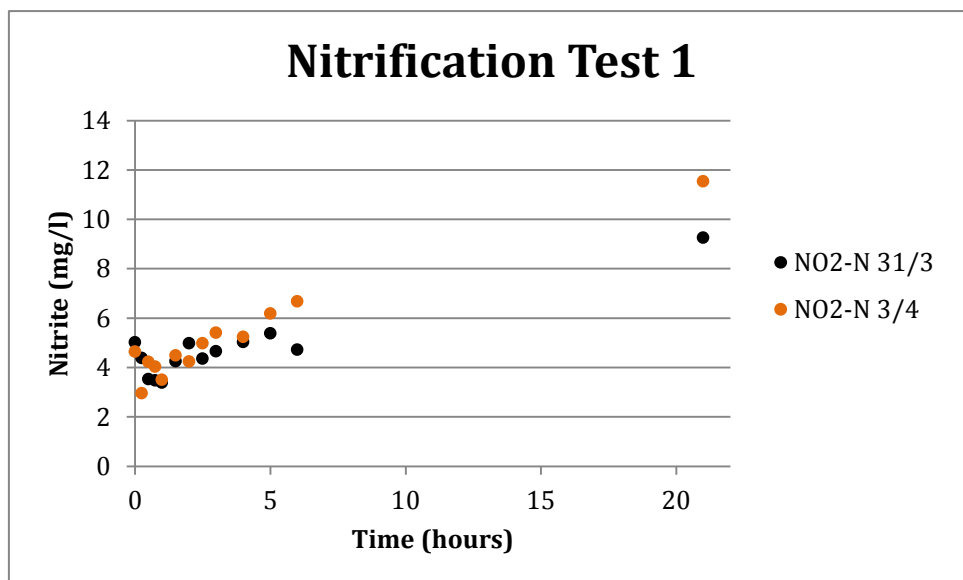
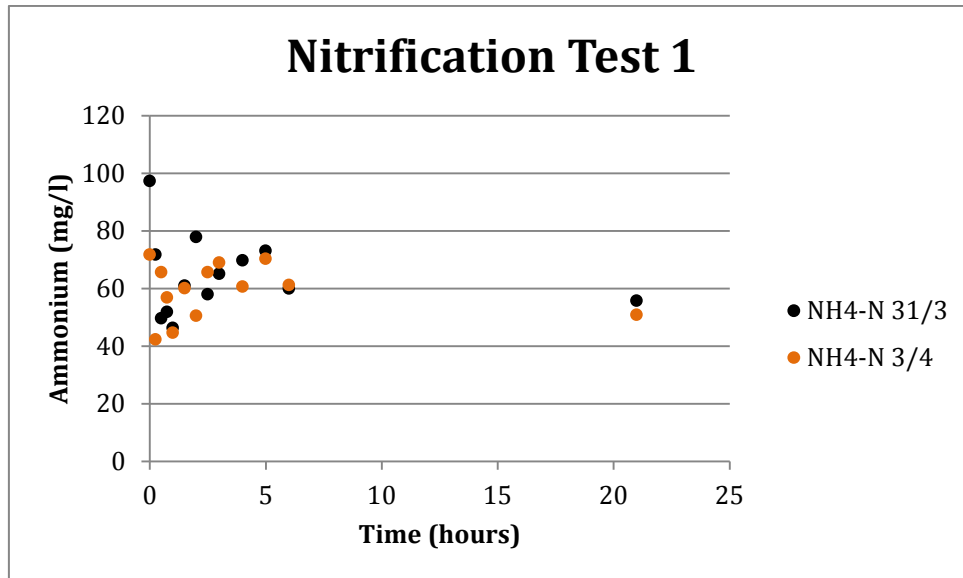
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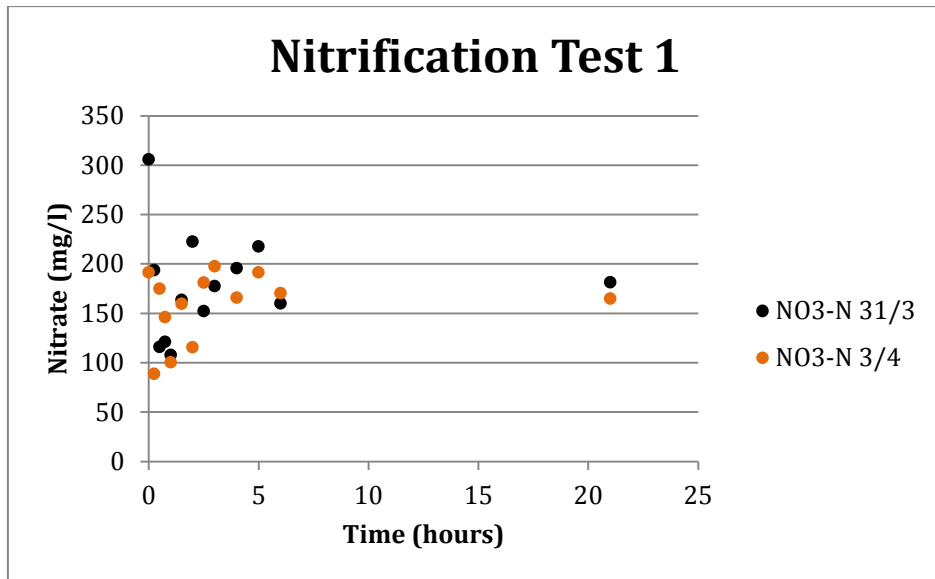
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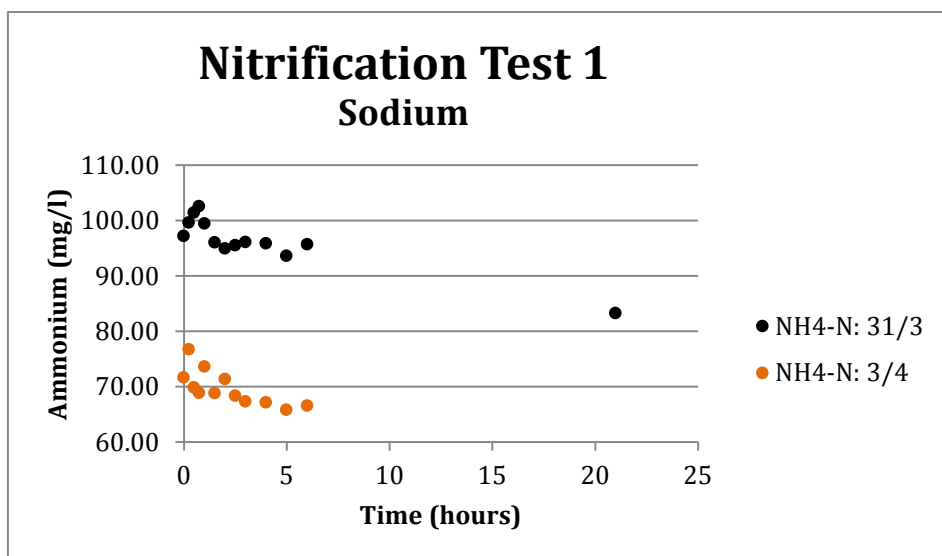
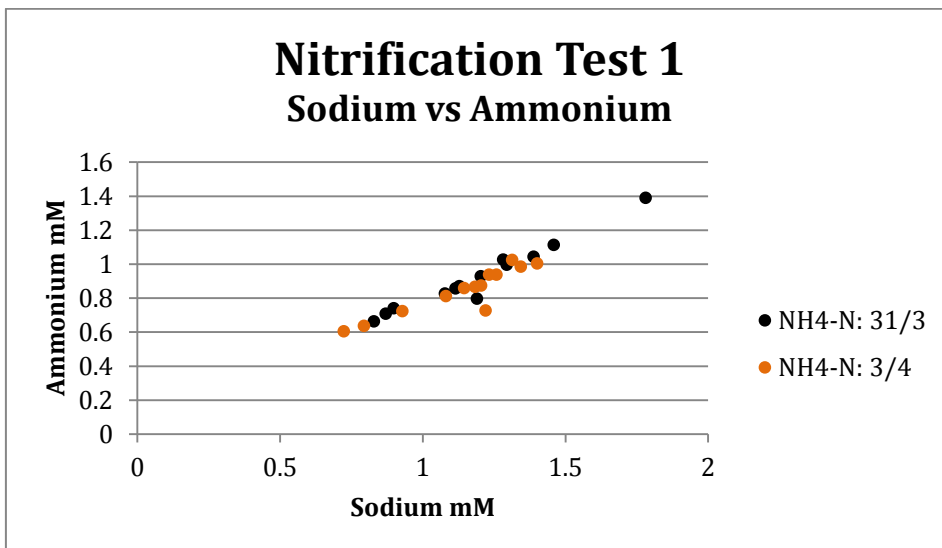
APPENDIX 1 – NITRIFICATION TEST 1

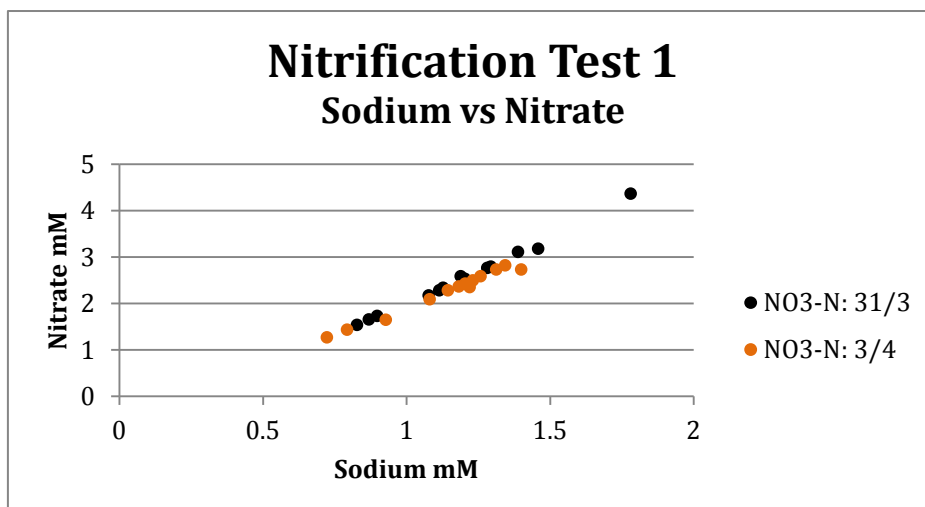
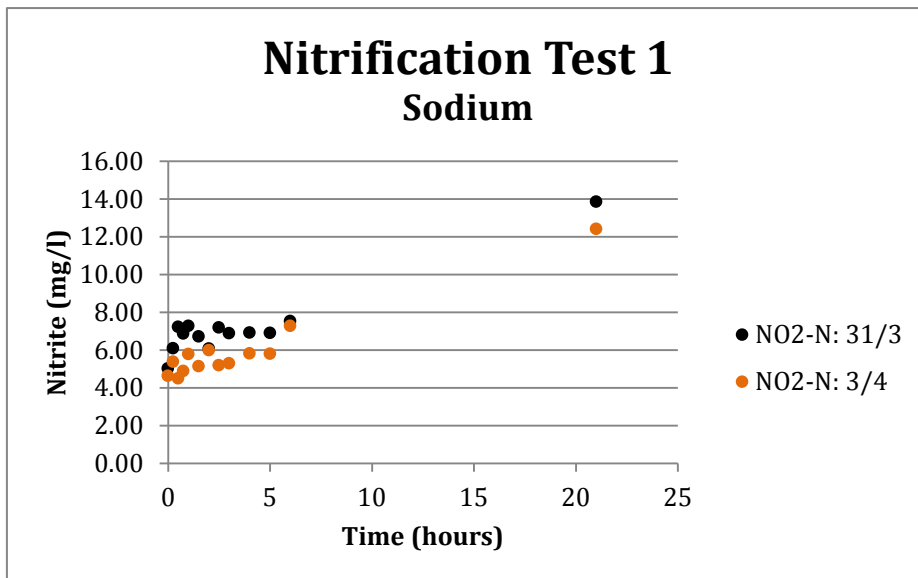
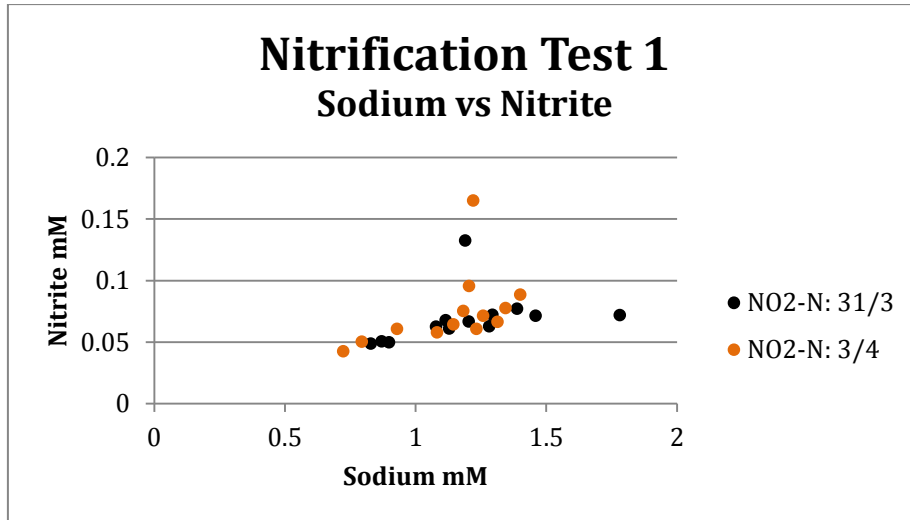
TRENDS BEFORE NORMALIZATION

