Ammonia Wastewater Treatment by Immobilized Activated Sludge

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Abstract

The increase in urbanization has created the need for proper management and treatment of wastewater. The activated sludge process is an alternative method to treat high strength ammonia wastewater. In this project, the effect of temperature and the nitrification performances in treating ammonia wastewater by immobilized activated sludge in both batch culture and continuous mode methods were examined. Batch culture operated at higher temperature had a positive effect on the reduction of ammonia concentration by 75% of a specific sample.

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Table of Contents

Table of Tables

Table of Equations

Table of Figures

1.0 Introduction

One of the many byproducts of civilization is waste. Waste arises from households, industrial factories, and other facilities. To purge the unwanted wastes, sewage systems were created in populated areas. Sewage systems wash down the waste with water, disposing the resulting wastewater in the desired locations.

The increase in population and the expansion of cities have led to a greater disposal of wastewater into the environment. Improper disposal of wastewater has led to outbreaks of disease arising from wastewater in many parts of the world. These outbreaks increased the need for wastewater management and treatment, driving the demand for wastewater treatment to higher levels. The spark in the demand for wastewater treatment led to new innovations in the wastewater treatment field, creating new treatment technologies and system processes.

Eutrophication due to high levels of ammonia and nitrogen in wastewater is one of the main environmental problems associated with the improper dispose of wastewater. Eutrophication may further bring serious concerns such as the increase of chemical concentration in the ecosystem, the aggravation of water quality, and consequently these risk animal and human life (Qiao, Chen, & Zhang). One of the alternative methods to reduce eutrophication is the use of the activated sludge process to treat wastewater. This method generates an activated mass of microorganisms, which is used to stabilize wastes (McGraw-Hill). Immobilization of cells is another technique used for nitrogen removal in wastewater. The use of cell immobilization in treating wastewater has been the solution to many problems encountered in other types of wastewater treatment methods. This technology has a great impact on the

nitrification process, which result in an enhanced nitrification performance and consequently improvements in water quality.

Researches point out some advantages and disadvantages of using only one technology to treat wastewater. Three major breakthroughs were the creation of the activated sludge process, immobilization of cells technique, and the use of ultraviolet technology in wastewater treatment. However, all of these technologies have some disadvantages related to them. A more effective wastewater treatment system is one that embraces different technologies and applies them with the purpose to bring an effective solution to a problem. In this research, activated sludge, the immobilization system and UV technology are used in order to treat synthetic ammonia wastewater. Activated sludge was immobilized by UV technology in order to treat ammonia wastewater.

The purpose of this research is to study the immobilization of activated sludge in poly (ethylene glycol) by ultraviolet technology, its application in treating ammonia wastewater by examining nitrification performances, and to analyze the effect of temperature in wastewater treatment. Both batch culture and continuous mode methods were implemented for comparison and observation of nitrification performances on the reduction of ammonia concentration. Temperature was also manipulated in order to examine the effect of temperature in treating ammonia wastewater using batch and continuous mode methods. Furthermore, total organic compound (TOC), total nitrogen (TN) and the hydraulic retention time (HRT) were analyzed for continuous mode method.

2.0 Background

In order to fully understand the effect of using immobilization of activated sludge in polyethylene glycol by UV technology and its application in micro-polluted wastewater, it is indispensable the study involving such technologies. This section covers the three main types of technologies associated to the project proposal. Wastewater treatment technologies discussed include activated sludge process, immobilization techniques and ultraviolet (UV) radiation system.

2.1 Activated Sludge Process:

The increase in population and the development of new cities have created the need for the proper management and treatment of wastewater. In the middle of the nineteenth century, waterborne diseases were widespread in England causing many deaths (Bitton). These deaths helped increase the awareness of microorganisms in diseases, creating a greater demand for wastewater treatment. To meet this growing demand for wastewater treatment legislations were passed to ensure and encourage proper treatment and disposal of wastewater with the construction of wastewater treatment plants.

Wastewater can arise from both household sewage, and industrial wastes. For both cases it is essential to treat the waste water before disposing in nature. The wastewater is further classified as nontoxic or toxic wastes (Obayashi & Gorgan). Nontoxic wastes are mainly food industry waste, and domestic sewage. While toxic wastes are from coal processing, petrochemical, pesticide, pharmaceutical, and electroplating industries. These two types of

wastewater are treated differently, each one of them requiring various steps for the cleansing process.

Treating wastewater is done by physical forces, chemical and biological processes (Bitton). Physical forces treatment methods or unit operations include screening, sedimentation, filtration, and flotation. Chemical and biological methods or unit processes include disinfection, absorption, precipitation, degradation of organic matter, and removal of nutrients.

The wastewater treatment process objective is to reduce the organic content of wastewater, remove or reduce nutrients, remove or inactivate pathogenic microorganisms and parasites (Bitton). To achieve these objectives, there are four major steps that should be taken:

> 1. Preliminary treatment. The objective of this operation is to remove debris and coarse materials that may clog equipment in the plant.
>
> 2. Primary treatment. Treatment is brought about by physical processes (unit operations) such as screening and sedimentation.
>
> 3. Secondary treatment. Biological (e.g.,activated sludge, trickiling filter, oxidation ponds) and chemical (e.g., disinfection) unit processes are used to treat wastewater. Removal of nutrients also generally occurs during secondary treatment of wastewater.
>
> 4. Tertiary or advanced treatment. Unit operations and chemical unit processes are used to further remove BOD, nutrients, pathogens and parasites, and sometimes toxic substances.
>
> (Bitton)

A schematic of the wastewater process is shown below:
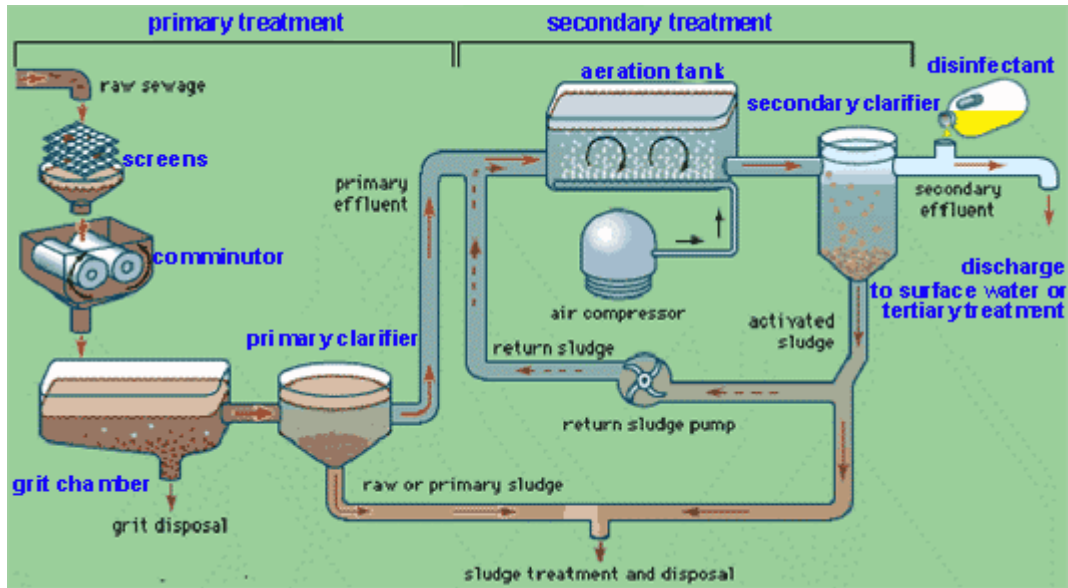
Figure 1 Schematic of the Wastewater Treatment

(Earthpace)