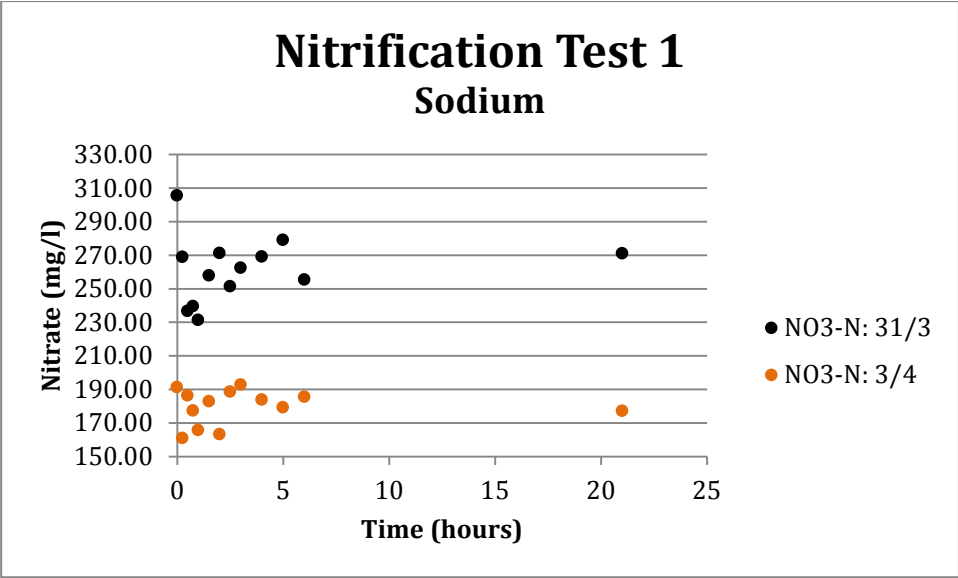




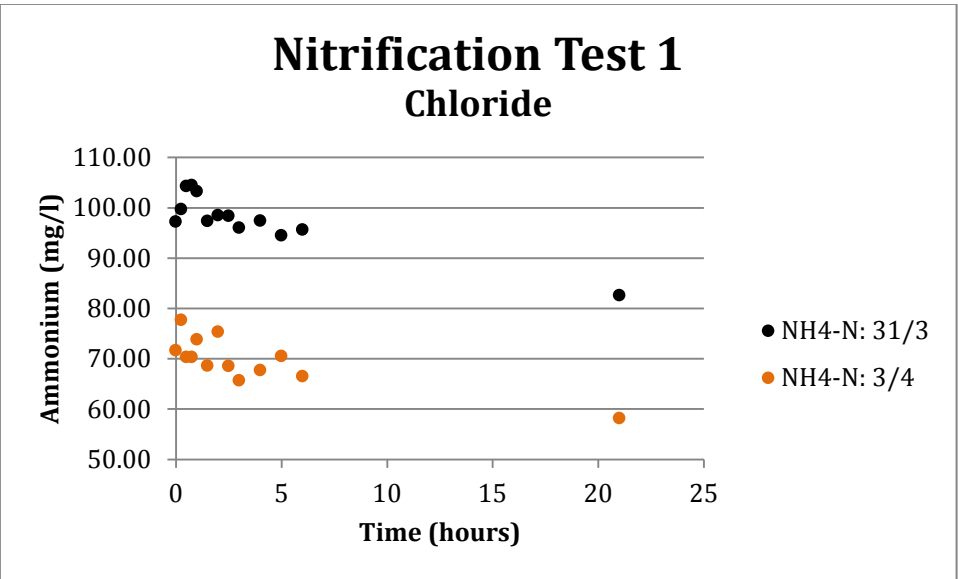
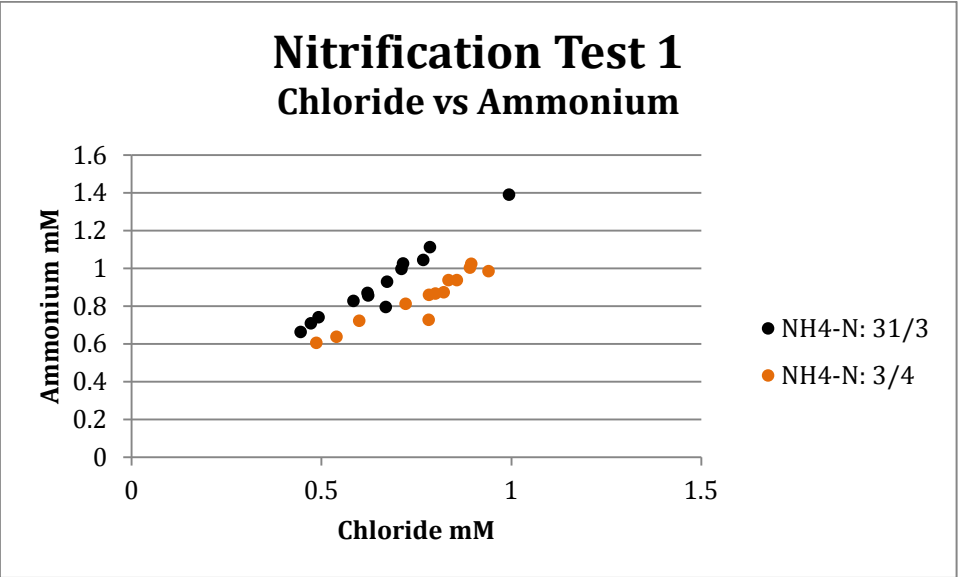
NORMALIZATION WITH SODIUM

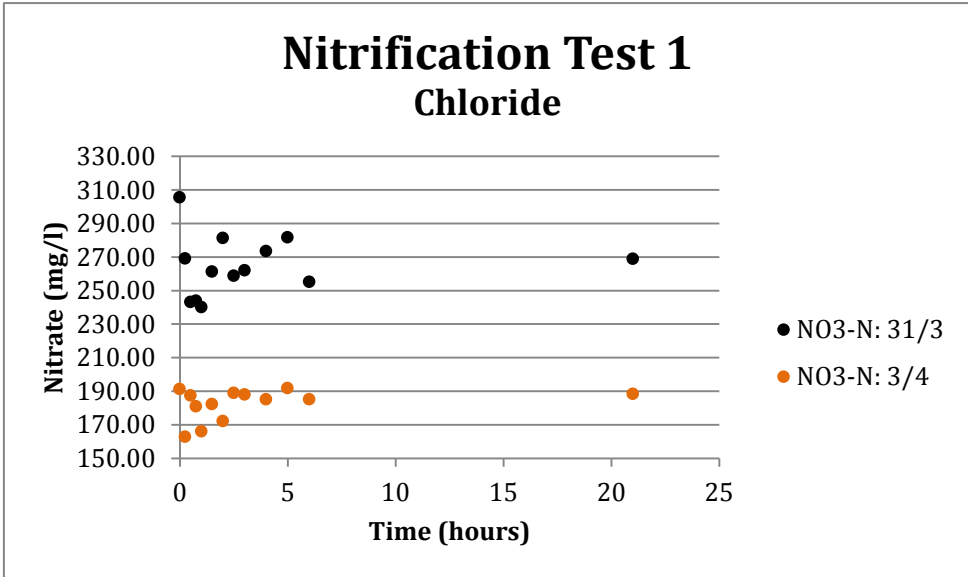
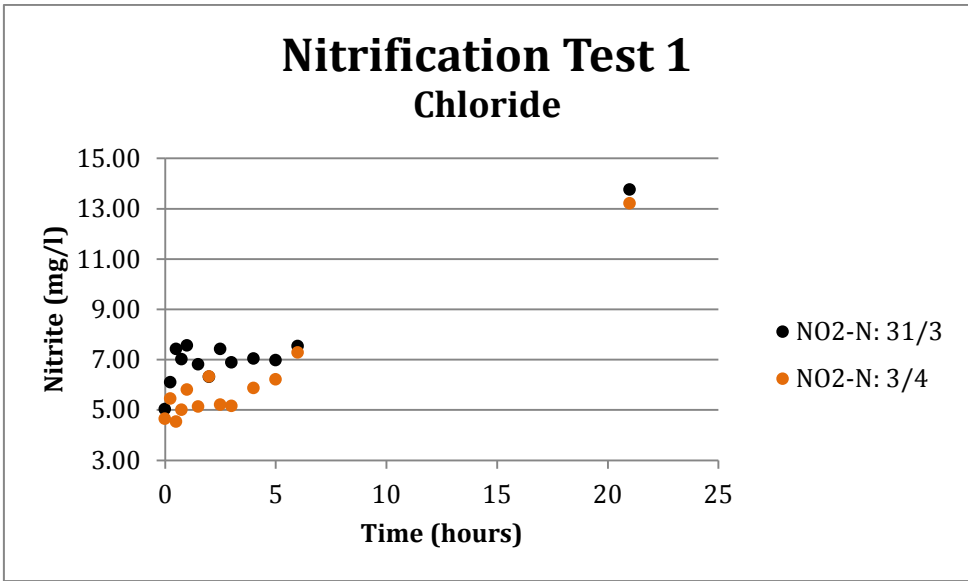






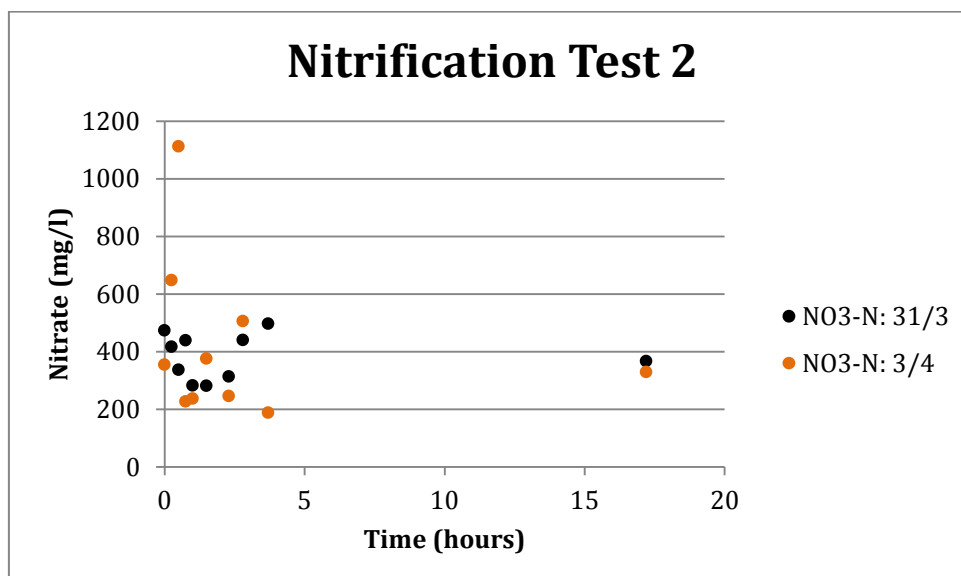
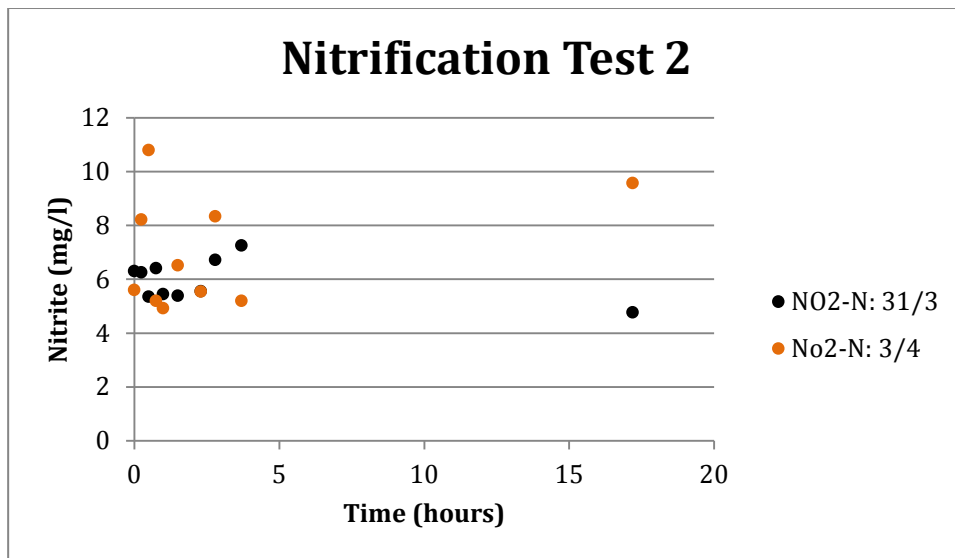
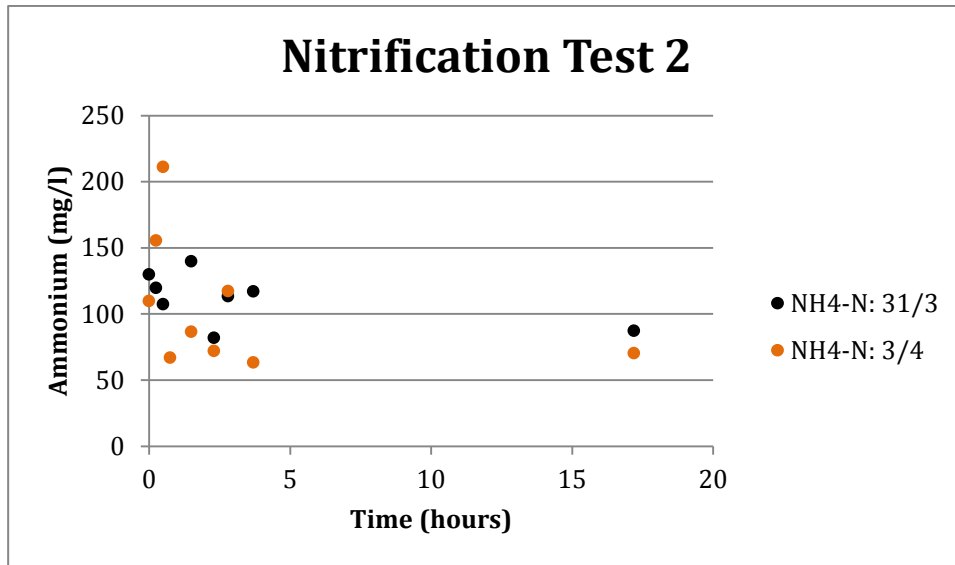
NORMALIZATION WITH CHLORIDE



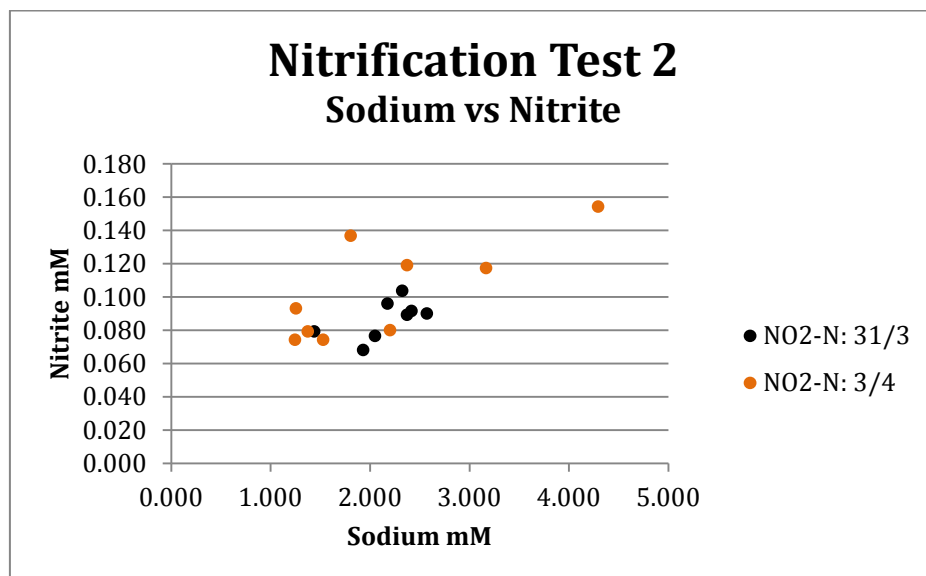
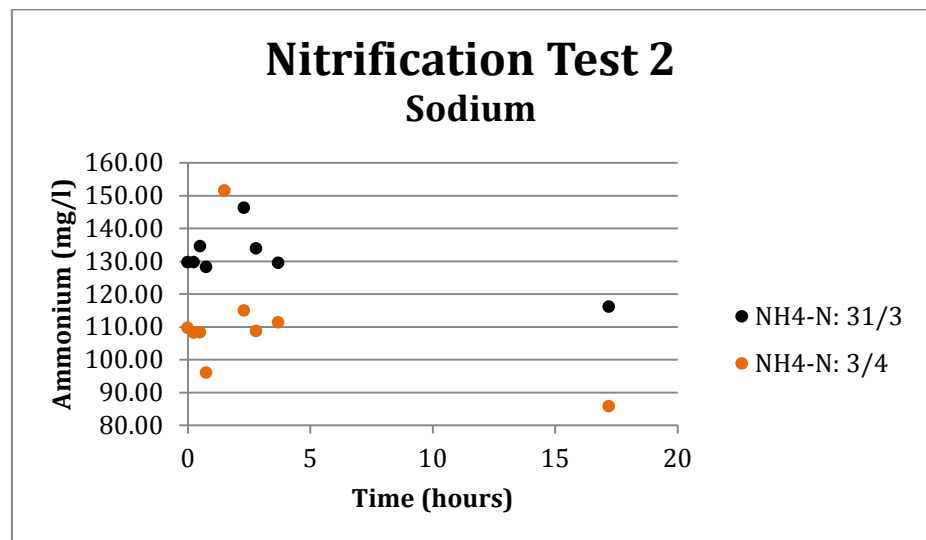
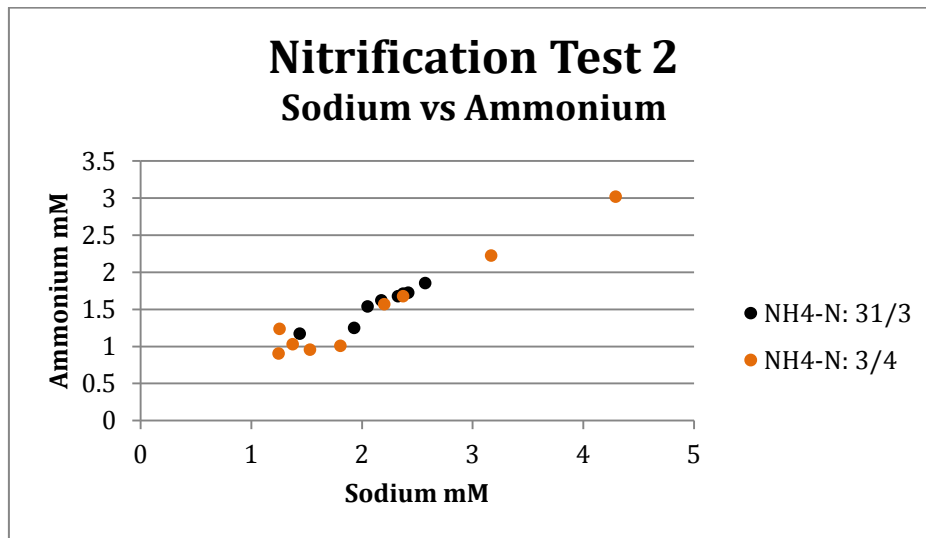