The process of activated sludge first started in England in 1914 by Ardern and Lockett and then this idea was spread worldwide (Bitton). This new method of treating wastewater was known as activated sludge since this process generated an activated mass of microorganisms, which was used in stabilizing a waste (McGraw-Hill). Activated sludge is a suspended-growth process that consists of aerobic treatment that oxidizes organic matter to CO_2 and H_2O, NH_4, and new cell biomass. It is commonly used as a secondary biological treatment for domestic wastewaters. Activated sludge process includes an aeration tank, sedimentation tank, mixed-liquor suspended solids (MLSS), mixed liquor volatile suspended solids (MLVSS), and can furthermore be used to calculate the food-to-microorganism ratio, hydraulic retention time (HRT), and sludge age (Bitton). Figure 2 below is a picture of a wastewater treatment plant where the activated sludge process is implemented.
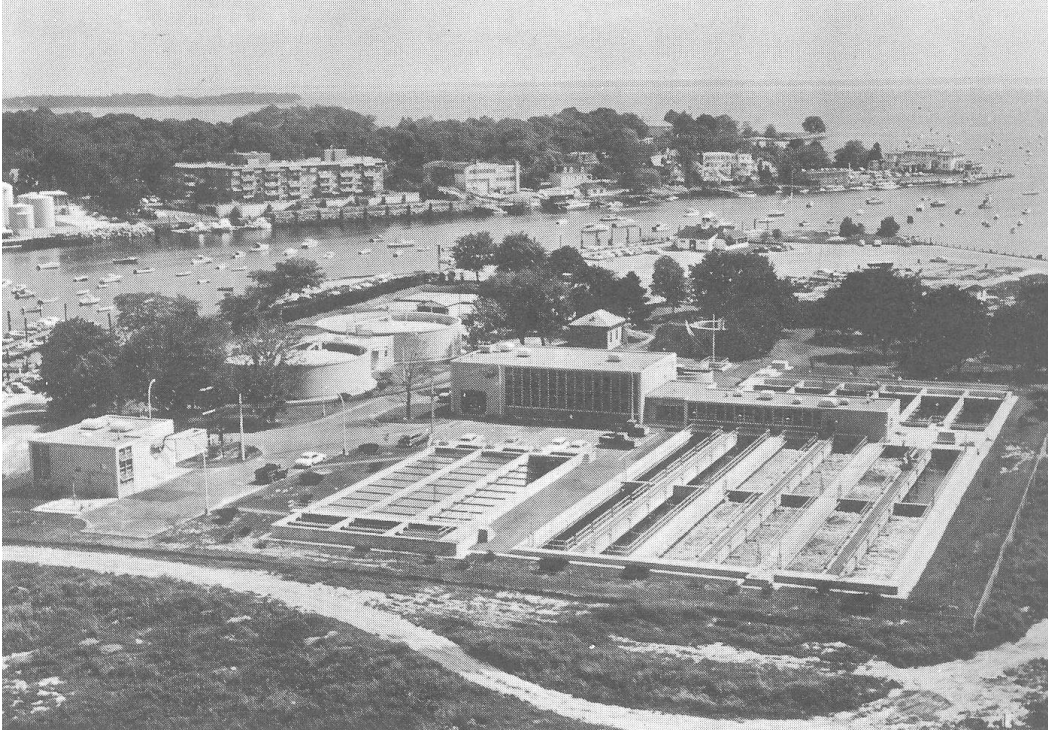
Figure 2 Activated Sludge Process Treatment Plant

(McGraw-Hill)

The activated sludge process is a system used for the treatment of sewage and industrial wastewaters that involves the mixture of biological mass and wastewater. In activated sludge process, organic waste is fed to the system and leaves the process depending on the desired treatment efficiency set by the operator. The process begins by mixing the biological waste present in industrial wastewater or sewage with an aerobic bacterial culture in the reactor and air (Eckenfelder). This mixture is known as the mixed liquor. Once in the reactor, the mixed liquor is aerated for a particular period of time in order to ensure that this solution is fully mixed (McGraw-Hill). Furthermore, this mixture undergoes separation through the gravity clarifier, where the waste activated sludge is removed from the treatment and mixed with primary treated wastewater before it is recycled back to the beginning of the process in order to maintain the desired concentration of organisms and sludge (Eckenfelder). Lastly, the sludge goes through

6

further treatment and the result of all this process is the treated wastewater that can be safely

disposed to nature. This process is illustrated in Figure 3 below.
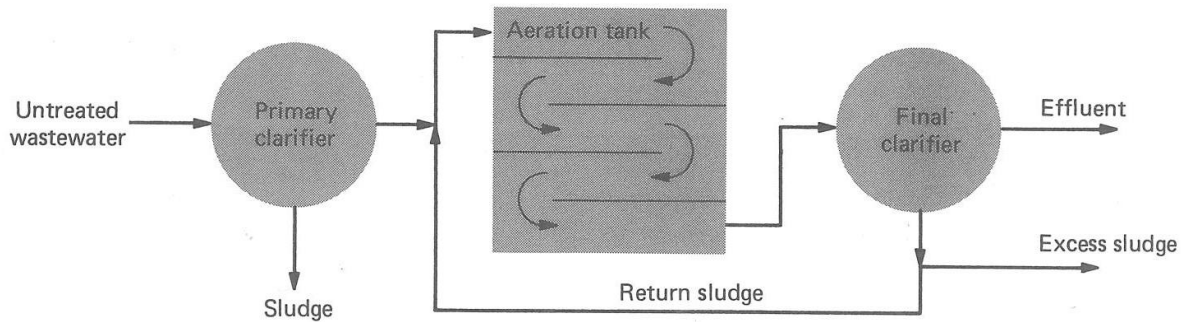
Figure 3 Conventional Activated Sludge Process

(McGraw-Hill)

When choosing the activated sludge process for wastewater treatment, special attention

needs to be paid for the amount of substrate removal such as the biological oxygen demand

(BOD) and chemical oxygen demand (COD). Other essential variables include mass of

microorganisms in the system and the non-biodegradable inert suspended solids in the influent to

the system (Turovskiy & Mathai). A relationship can be established between these variables in

the following equations:

Equation 1 Net Growth of Biomass

$$P_x = Y(S_0 - S) - k_d X$$

Equation 2 Total Waste Activated Sludge Solids

$$WAS = P_x + I_0 - E_t$$

Where:

P_x = net growth of biomass expressed as volatile suspended solids (VSS), kg/d or lb/d

Y = gross yield coefficient, kg/kg or lb/lb

S_0 = influent substrate (BOD or COD), kg/d or lb/d

S = effluent substrate (BOD or COD), kg/d or lb/d

k_d = endogenous decay coefficient, d^{-1}

X= biomass in aeration tank (MLVSS), kg or lb

WAS = total waste activated sludge solids, kg/d or lb/d

I_0= influent nonvolatile suspended solids, kg/d or lb/d

E_t = effluent suspended solids, kg/d or lb/d

(Turovskiy & Mathai)

Figure 4 shows how these variables are related in an activated sludge process.

Influent
Q = Flow
S_0 = Substrate (BOD or COD)
M_0 = Total Suspended Solids (TSS)
I_0 = Inert (Nonvolatile) TSS

Aeration Tank
V = Volume
M = Mixed Liquor
    Suspended Solids
    (MLSS)
X = Biological Solids
    (MLVSS)

Final Clarifier

Effluent
Q_e = Flow
S = Substrate
I_e = TSS

Return Activated Sludge (RAS)
Q_r = Flow

Waste Activated Sludge (WAS)
Q_w = Flow
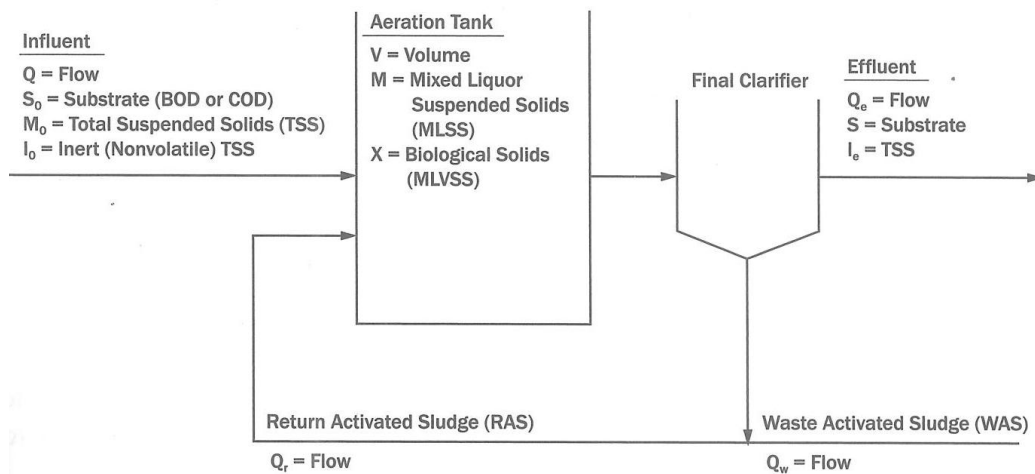
Figure 4 Flow Diagram of a Typical Activated Sludge System

(Turovskiy & Mathai)

The activated sludge method is applicable to a series of specific wastewater treatment procedures. One application of this method is on treating high strength ammonia wastewater. The treatment of high strength ammonia wastewater is crucial to the environment since improper treatment may bring serious environmental problems or eutrophication, which results from the increase of chemical concentration in the ecosystem thus endangering animal life and affecting water quality significantly (Qiao, Chen, & Zhang). Researches including cost balance analysis

and efficiency suggest that the activated sludge process is a favorable method since it provides

high efficiency and low cost (Qiao, Chen, & Zhang).