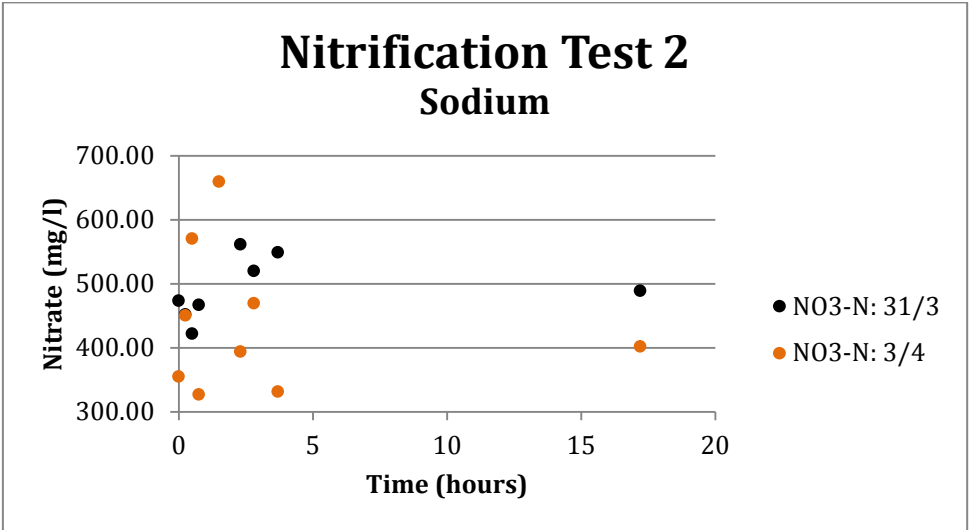
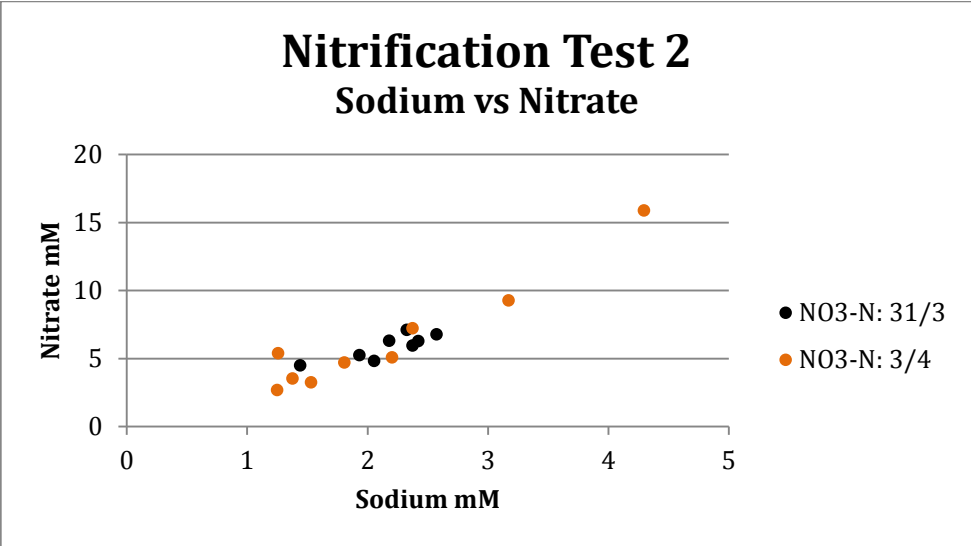
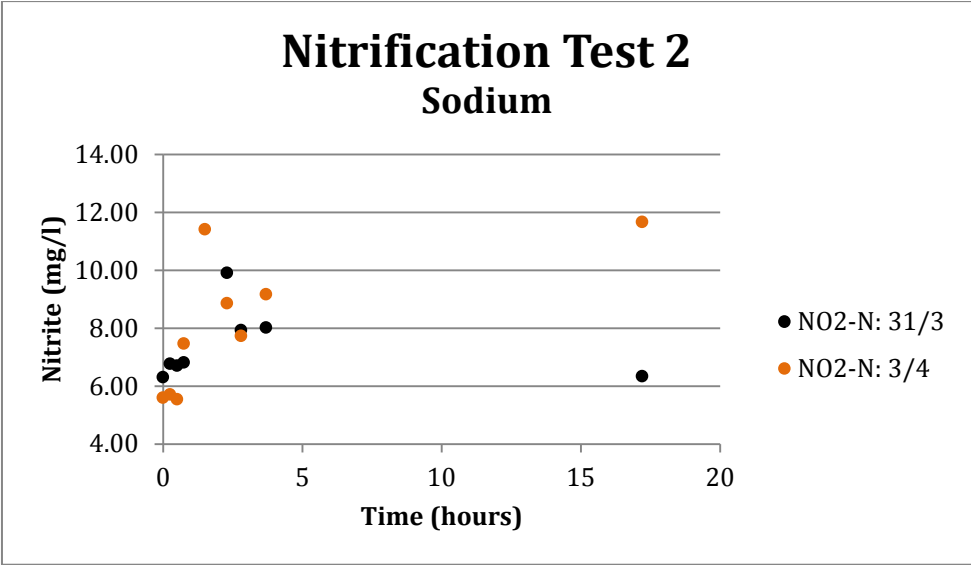
APPENDIX 2 – NITRIFICATION TEST 2

TRENDS BEFORE NORMALIZATION

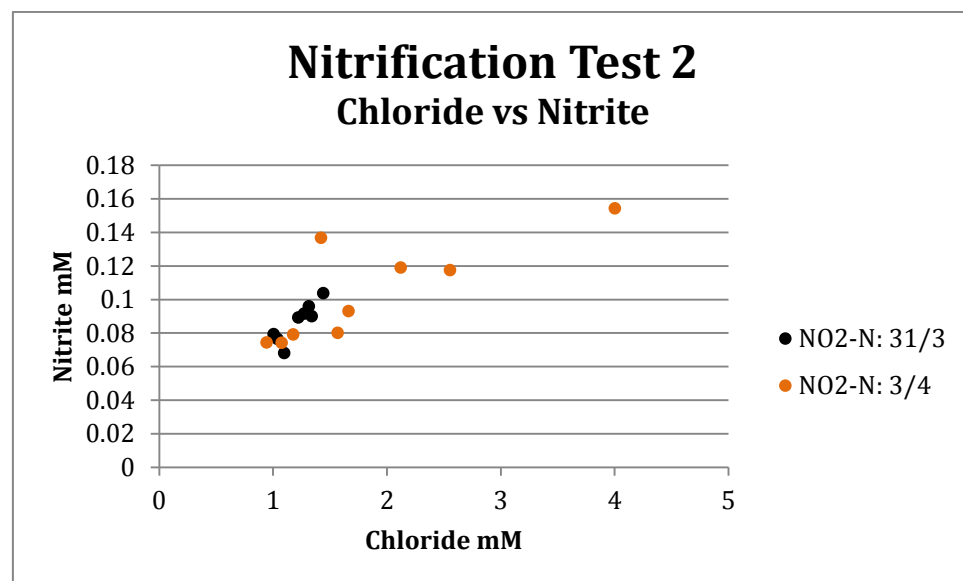
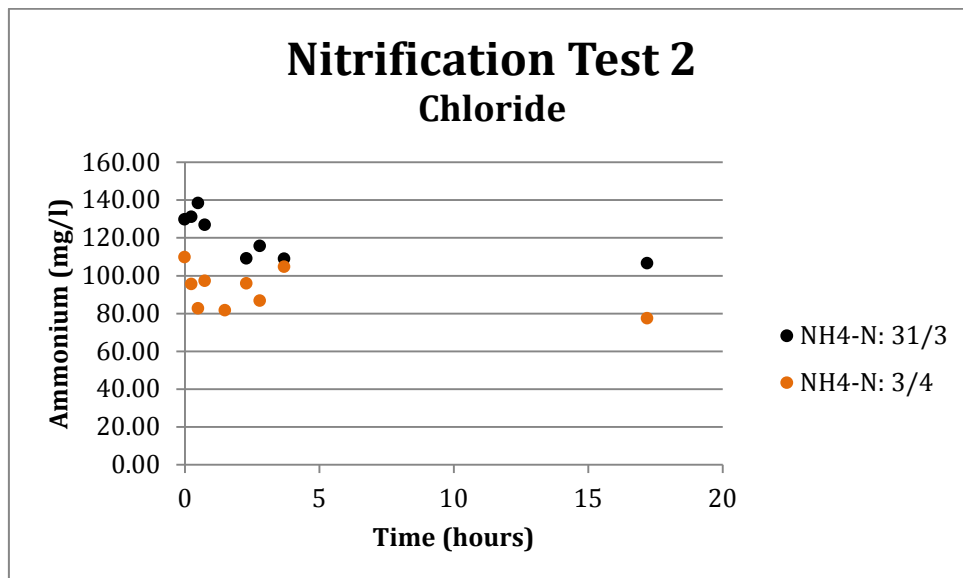
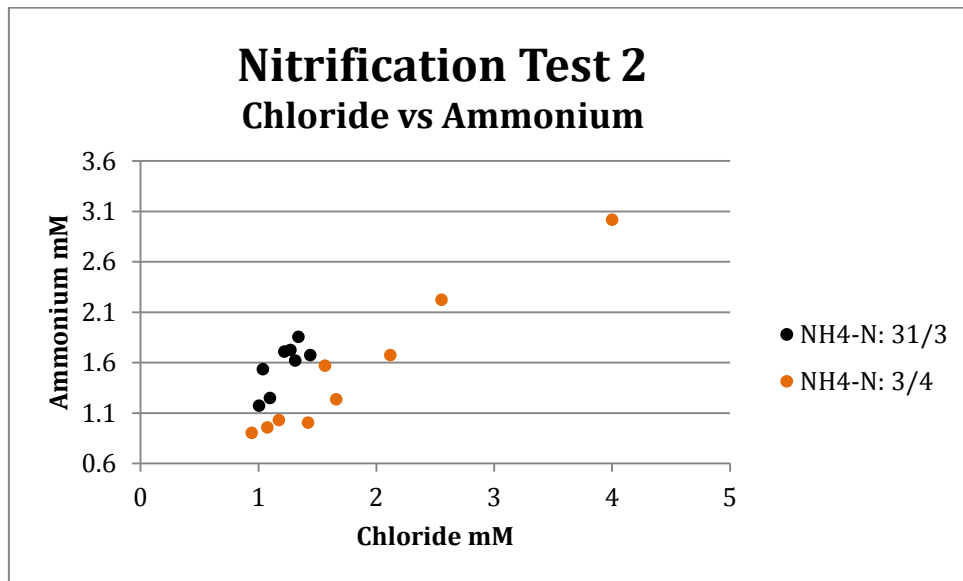