2.1.1 Types of Activated Sludge Processes

The activated sludge is characterized into two different processes: single-stage and two-stage processes. In both processes, four key elements are essential and if any of them malfunction, the whole process may be put in jeopardy. This is further explained in the table below:

Table 1 Four Elements in Activated Sludge Plant

- **An aeration tank equipped with appropriate aeration equipment, in which the biomass is mixed with wastewater and supplied with oxygen.**
- **A final clarifier, in which the biomass is removed from the treated wastewater by settling or other means.**
- **Continuous collection of return sludge and pumping it back into the aeration tank.**
- **Withdraw of excess sludge to maintain the appropriate concentration of mixed liquor.**

(Kayser)

2.1.1.1 Single Stage Process

The single stage process draws its history back to 1910, when the concept of the aeration technique in treating wastewater was first introduced. In this process, wastewater undergoes aeration for a specific period with regular stops, so that the suspended wastewater may settle and more wastewater can be added to the system before aeration starts again (Kayser). This cycle is repeated until the effluent is fully nitrified and a desired amount of activated sludge or settled

sludge is achieved. This whole process may take about six hours for total completion (Kayser).

The diagram (Figure 5) shows a conventional mode for an activated sludge plant:
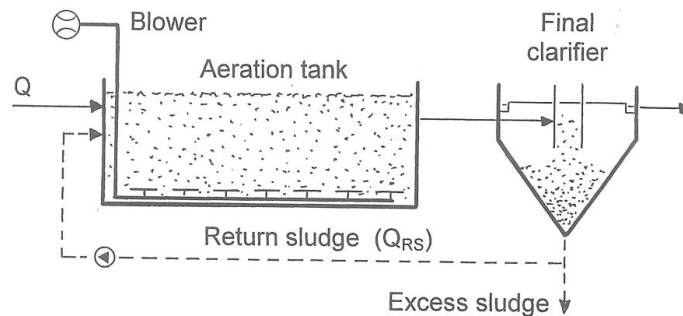
Figure 5 Schematic of Single Stage Activated Sludge Process

(Kayser)

2.1.1.2 Two Stage Process

Two stage process implements the idea proposed by the single stage process in addition to adding a second activated sludge plant to the system. Basically, the two stage process is a combination of two independent activated sludge plants which work in series (Kayser). It is designed in such a way that the first activated sludge plant has a higher sludge loading rate (F/M), since it also receives excess sludge from the second stage, on the other hand, the second plant has a lower sludge loading rate (F/M) (Kayser). Figure 6 is a flow diagram of the two stage process.
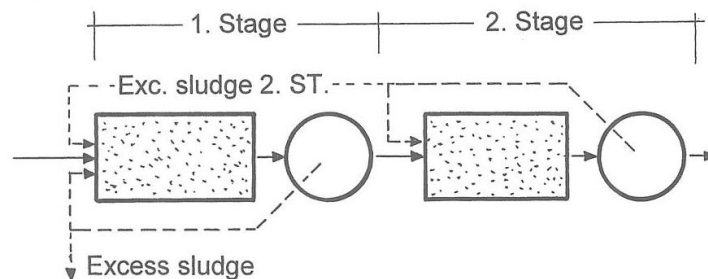
Figure 6 Schematic of Two Stage Activated Sludge Process

(Kayser)

The two stage process has some advantages when comparing it to the single stage process. Since it contains two independent activated sludge plants, it is more efficient in removing harmful substances. These substances are mostly removed in the first stage and if any trace is left, it can be completely removed in the second stage. Removal of harmful substances is extremely important in the treatment of industrial wastewater. In addition to this, bulking sludge formation is rarely noticed and high sludge age microorganism may facilitate the removal of biodegradable organisms and also oxidize ammonia (Kayser).

Since the creation of the original activated sludge process, several other processes are used today. These different versions of activated sludge systems according to specific uses are listed in Table 2:

Table 2 Activated Sludge Processes

Type	Common Name	Use
Aerobic processes: Suspended growth – Activated sludge processes	Conventional (plug flow)	Carbonaceous BOD removal (nitrification)
	Continuous – flow stirred tank	
	Step aeration	
	Pure oxygen	
	Modified aeration	
	Contact stabilization	
	Extended aeration	
	Oxidation ditch	

(McGraw-Hill)

2.1.2 Aeration Systems

There are innumerous types of aerators adapted to activated sludge processes. Some of the different types of aeration systems include the conventional, continuous-flow stirred-tank and pure oxygen processes.

Conventional

The conventional activated sludge process is the simplest process described earlier in the activated sludge process section. Equipments used in this system include an aeration tank, where influent wastewater and recycled sludge are mixed, a secondary clarifier and a sludge recycle line (McGraw-Hill). Plug flow with cellular recycle model is applied, where both the influent wastewater and the recycled sludge are aerated for a time period of six hours (McGraw-Hill).

Continuous-flow stirred-tank

In the continuous-flow stirred-tank process, the influent wastewater and the return sludge pass through several parts of the aeration tank. The mixed liquor is also aerated, which helps to balance the organic load in the system (McGraw-Hill). When the influent wastewater and the return sludge exit the tank, they are deposited into the activated sludge the settling tanks. Figure 7 below is a schematic of the continuous-flow stirred-tank activated sludge process.
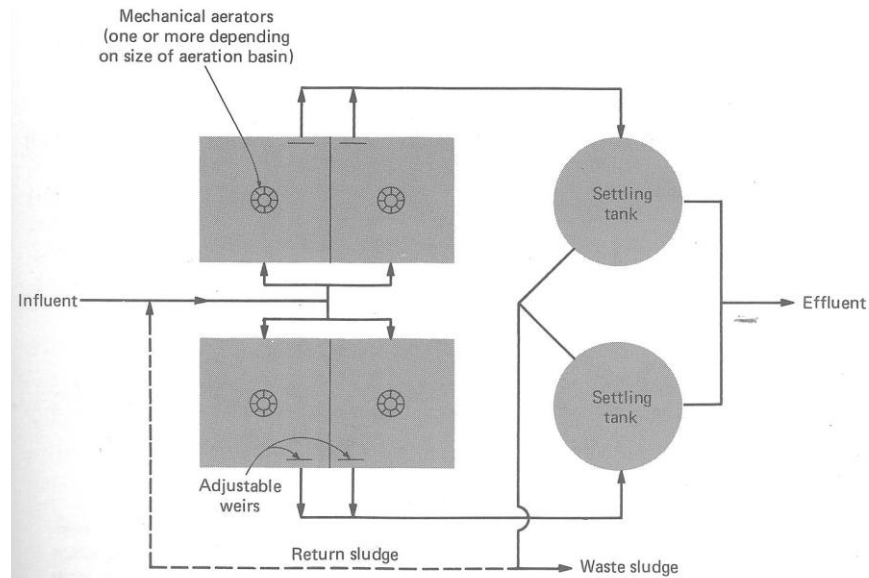
Figure 7 Schematic of the Continuous-Flow Stirred-Tank Process

(McGraw-Hill)