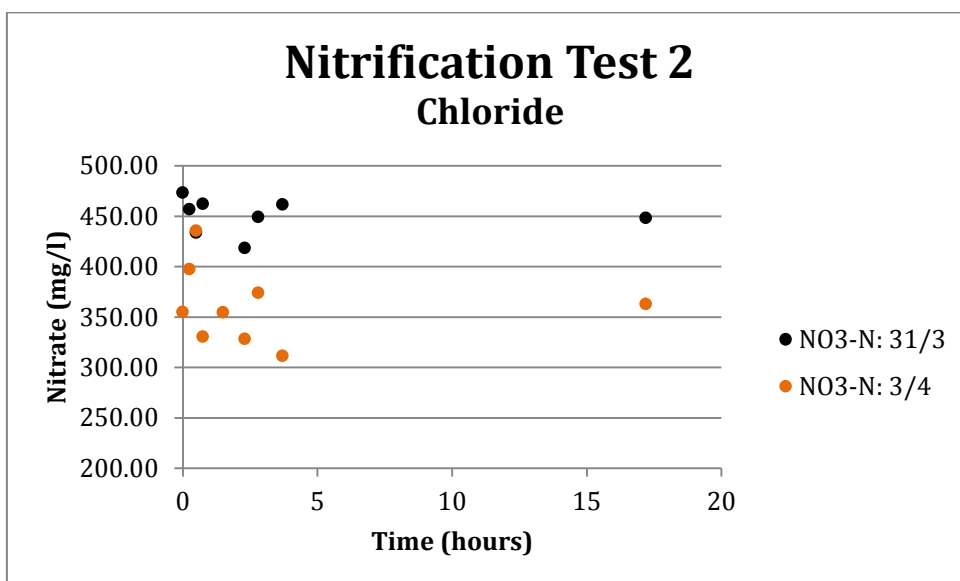
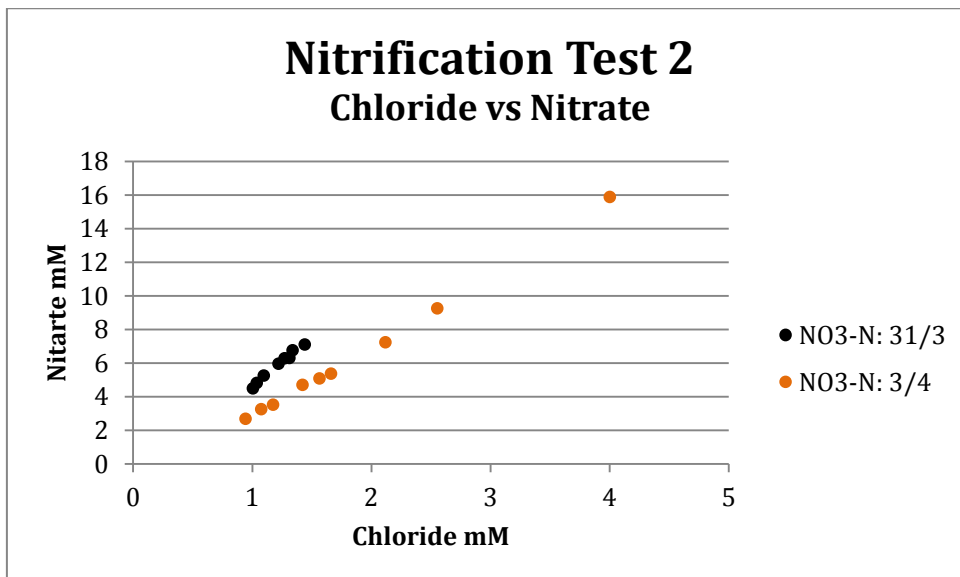
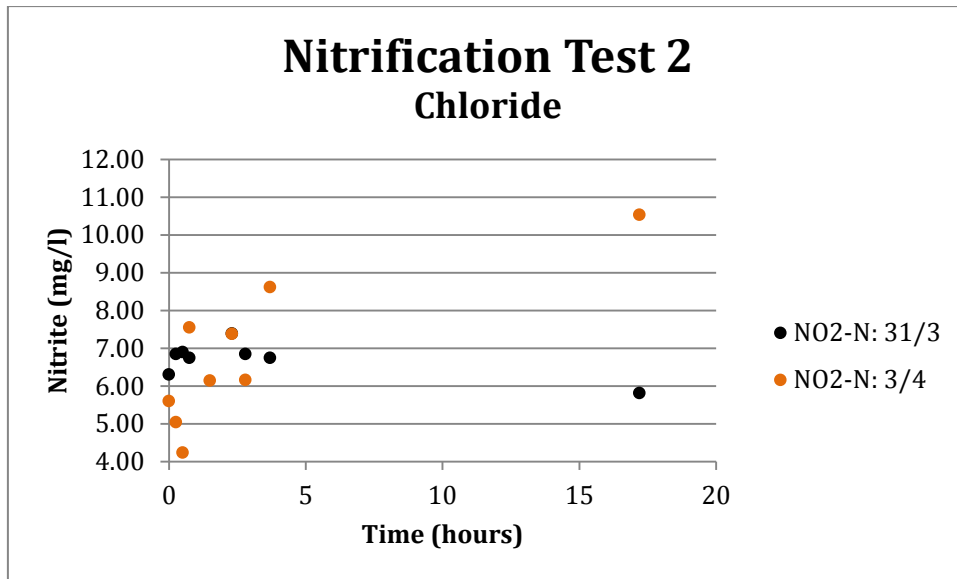
NORMALIZATION WITH SODIUM





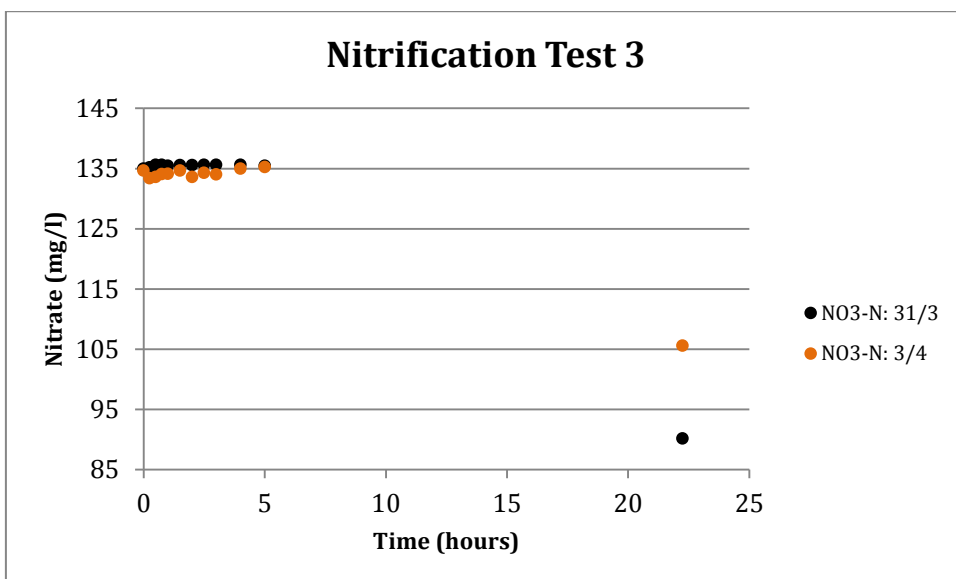
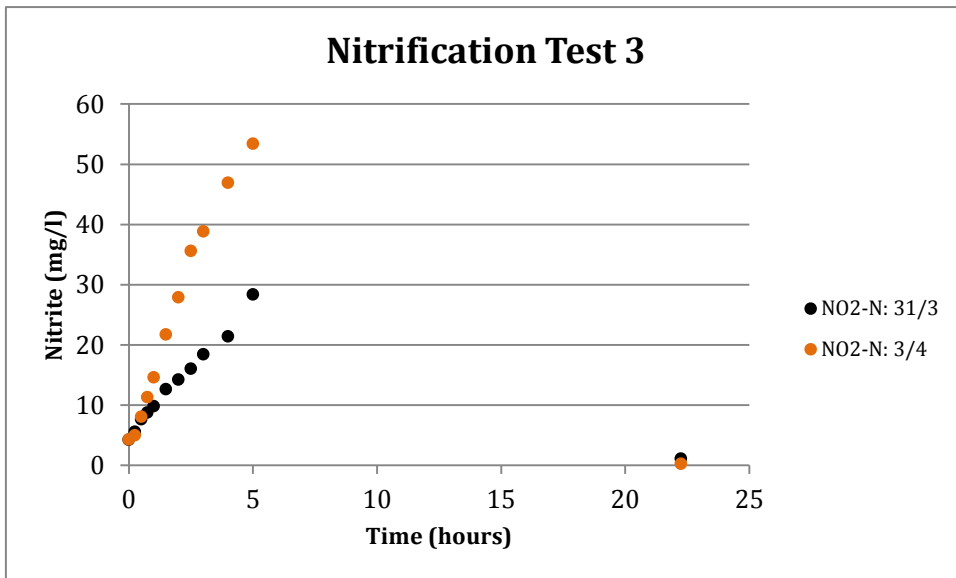
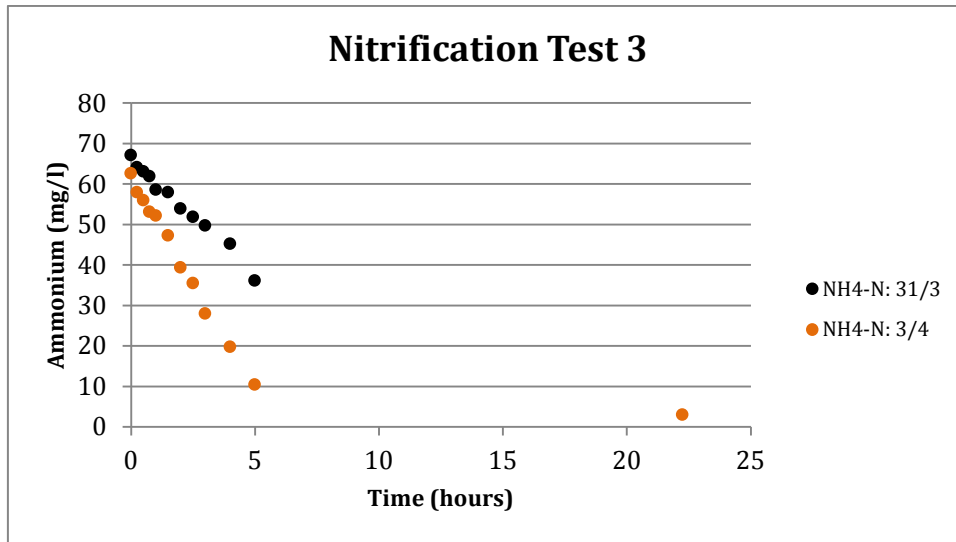
NORMALIZATION WITH CHLORIDE



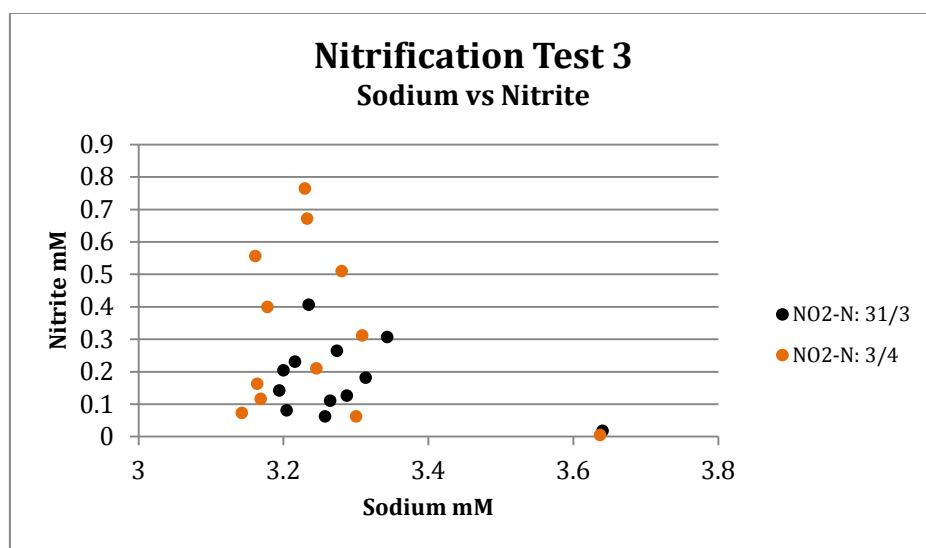
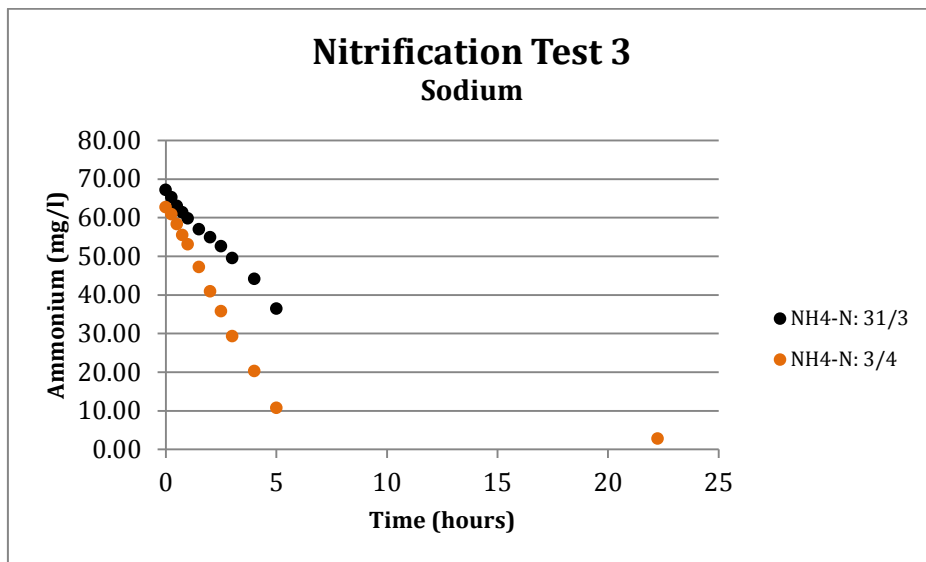
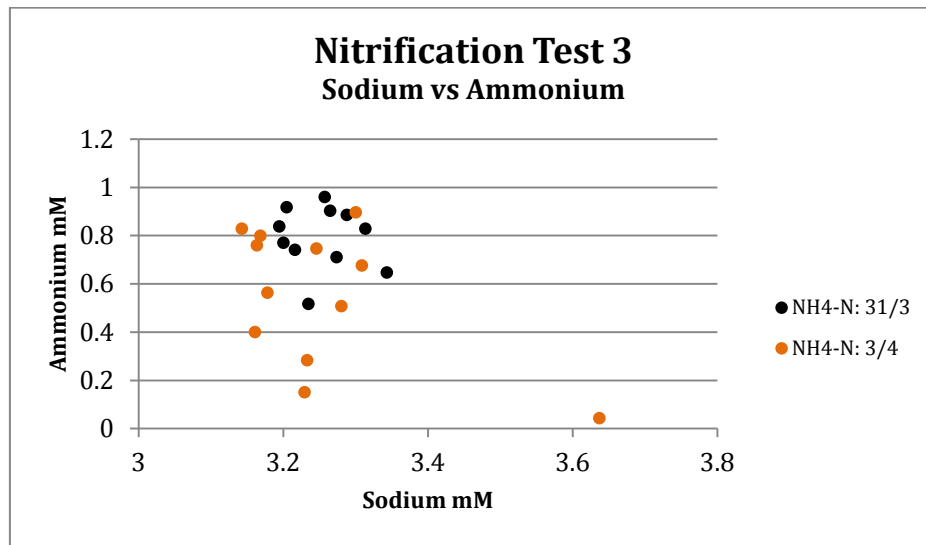


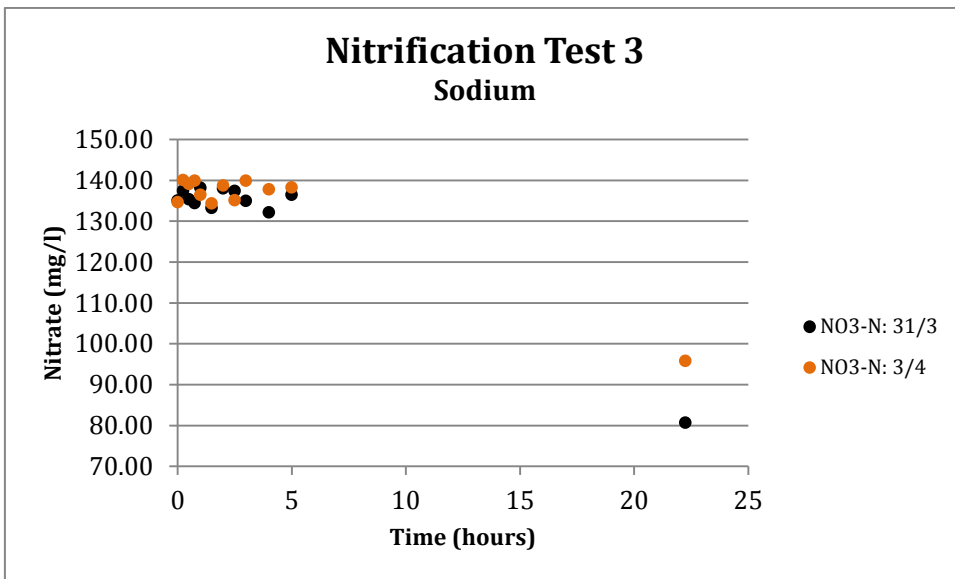
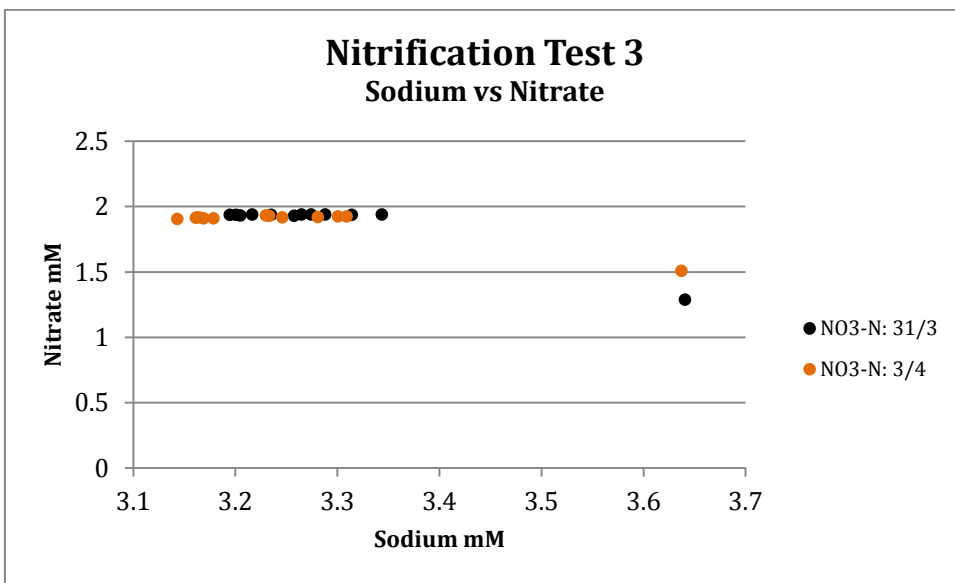
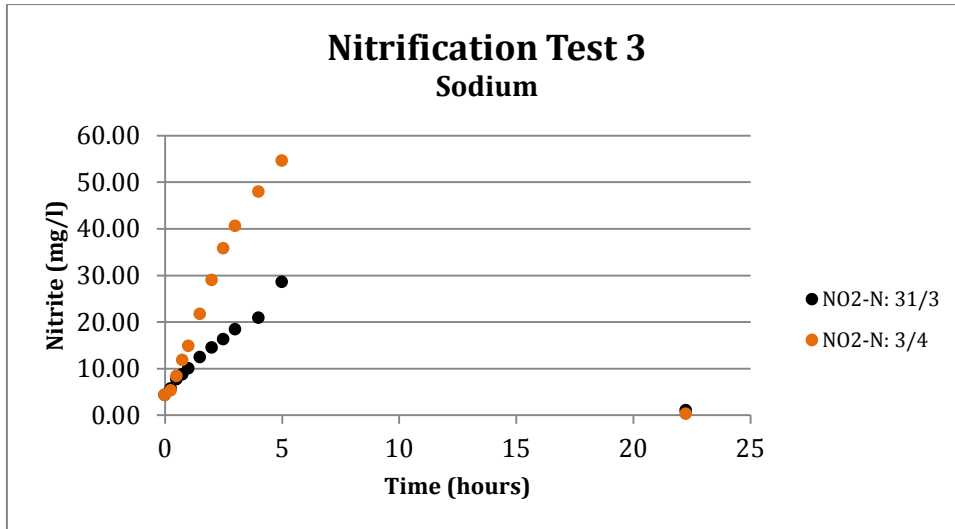
APPENDIX 3 – NITRIFICATION TEST 3

TRENDS BEFORE NORMALIZATION

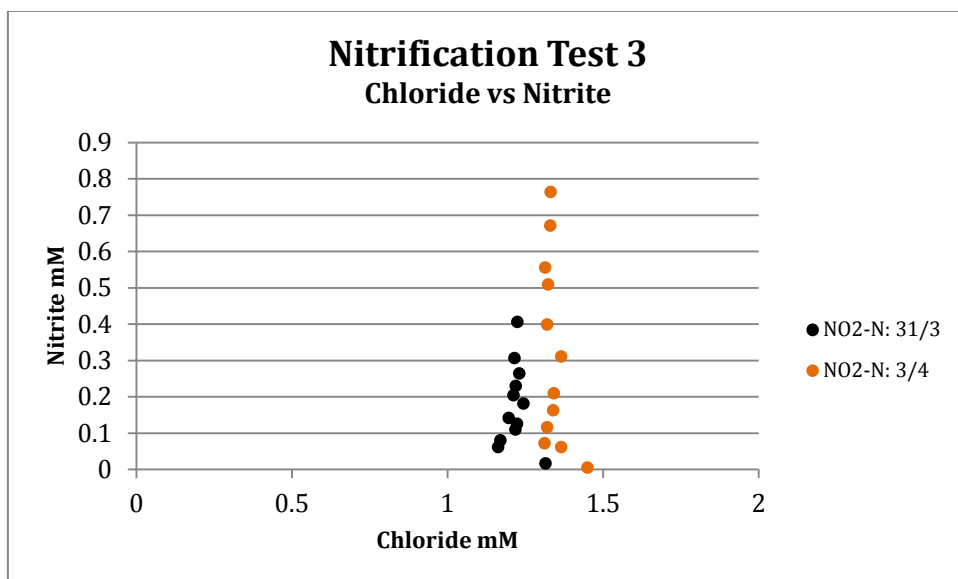
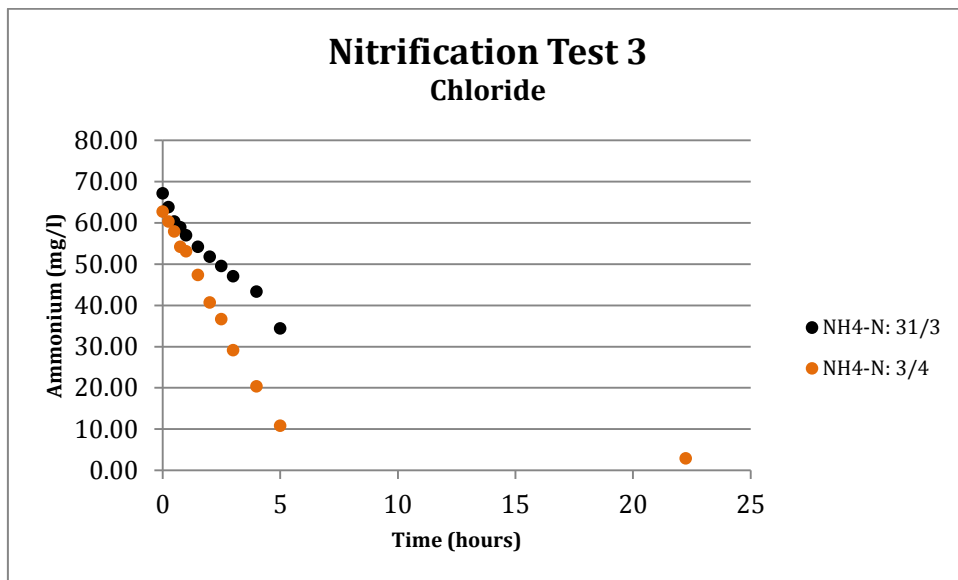
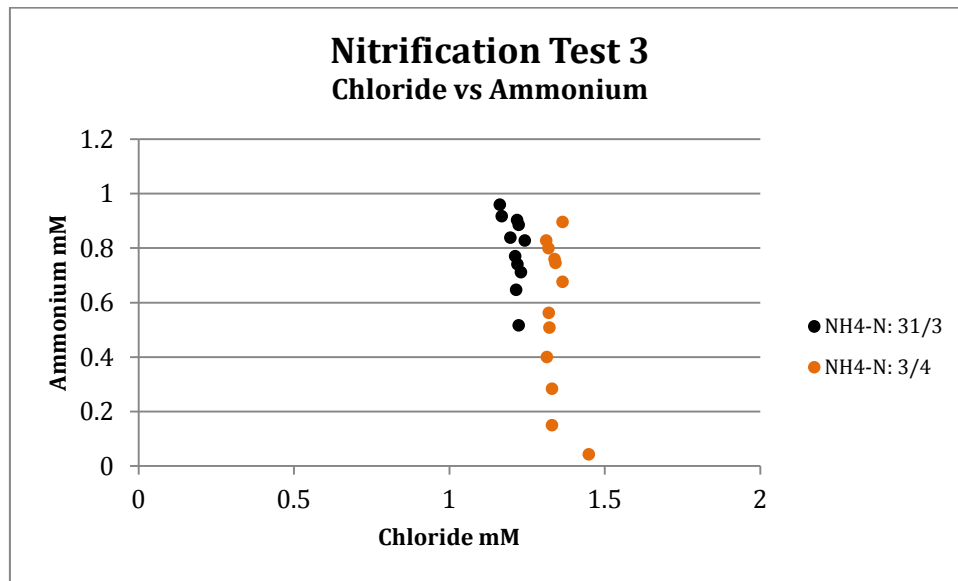