Pure oxygen

The pure oxygen system uses a series of covered continuous-flow stirred-tank reactors, where oxygen is constantly circulating throughout the process. The idea of using pure oxygen instead of air, as it was implemented in traditional activated sludge process, was adopted by several treatment plants since 1970 due to its high performance in treating wastewater (McGraw-Hill). In this process, carbon dioxide is released and more oxygen needs to be added depending on how much oxygen the microorganisms need for their activity. Furthermore, Henry's Law suggests that given the mole fraction of oxygen above the liquid to be 0.8, the amount of oxygen in the liquid needs to be four times the amount of air put in the traditional activated sludge system (McGraw-Hill). Figure 8 below is a schematic of pure oxygen activated sludge process in series.
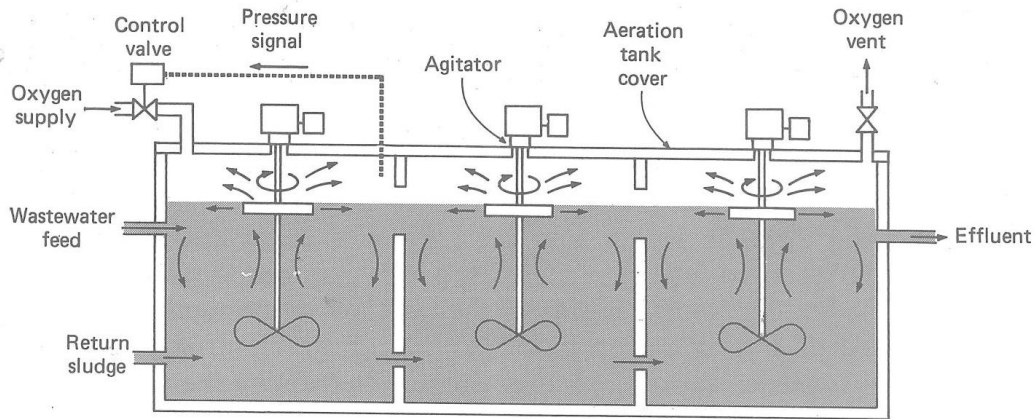
Figure 8 Schematic of Pure Oxygen Process

(McGraw-Hill)

2.1.3 Nitrification and Denitrification

Nitrate brings serious harms to the environment including eutrophication, humans and drinking water. This concern was further reinforced with the development of technologies to remove total nitrogen from wastewater (The Water Planet Company). Bacteria are used to convert ammonia and nitrate to gaseous nitrogen, so that it can be released into the air. Nitrification and denitrification processes are carried out in the wastewater treatment system to remove nitrogen from wastewater (Kayser).

The biological conversion of ammonia to nitrogen gas can be accomplished by a two step process in nitrification. The first step is to convert ammonia and ammonium to nitrite by the bacteria *Nitrosomonas* and then the nitrite is converted to nitrate by the bacteria *Nitrobacter* (U.S. Environmental Protection Agency (EPA)). Both of these bacteria work under an aerobic environment. The overall reactions are shown below:

Equation 3 Nitrification Step 1

$$NH_4^+ + 1.5\ O_2 \rightarrow NO_2^- + H_2O + 2H^+$$

14
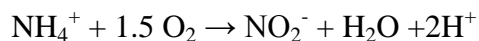
Equation 4 Nitrification Step 2

$$NO_2^- + 0.5\ O_2 \rightarrow NO_3^-$$
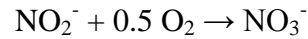
Equation 5 Nitrification Final Step

$$NH_4^+ + 2.0\ O_2 \rightarrow NO_3^- + H_2O + 2H^+$$
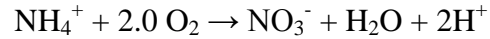
(Kayser)

After the nitrate is formed, it undergoes denitrification to be reduced to nitrogen gas. This process also involves the use of bacteria, however, it requires the dissolved oxygen (DO) level to be near or equal to zero. The reaction for denitrification is shown below. It is important to note that for a wastewater treatment plant that adopts the nitrification and denitrification processes to be effective, there is a need to design a system that will specific sections for aerobic and anaerobic processes.

Equation 6 Denitrification

$$2\ NO_3^- + 2H^+ \rightarrow N_2 + H_2O + 2.5\ O_2$$
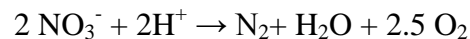
(Kayser)

2.2 Immobilization of Cells in Wastewater Treatment:

Cell immobilization techniques for removing unwanted chemicals in wastewater have been used since the 1980's. Cells taken from the activated sludge are entrapped in a support matrix where they go through the process of immobilization. This method has been studied in laboratories and applied to a sewage treatment plant in the 1990's, where immobilized cells were used in the removal of nitrogen (Chen, Lee, Chi, & Houng).

One of the uses of the immobilization of cells technique in wastewater treatment is the immobilization of activated sludge. Although the activated sludge process is seen as a favorable

wastewater treatment, there are also some disadvantages related to it. One of the negative points in using activated sludge is that most of the times slow growing organisms such as nitryifiers and anaerobic methane producers take long to be entrapped by the system. This process also requires a greater biomass concentration for a higher efficiency to be attained. However, the result of municipal wastewater treatment provides a much diluted feed stream with very low biomass concentration. A solution to this problem is to use the cell immobilization system in activated sludge process which will increase biomass retention time, thus enabling the reactor to achieve a higher capacity and efficiency in the system (Chen, Lee, Chi, & Houng). Cell immobilization process may also assist in eliminating unwanted elements in a wastewater treatment, in facilitating a solid-liquid separation in a settling tank thus avoiding bulking (Chen, Lee, Chi, & Houng).

The use of cell immobilization in treating wastewater has been the solution to many problems encountered in other types of wastewater treatment methods. Some wastewater treatment plants use the biological nutrients removal (BNR) process for nitrogen removal as it brings water quality concerns. In BNR process, suspended biomass is used to remove organic carbon from the wastewater entering the treatment process. Although some treatment plants adopted this process since it is fairly economical, it has been noted that it brings several limitations that affect the process's efficiency (Chang, Kim, & Nam). One of the most alarming concerns about using BNR process is that slow growing nitrifying bacteria require a long sludge retention time (Chang, Kim, & Nam). A solution to this problem is to use immobilization techniques since it was proven that this form of treating wastewater has brought many contributions to the efficiency in wastewater treatment. Some of the positive features about the immobilization system are that it maintains a high cell concentration, has a better solid-liquid

separation and is less sensitive to temperature (Chang, Kim, & Nam). All of these advantages have a great impact on the nitrification process, which result in an enhanced nitrification and consequently improvements in water quality.

2.2.1 Types of Immobilization Techniques

Cells can undergo the immobilization technique by either encapsulation, which is sometimes known as entrapment, and attachment.

Immobilization by encapsulation has been mostly chosen for immobilization of living cells. In this method, cells are immobilized with the use of porous polymeric materials such as alginate, agar, polyacrylamide, carrageenan, cellulose acetate and poly-vinyl alcohol (PVA), which is carried out by ionotropic or thermal gelation (Orive, Ponce, Hernandez, Gascon, Igartua, & Pedraz) . Other materials used in encapsulation method also include polymer gels, microcapsules, liposomes, hollow fibers and ultrafiltration membranes (Tanaka & Nakajima). According to a study on the structure of the materials used in immobilization, "The porous structure of polymers allows substrates and oxygen to diffuse into the internal pores where nitrification is carried out by the entrapped cells" (Chang, Kim, & Nam). There is a wide range of choices for materials that can be used in immobilization by encapsulation method; however, PVA is the most used material. PVA is preferred over other materials since the freezing-thawing method, a simple technique that does not require chemical initiation, can be applied (Serrano, Palacio, Trevino, & Esparza). In freezing-thawing, the cross-linking procedure produces an elastic and non-water-soluble hydrogel (Tanaka & Nakajima). If PVA is used in treating wastewater with the encapsulation method, some physical stability requirements such as

solubility, biodegradability, diffusivity and mechanical stability, need to be met (Chang, Kim, &
Nam). Special consideration need to be made on the solubility of PVA to water since PVA is
considered to be hydrophilic.

The other type of immobilization is by attachment, which has been favored over
encapsulation for wastewater treatment. The attachment method involves the attachment of
biomass to porous support materials such as polyurethane foam and inorganic matrix (Chang,
Kim, & Nam). One advantage in using the attachment method is that are no need for chemical
additions. Although the attachment method has been preferred, the encapsulation method can
produce much higher cell concentrations and higher nitrification rates for PVA (Chang, Kim, &
Nam, 2005).