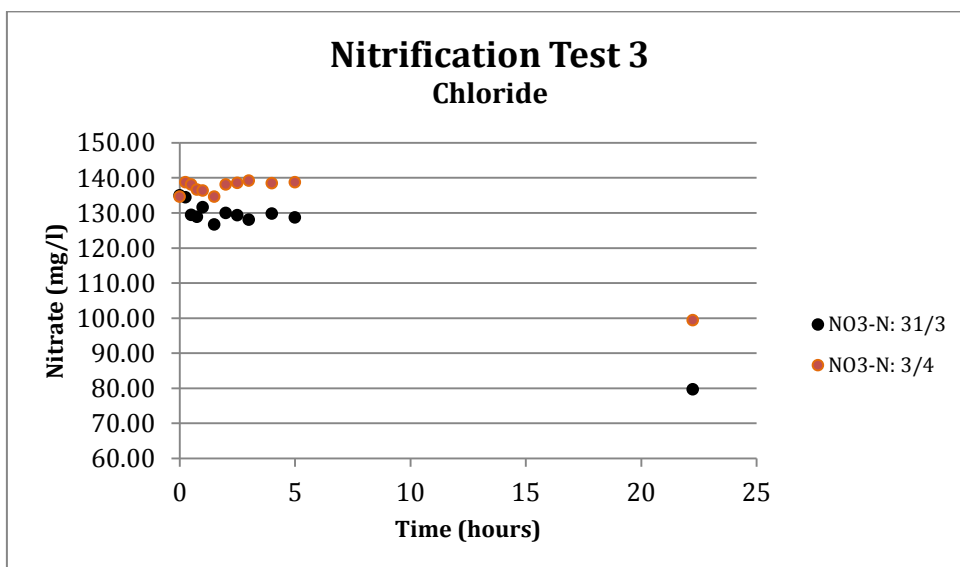
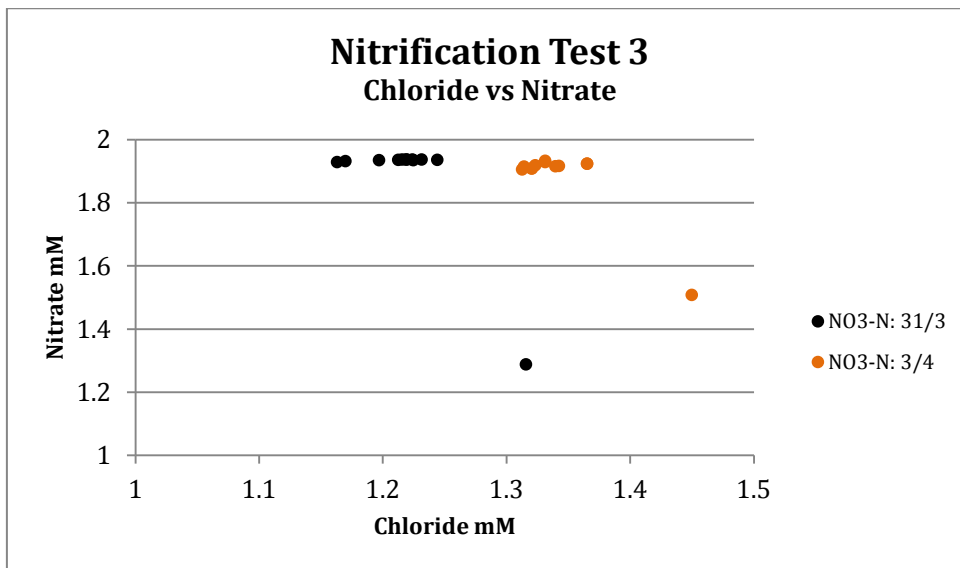
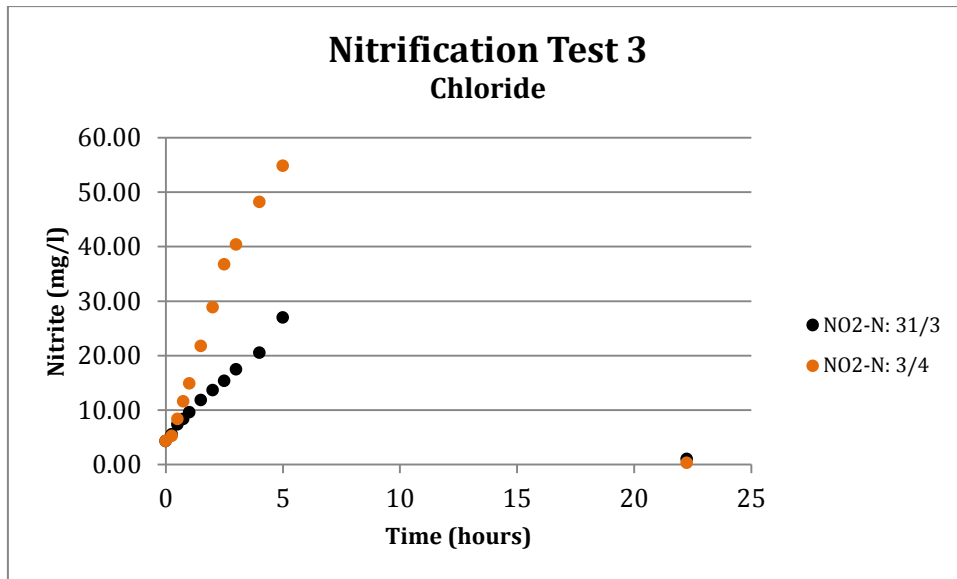
NORMALIZATION WITH SODIUM