2.2.2 Advantages and Disadvantages of the Immobilization System

There are a list of advantages and disadvantages associated with the use of the
immobilized cell system in wastewater treatment. This is presented in Table 3 below. As it is
seen from the table, although there are a number of disadvantages in using this technology to
treat industrial/sewage water, the positive points about using it may overcome the limitations
defined in the disadvantages section. "Some of these limitations can be overcome, especially in
the case of immobilization of nonviable cells, where the cells are used mainly as sources of
catalysts in the bioconversion" (Tyagi & Vembu).

Table 3 Advantages and Disadvantages of the Immobilization System

Advantages	Disadvantages
• It allows the recycling of the biological catalyzers	• The undesirable side reactions
• It allows the reactor to function at very high cell concentration, without rheological or mass transfer limitations	• Inhibition of certain metabolic activities due either to product accumulation or some toxic substances accumulation
• There is a decrease in the metabolic regulation effect due to product accumulation	• The diffusional limitation of the substrates, mainly those of high molecular weight. This is one of the major limitations in the case of entrapped cells
• A better utilization of the substrate even at low concentrations, thanks to the localized concentration of nutrients and hydrolytic coenzymes at the support-substrate/interface	• The cell leaking from the solid support
• The possibility of using the cells in their stationary phase where only the metabolic chains are active	

(Tyagi & Vembu)

2.3 Ultraviolet Technology in Wastewater Treatment:

The Ultraviolet (UV) radiation system has been used to treat water by destroying pathogens and bacteria found in wastewater. This system emits electromagnetic energy and operates at low or medium pressure mercury lamps protected by a quartz tube layer (Bitton). UV technology is classified into two main types: UV technology using low pressure lamp and UV technology using medium pressure lamp with high or low intensities. For the low pressure lamp technology, there is a monochromatic UV output of 254nm and the system involves an open

channel with immersed lamps in the water. On the other hand, for medium pressure lamp technology, there is a polychromatic UV output of 185-400nm and consists of a closed pipe system with lamps set along the treatment chamber (Berson UV-techniek). In general, UV radiation travels with a wavelength of a peak of 265nm and penetrates the cell membrane of pathogens, thus destroying their genetic material (DNA and RNA) and disabling them from reproducing (U.S Environmental Protection Agency (EPA)).

2.3.1 Low Pressure Lamp UV Technology

Low pressure lamp UV technology designed in open channel has been preferred over medium pressure lamp for a long time. For this type of system, UV radiation travels at the germicidal wavelength of 2,537 A and inactivates the microbial organisms (Bitton). Inactivation of pathogens by UV radiation is due to thymine dimerization, which destroys their DNA and causes inefficiency for DNA to reproduce, thus avoiding them from spreading through the effluent. This inactivation can be calculated using the following equation:

Equation 7 Inactivation of Pathogens by UV

$$\frac{N}{N_0} = e^{-KPdt}$$

Where:

N_0=initial number of microorganisms (#/mL)

N= number of surviving microorganisms (#/mL)

K= inactivation rate constant ($\mu W*s/cm^2$)

P_d = UV light intensity reaching the organisms ($\mu W/cm^2$)

t= exposure time (s)

(Bitton)

According to the EPA, "the effectiveness of a UV disinfection system depends on the characteristics of the wastewater, the intensity of UV radiation, the amount of time the microorganisms are exposed to the radiation, and the reactor configuration" (U.S Environmental Protection Agency (EPA)). Studies show that the efficacy of UV disinfection varies from types of pathogens and their resistance to UV radiation. Their resistance can be classified as follows: protozoan cysts> bacterial spores> viruses> vegetative bacteria (Bitton). This classification and further details on 90% of inactivation of pathogens can be observed on Table 4:

Table 4 Dosage for 90% Inactivation of Selected Microorganisms by UV

Microorganism	Dosage (μW-s/cm^2)
Bacteria	
E. coli	3,000
Salmonella typhi	2,500
Pseudomonas aeruginosa	5,500
Salmonella enteritis	4,000
Shigella dysenteriae	2,200
Shigella paradysenteriae	1,700
Shigella flexneri	1,700
Shigella sonnei	3,000
Staphylococcus aureus	4,500
Legionella pneumophila	380
Vibrio cholera	3,400
Viruses	
Poliovirus 1	5,000
Coliphage	3,600
Hepatitis A virus	3,700
Rotavirus SA 11	8,000
Protozoan cysts	
Giardia muris	82,000
Giardia lamblia	63,000
Acanthamoeba castellanii	35,000

(Bitton)

2.3.2 Medium Pressure Lamp UV Technology

Medium pressure lamps offer more advantages both economically and effectively than low pressure lamps. For a long time low pressure lamps have being preferred and adapted to wastewater treatment plants, however, new developments in medium pressure lamps are

changing this scenario. Since they are designed in compact closed-pipe system, they are much easier to adapt to different environments and occupy less space than the designed open-pipe low pressure lamp system (Berson UV-techniek). A small working space required is a crucial benefit for medium pressure lamps since a large treatment plant is not needed. This reflects a plus in the plant's cost since a larger treatment plant will require more space and money to be invested. Medium pressure lamps also produce higher UV intensity (15 to 20 times more) when compared to low pressure lamps, which adds favorability over low pressure lamps (Berson UV-techniek). With a higher germicidal UV intensity produced, disinfection is faster and fewer amounts of lamps is needed, thus saving more on investments. Unfortunately, the disadvantage of a lamp that has a higher intensity is that it also requires higher temperatures which lead to a higher energy consumption (U.S Environmental Protection Agency (EPA)).

Other factors that favor medium pressure lamps include maintenance and design. Closed-pipe medium pressure systems give better accessibility for workers to clean and to do maintenance. These pipes are enclosed by a quartz layer, which needs to be cleaned from time to time to avoid fouling and to ensure a high performance (Berson UV-techniek). For closed-pipe systems, a mechanical wiper that moves up and down, which makes it very convenient and easy to clean, is used. In open-channel system the same kind of cleaning aid cannot be used and chemical cleaning is indispensable, which demands more investment and time (Berson UV-techniek). For this type of system, personnel need to manually clean the lamps and remove them for further cleaning in an acid bath, which poses a great danger in exposure to UV light (Berson UV-techniek). In addition to a more favorable maintenance system, the design of a closed-pipe system also provides a safer environment to the workers in the treatment plant due to an enclosed

system which may prevent people from being exposed to ultraviolet radiation (Berson UV-techniek).

Studies show that medium pressure lamps are more efficient in destroying microorganisms since they suggest that damages are irreversible after being treated by UV technology compared to low pressure lamps where these microorganisms may regenerate (Berson UV-techniek). Furthermore, low pressure lamps generally use amalgam lamp types, which use electronic ballasts that need to be replaced at a regular basis since they were proven to constantly give problems to the system (Berson UV-techniek). Taking into account that low pressure lamp systems require a greater number of lamps to attain the same intensity provided by medium pressure lamp system, failures in the electronic ballasts may increase the plant's cost exponentially. Moreover, researches show that low pressure lamps are not capable to perform well when operated at the extremes high or low water temperatures and their hydraulic system is not at its high performance, resulting in a not well dispersed UV radiation throughout the effluent (Berson UV-techniek).

2.3.3 Advantages and Disadvantages of Using UV Technology

UV technology has advantages and disadvantages related to its effects in treating wastewater. The following table from EPA Fact Sheet on ultraviolet disinfection summarizes the main advantages and disadvantages of using UV technology.