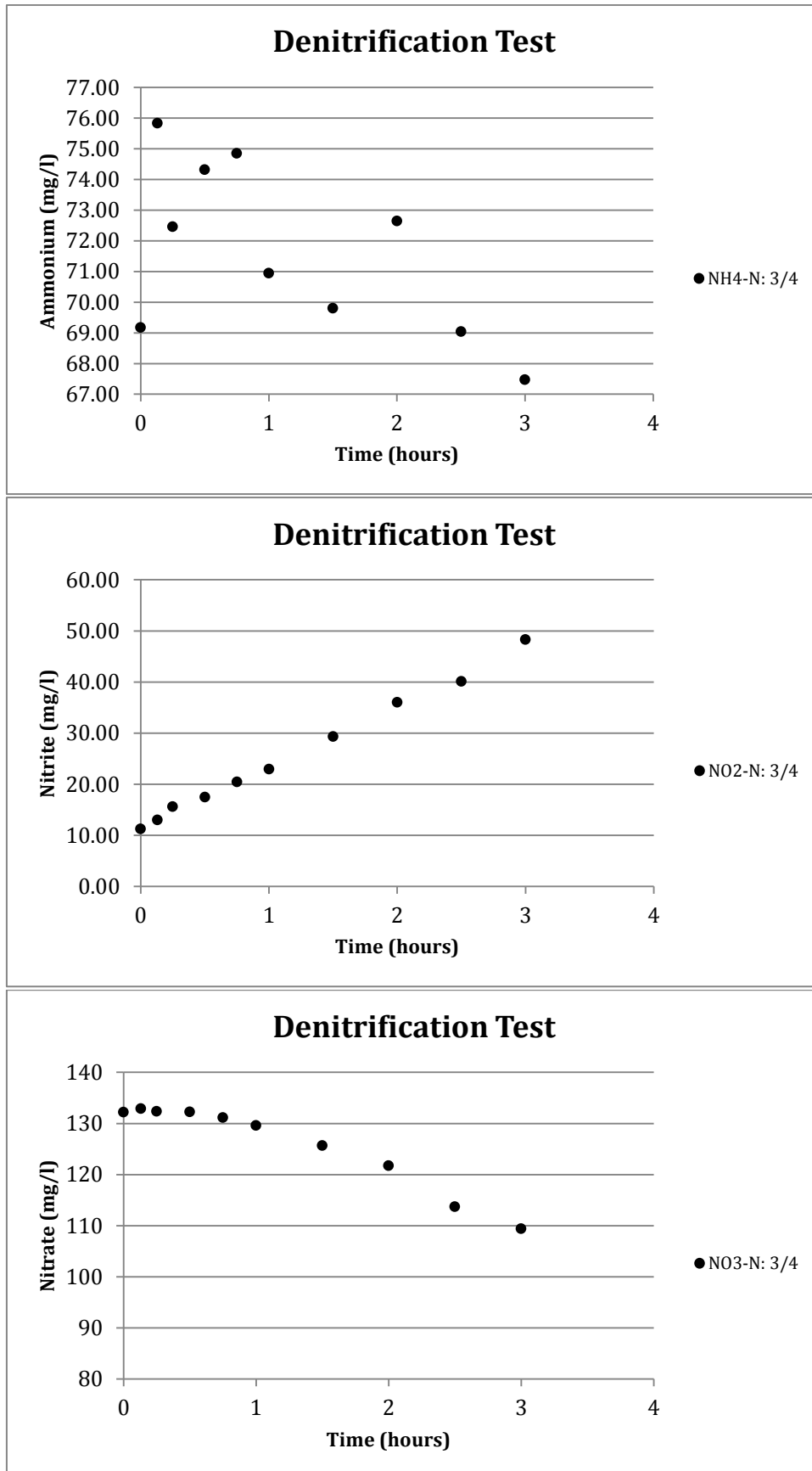
NORMALIZATION WITH CHLORIDE



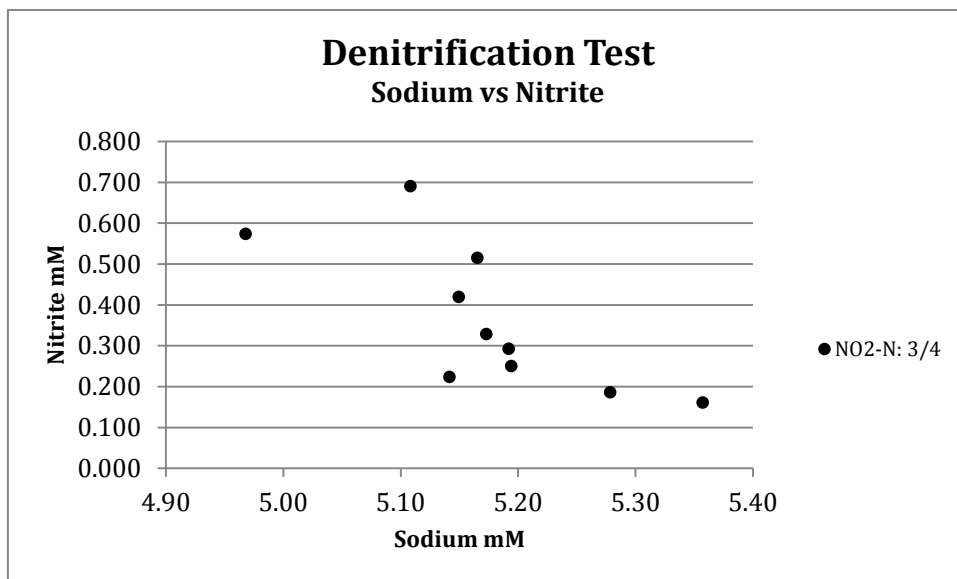
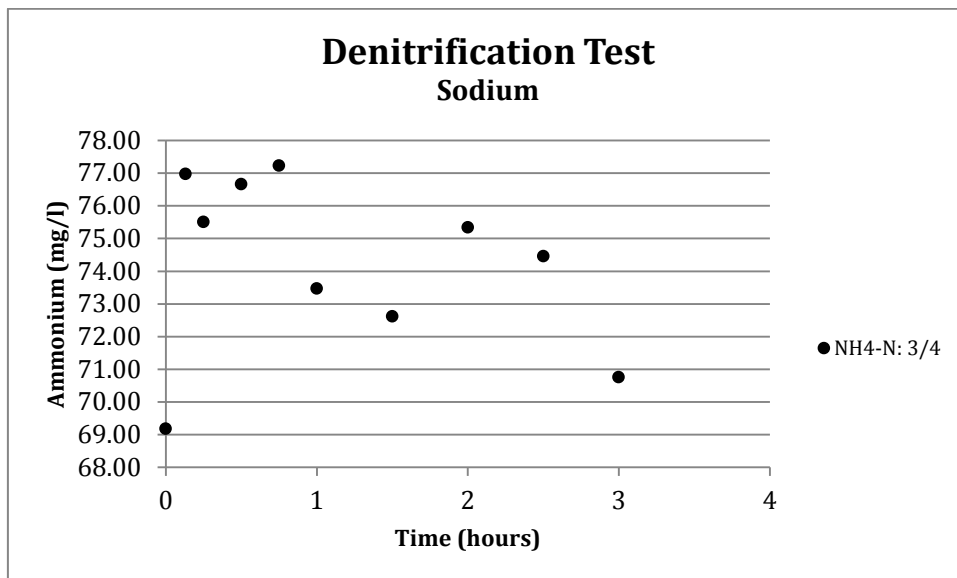
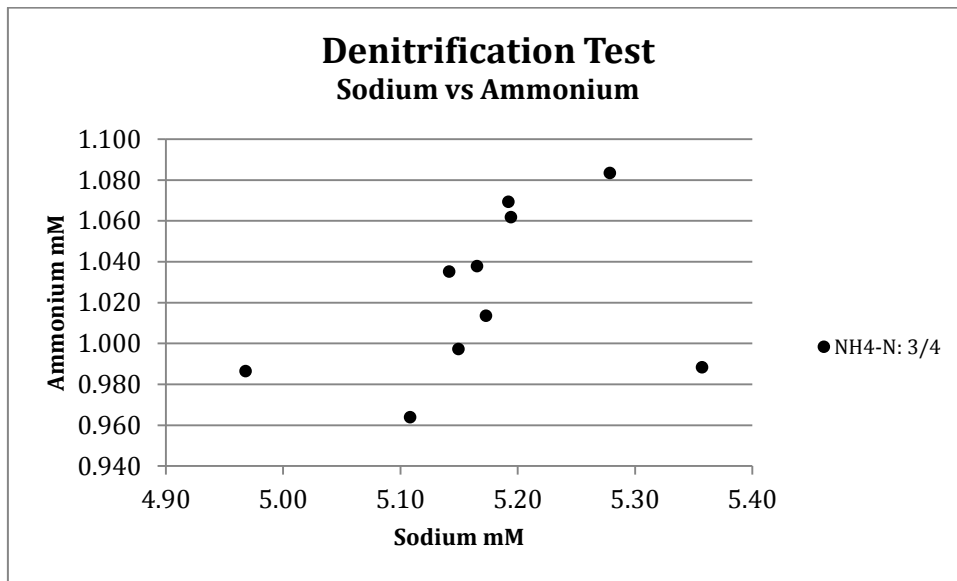


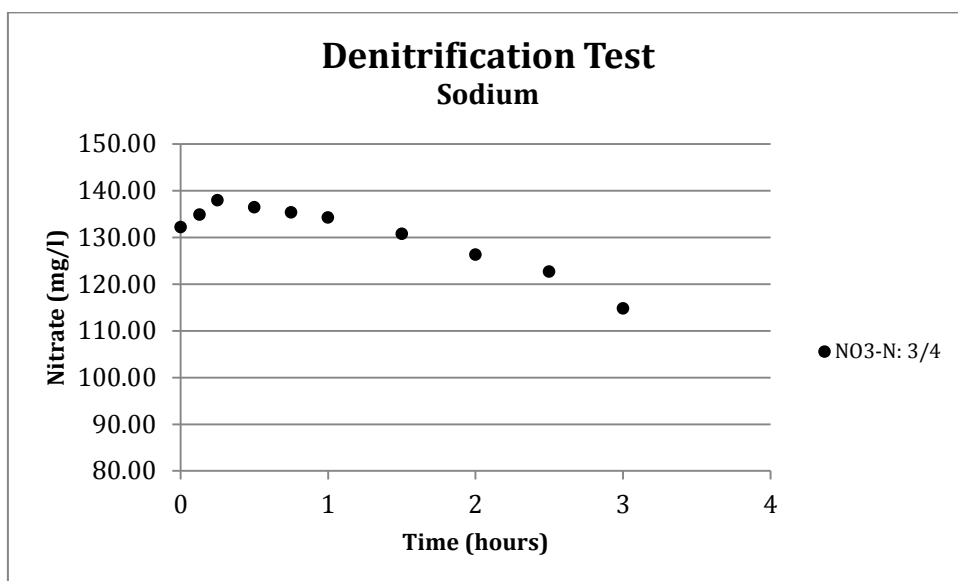
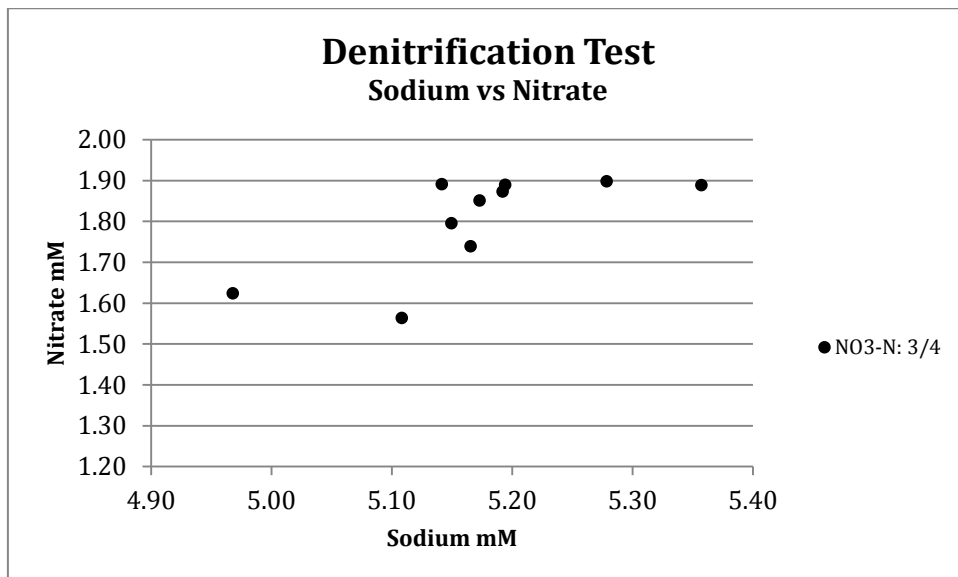
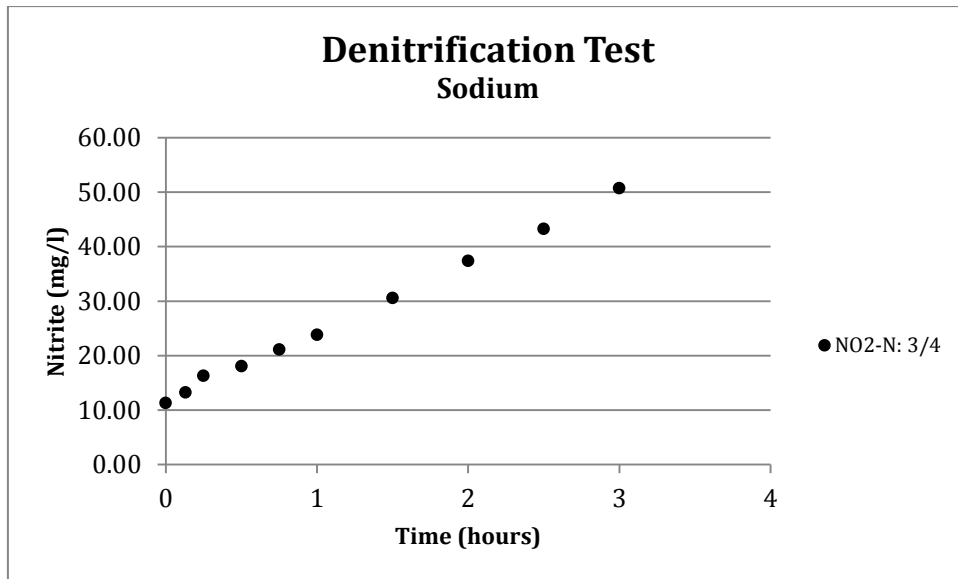
APPENDIX 4 – DENITRIFICATION TEST

TRENDS BEFORE NORMALIZATION



NORMALIZATION WITH SODIUM





NORMALIZATION WITH CHLORIDE