Table 5 Advantages and Disadvantages of Using UV Disinfection

Advantages	Disadvantages
• UV disinfection is effective at inactivating most viruses, spores, and cysts.	• Low dosage may not effectively inactivate some viruses, spores, and cysts.
• UV disinfection is a physical process rather than a chemical disinfectant, which eliminates the need to generate, handle, transport, or store toxic/hazardous or corrosive chemicals.	• Organisms can sometimes repair and reverse the destructive effects of UV through a "repair mechanism," known as photo reactivation, or in the absence of light known as "dark repair."
• There is no residual effect that can be harmful to humans or aquatic life.	• A preventive maintenance program is necessary to control fouling of tubes.
• UV disinfection is user-friendly for operators.	• Turbidity and total suspended solids (TSS) in the wastewater can render UV disinfection ineffective. UV disinfection with low-pressure lamps is not as effective or secondary effluent with TSS levels above 30 mg/L.
• UV disinfection has a shorter contact time when compared with other disinfectants (approximately 20 to 30 seconds with low-pressure lamps).	• UV disinfection is not as cost-effective as chlorination, but costs are competitive when chlorination dechlorination is used and fire codes are met.
• UV disinfection equipment requires less space than other methods.	

(U.S Environmental Protection Agency (EPA))

2.3.4 UV Technology in China

UV Disinfection treatment technology, a clean technology with no unwanted by-products, is common place in first world countries such as Europe and North America. The UV treatment technology has been improving in China. China first installed a municipal UV system

in 2001, yet in eight years the number of UV system in China jumped from one to one hundred, showing a rapid growth (Berson UV-techniek). UV disinfection treatment technology is mostly used to remove harmful pathogen from the drinking water. Recent breakthroughs in the UV technology have expanded the treatment to reduce pesticides and other contaminants from underground water (Berson UV-techniek).

3.0 Methodology

The goal of this project is to examine the effect of temperature and nitrification performances in treating ammonia wastewater using both batch culture and continuous mode methods. Background research was essential for the understanding of the main technologies related to this project: immobilization of cells, activated sludge and the use of ultraviolet radiation in wastewater treatment. In order to carry on this research project, a methodology was developed with the purpose to identify the details involved in the process of studying this particular immobilization of activated sludge process. This chapter will focus on steps taken in examining the nitrification performances in batch culture and continuous mode methods. Furthermore, total organic compound (TOC), total nitrogen (TN) and the hydraulic retention time (HRT) were analyzed for continuous mode method.

3.1 Preparation of the Immobilized Pellets

The immobilized pellets were prepared prior to the experiment under the described procedure. Samples of concentrated activated sludge were collected from a municipal wastewater treatment plant facility in Minhang, Shanghai and were used as the basis for immobilization. After collection, a portion of the activated sludge, which represented approximately 4.8%, m/m, was added in a polyethylene glycol (PEG) pre-polymer solution (14%, m/m). The PEG solution contained a cross-linker N, N'-Methylenebisacrylamide (MBA) (0.5%, m/m). The specified

portion of activated sludge and the PEG solution were mixed thoroughly with a photo-initiator, Benzoin Dimethyl Ether (0.1%, m/m)[1]. Afterwards, the mixture was immediately put under the UV rays for 4 min to form an elastic gel. This gel was then solidified and cut into 3mm x 3mm x 3mm cubic pellets. After undergoing this process, the pellets were then known to be the immobilized activated sludge pellets. These pellets were prepared with components to enable them to undergo aeration. Their density was equivalent to 1.02 g/cm^3, which facilitated their suspension and movement in water while being aerated since it is only a little denser than water (Qiao, Chen, & Zhang).

3.2 Purpose of the Experiment

The purpose of this experiment was to observe nitrification performances and the effect of temperature using batch culture and continuous mode methods. This was accomplished by first acclimating immobilized activated sludge pellets prepared prior to the experiment with 40 mg/l synthetic ammonia wastewater in a 250 ml up-flow aeration bioreactor in both batch culture and continuous mode methods. Then, we examine ammonium nitrogen (NH_4+-N), nitrite nitrogen (NO_2^--N), and nitrate nitrogen ($NO3^-$-N) by salicylic acid hypochlorite spectrophotometer, N-(1-Naphthyl) ethylene diamine spectrophotometer and UV spectrophotometer. These methods were used to determine trace amounts of nitrite in all water samples tested. All reagents used to help in analyzing NH_4+-N, NO_2^--N, $NO3^-$-N were purchased

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[1] Professor Xiangli Qiao

from Shanghai chemical reagents Co., China and were directly used without any further treatment.

3.3 Pre laboratory Procedure

This section addresses the required preparation of immobilized activated sludge prior to analyzing the performances in treating ammonia wastewater in batch culture and continuous mode.

Set Up Procedure

Four up-flow aeration bioreactors were set up for this experiment. Each of them contained different kinds of immobilized activated sludge pellets (sample 02, sample 04, sample 05 and sample 06) and consequently they diverged in properties. The design of the reactors included a double walled glass column, where the inside wall was filled with 10% (v/v) of immobilized cell pellets which is approximately a volume of 250 ml (Refer to Figures 9 and 10). Hot water circulated between the space between the outer and the inner wall (Urra, Sepulveda, Contreras, & Palma). The hot water temperature was controlled by the temperature controllers and was manipulated to observe different responses and the effect of temperature in treating the synthetic ammonia wastewater. The system was oxygenated with a rate of dissolved oxygen (DO) equal to 5.2 mg/L.

Figure 9 Set Up of Aeration System

Figure 10 Up-Flow Aeration Bioreactors

Aeration in the reactors started by the circular movement of immobilized pellets from the center bottom to the top of the reactor, forming an up-flow inner circulation (Qiao, Zhang, Chen, & Chen). Pellets rose to the top of the reactor and then settled down with gravity. This established inner circulation helps in the mixing the synthetic ammonia wastewater and the activated sludge pellets completely. Pellets were aerated with the prepared ammonia wastewater

29

for a specific amount of time and final treated wastewater was removed from the process from a tube attached to the outside wall of the reactor.

In continuous mode method, there was an addition of a pump to the system. Since the pump could only be attached to one reactor at a time, only one kind of activated sludge was tested. The same set up was used; however, there was a tube attached to another outlet in the outside wall of the reactor where the prepared wastewater could be pumped to the bioreactor. See Figure 11 for continuous mode set up.
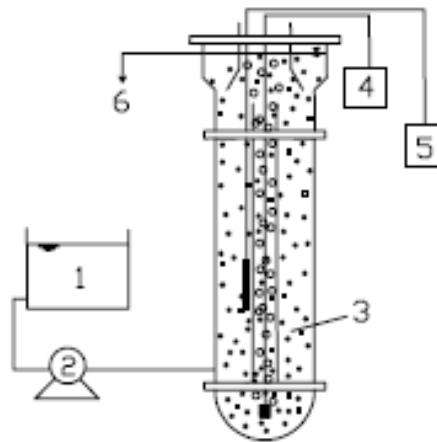
Figure 11 Schematic of Set Up for Continuous Mode Method

1. Feed tank
2. Pump
3. Bioreactor with immobilized pellets
4. Aeration
5. Temperature controller
6. Outlet (Effluent)

(Qiao, Zhang, Chen, & Chen)

Preparation of Synthetic Ammonia Wastewater

40 mg/L of Ammonia wastewater was prepared using the following steps:

1. Measure bucket volume using a 2 L beaker. Pour tap water 2 L of water into the beaker and then transfer it to the bucket. Repeat this step five times until a total volume of 10 L is obtained

   a. Note: use distilled water for batch culture method and do not add glucose to the synthetic ammonia wastewater

2. Mark the water level of 10 L in the bucket

3. Use the following table to calculate the appropriate weight for each substance:

Substance	Density (mg/L)	Weight (g)
NH_4Cl	153.2	1.532
$NaHCO_3$	468	4.68
$Na_2HPO_4 * 12 H_2O$	46.4	0.464
$NaCl$	20.5	0.205
KCl	9.6	0.096
$CaCl_2 * 2 H_2O$	9.6	0.096
$MgSO_4 * 7 H_2O$	33.6	0.336
$C_6H_{12}O_6$	25	0.25

Sample calculation for weight:

<p align="center">Equation 8 Calculation for Substance's Weight</p>

$$Weight = \frac{Density}{Volume} = 153.2 \frac{mg}{L} * 10\,L * 10^{-3} = 1.532\,g$$

4. Weigh each substance using the digital scale and pour it into the bucket

5. Mix all substances in the bucket with a stirring rod until they are completely dissolved

6. Reserve the mixture

Preparation of 1mol/L of HCl

1. In order to prepare 1 mol/L of HCL, the following calculation was done:

Equation 9 Calculation for 1 mol/L of HCL

$$\frac{250\ ml}{12 mol/L} = 20.8\ ml$$

2. Pour 20.8 ml of pure HCl into a flask and complete the rest with distilled H_2O so that the total volume of the mixture is 250 ml

3. Mix it well and close the flask

Preparation of H_3NO_3S

1. Calculate the mass required for a 0.8 % of H_3NO_3S:

Equation 10 Calculation for Mass of 0.8% of H_3NO_3S

$$Mass = 200\ ml * 0.08 = 1.6\ g$$

2. Clean the beaker with distilled water and fill it with 200 ml of distilled water

3. Pour it into a flask

4. Weigh 1.6 grams of H_3NO_3S, mix it with the water and pour it into the flask

5. Keep the mixture in the refrigerator

Preparation of Na_2 [Fe(CN)$_5$NO] * 2H$_2$O Reagent

1. Mix 0.1grams of $Na_2[Fe(CN)_5NO] * 2H_2O$ with 10 ml of distilled water in a test tube

2. Wait for $Na_2[Fe(CN)_5NO] * 2H_2O$ to completely dissolve in water before using it

3.4 Preparation of Batch Aeration – Method: Batch Culture

1. With the four prepared immobilized cells samples, measure the volume of the solid immobilized cells -> this was taken to be 30 ml in the graduated cylinder

2. Mix the immobilized cells with 230 ml of the prepared synthetic ammonia wastewater (except the addition of glucose) and pour it to the set up system

3. Dissolved oxygen (DO) used was 5.2 mg/L

4. Turn on the switch to start aeration

5. Wait for 1 hour before taking samples

6. Take samples at different time periods (preferably every 2-3 hours)

Collecting samples:

1. Label three test tubes for each immobilized cell so that we can analyze the concentrations of NH_4^+- N, NO_2^- - N and NO_3^- - N contained in each of them

2. Turn off aeration and let immobilized pellets settle down

3. Place a membrane paper on the syringe

4. Collect liquid sample and pass it three times through the membrane before pouring it to the test tubes

5. Collect 1 ml of liquid sample with the syringe and pour it to the test tube

6. Repeat steps 4 and 5 to all four immobilized cells

7. After collecting a total of 12 liquid samples in the test tubes, turn on the aeration

   Repeat this procedure in different time periods (40 min, 2 hours) to observe its behavior until most of the ammonia is removed from the solution

Preparing concentrations

NH4+- N Concentration

1. Add 1 ml of salicylic acid with pipette to all test tubes labeled with NH_4^+- N

2. Add 3 drops of 0.35% NaClO

3. Add 2 drops of $Na_2[Fe(CN)_5NO] * 2H_2O$. Note that $Na_2[Fe(CN)_5NO] * 2H_2O$ needs to be prepared daily

4. Complete the test tube with distilled water until it reaches the 50 ml mark

5. Wait for 1 hour for reaction to take place

NO2- - N Concentration

1. Add 1 ml of color indicator for NO_2^- - N with pipette to all test tubes labeled with NO_2^- - N

2. Complete the test tube with distilled water until it reaches the 50 ml mark

3. Wait for 20 min for reaction to take place

NO3- - N Concentration

1. Add 1 ml of the prepared 1 mol/L of HCL with pipette to all test tubes labeled with NO_3^- - N

2. Add 0.1 ml of the prepared 0.8% H_3NO_3S

3. Complete the test tube with distilled water until it reaches the 50 ml mark

Using UV Spectrophotometer

Use UV spectrophotometer to find absorptions for NH_4^+- N, NO_2^- - N and NO_3^- - N. For both NH_4^+- N and NO_2^- - N use the glass cuvette and for NO_3^- - N, use the quartz cuvette. Make sure to fill up to at least 2/3 of the cuvette and always have a blank sample with distilled water

for NH_4^+- N and NO_2^- - N or 1 mol/L HCL dissolved in 50 ml of distilled water for NO_3^- - N. For

NH_4^+- N set the wavelength to be 697 nm and for NO_2^- - N use 540 nm. Measure two different

wavelengths (220 nm and 275 nm) for NO_3^- - N. Record the absorbencies acquired for all

samples and repeat the same process of collecting samples from the reactors until most of the

ammonia is removed from the system. This will happen when the values for NH_4^+- N come very

close to zero, which corresponds to absorbencies of 0.005 or less.

Repeat the same procedure in order to test two different temperatures (27^oC and 32^oC) to

observe the effect of temperature in wastewater treatment using the batch culture method.

Analyzing Concentrations

With the absorbencies acquired from NH_4^+- N, NO_2^- - N and NO_3^- - N samples, use the

given standard curves for each nitrite and the equations of each specific curve to calculate

concentrations for all samples. The standard curves and equations below were used in order to

calculate concentrations for particular nitrites:
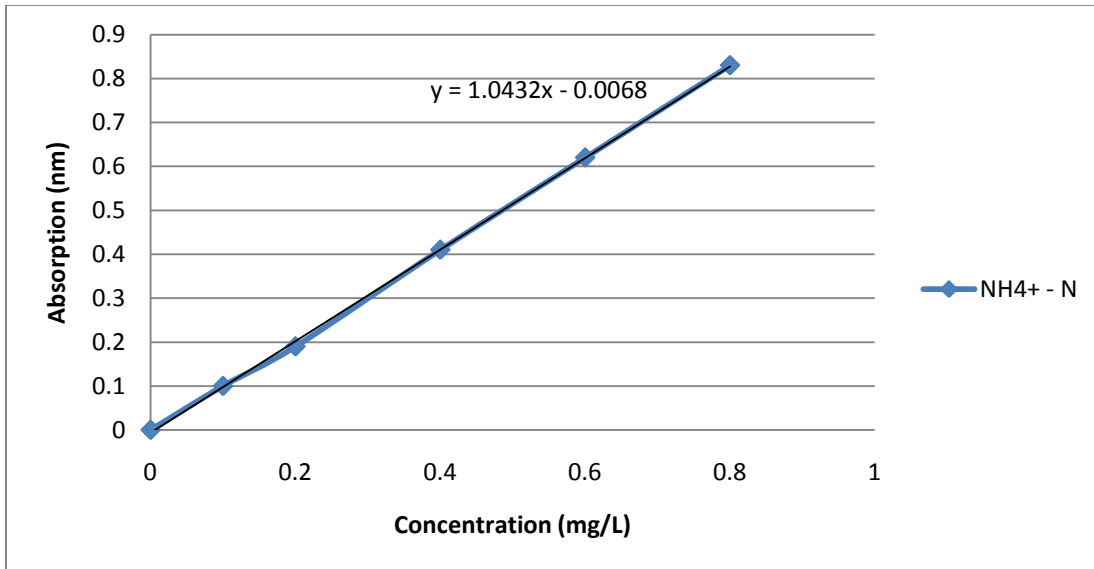
Figure 12 Standard Curve for NH$_4^+$ - N

In solving for concentration (x), with collected absorption values (y), this equation can be rewritten as:

Equation 11 NH$_4^+$ Concentration

$$x = \frac{y}{1.0432} + 0.0068$$
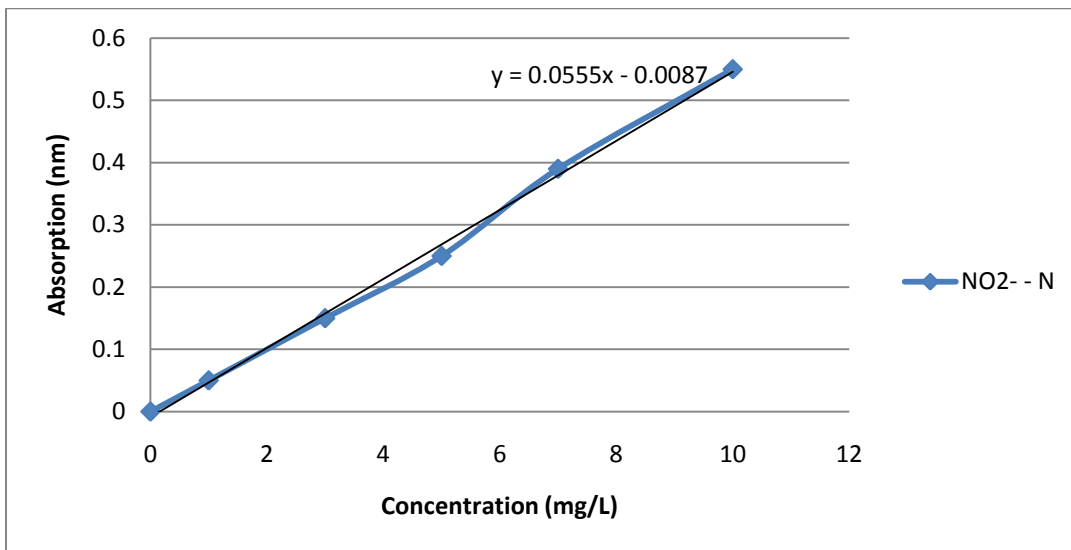
Figure 13 Standard Curve for NO$_2^-$ - N

Rewriting the equation in terms of concentration we have:

Equation 12 NO$_2^-$ Concentration

$$x = \frac{y}{0.0555} + 0.0087$$
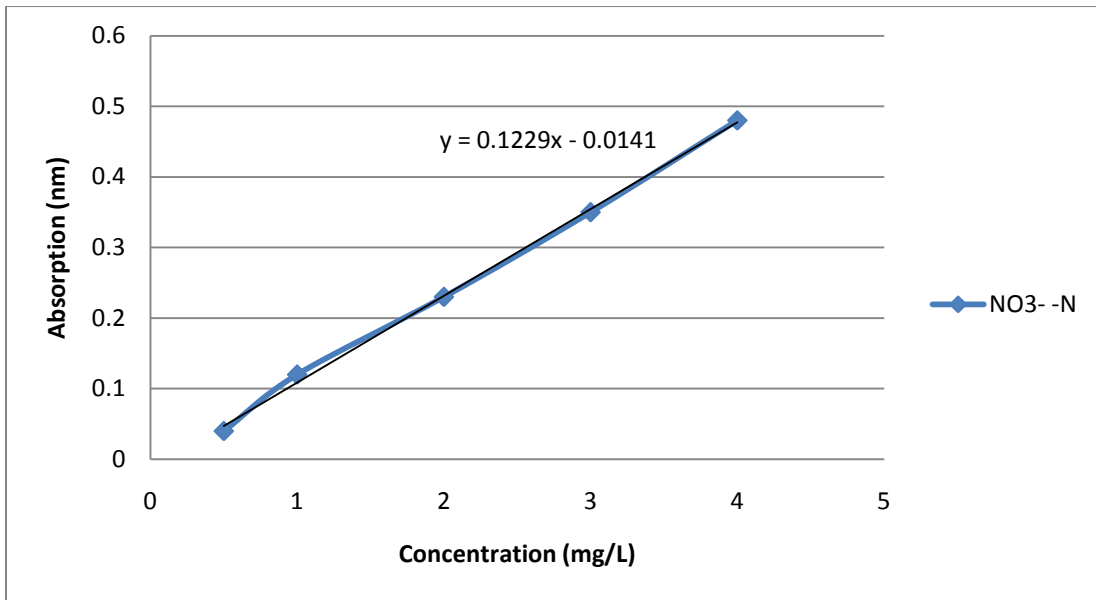
Figure 14 Standard Curve for NO$_3^-$ - N

Rewriting the equation in terms of concentration we have:

Equation 13 NO$_3^-$ Concentration

$$x = \frac{y}{0.1229} + 0.0141$$

With the values for concentration a plot was obtained of time and concentrations for each

nitrite to compare/contrast the behaviors of the four different types of immobilized activated

sludge at different temperatures. A graph showing the effect of temperature was also obtained in

order to notice any difference in conducting the same experiment at different temperatures.

3.5 Preparation of Continuous Aeration – Method: Continuous Mode

For continuous mode, a pump was added to the system so that it would continuously feed

the reactor with the synthetic ammonia wastewater. The same system set up for batch culture

was used, however, there were some modifications since the pump could only feed one reactor at

a time for continuous mode and for batch culture four reactors were used in the same time. In this method, the reactor containing the immobilized activated sludge pellets (sample 02) was chosen since it was more effective in treating the synthetic ammonia wastewater due to its relative high mechanical strength.

In this method, 40 mg/L of ammonia wastewater was prepared at a larger scale (10 liters) since wastewater needs to be constantly feeding the reactor. For continuous mode, glucose was added to the synthetic ammonia wastewater to observe the effect of an external carbon source in the treatment. Note that the bucket with the prepared ammonia wastewater can never be empty. The pump was set to work at 1.0 rpm and samples were collected daily instead of on an hourly basis due to the fact that concentration did not vary as much as in batch culture method. The same procedure for collecting samples, preparing concentrations, using the UV spectrophotometer and analyzing concentration were repeated until most of the ammonia was eliminated from the treated solution.

Based on the results from the batch culture method, I observed that I attain better results when the bioreactor is set at a higher temperature. Therefore, two temperatures ($30^{o}C$ and $32^{o}C$) were tested to detect any differences and the effect of temperature in wastewater treatment.

Hydraulic Retention Time (HRT)

The hydraulic retention time (HRT) is defined as the total time that a solution remains in a constructed bioreactor. It may also be considered as the time that is required for a whole wastewater treatment process to take place resulting in treated water that can be safely disposed to nature (Lenntech). I calculated HRT for the continuous mode method using the following relationship equation:

Equation 14 Hydraulic Retention Time (HRT) Calculation

$$HRT \text{ (min)} = \frac{Volume\ of\ the\ reactor\ (ml)}{\dfrac{Volume\ of\ the\ collected\ treated\ water(ml)}{Time\ (min)}}$$

The time in this equation refers to the time required to collect a specific amount of treated water. For this experiment, I collected 20 ml of the treated water in a graduated cylinder and this

corresponded to 7 minutes of collection. With all the values known, I was able to calculate the HRT for continuous mode using the above equation:

$$HRT \text{ (min)} = \frac{250 \; ml}{\frac{20ml}{7min}}$$

$$HRT = 87.5 \min \cong 1.46 \; hours$$

Total Organic Compound (TOC) and Total Nitrogen (TN)

Total organic compound (TOC) and total nitrogen (TN) were analyzed in the continuous mode method since there was an addition of glucose to the prepared synthetic wastewater. TOC was measured and used as an indicator of water cleanliness and the treatment's efficiency in removing carbon from the synthetic wastewater fed to the system. TN was analyzed in order to check if denitrification would take place in this experiment environment.

For TOC and TN analysis, 20 ml of the prepared synthetic wastewater and 20 ml of the treated wastewater collected from the outlet of the bioreactor were collected into two test tubes and taken to analysis in the laboratory.

4.0 Results and Discussion

In this section of the report, the information gathered in the experimental process is further analyzed and discussed. The results and discussion are broken down into two sections concerning the two conducted experiments using activated sludge to treat synthetic ammonia wastewater. The first section refers to the process using the batch culture method and the second section analyses the outcomes of the same process using the continuous mode method. Temperature effect was also observed for both methods, since it reveled to be a key component in the effectiveness of treating the synthetic ammonia wastewater. Furthermore, other considerations such as the effect of carbon in the synthetic ammonia wastewater preparation, and the total organic carbon (TOC) in the water after the aeration treatment were examined. All of the raw data and graphs collected in the experiment can be found in the appendix section.

4.1 Batch Culture

The batch culture method treats a fixed amount of wastewater fed to the system and gives a fast response to the treatment. Results vary very drastically within few hours of collection of each sample and in less than 24 hours all samples had reduced their NH_4^+- N concentration to nearly total. The hydraulic retention time (HRT) for a batch culture varies from 5 to 8 hours. When the experiment was conducted at $27\,^{o}C$, sample 02, sample 05, sample 06 and sample 04 had an ammonia reduction of 84%, 89%, 95% and 98% respectively. The average percentage of NH_4^+- N concentration reduction for these four samples was 91.5%, which is an excellent amount of ammonia removal from the water. This shows that the method used was relatively effective in treating the ammonia in the wastewater, therefore disposing water with almost fully

eliminated ammonia. Figure 16 below shows the behavior of ammonia reduction for batch culture method operated at $27\,^{\circ}$C over a period of one day.
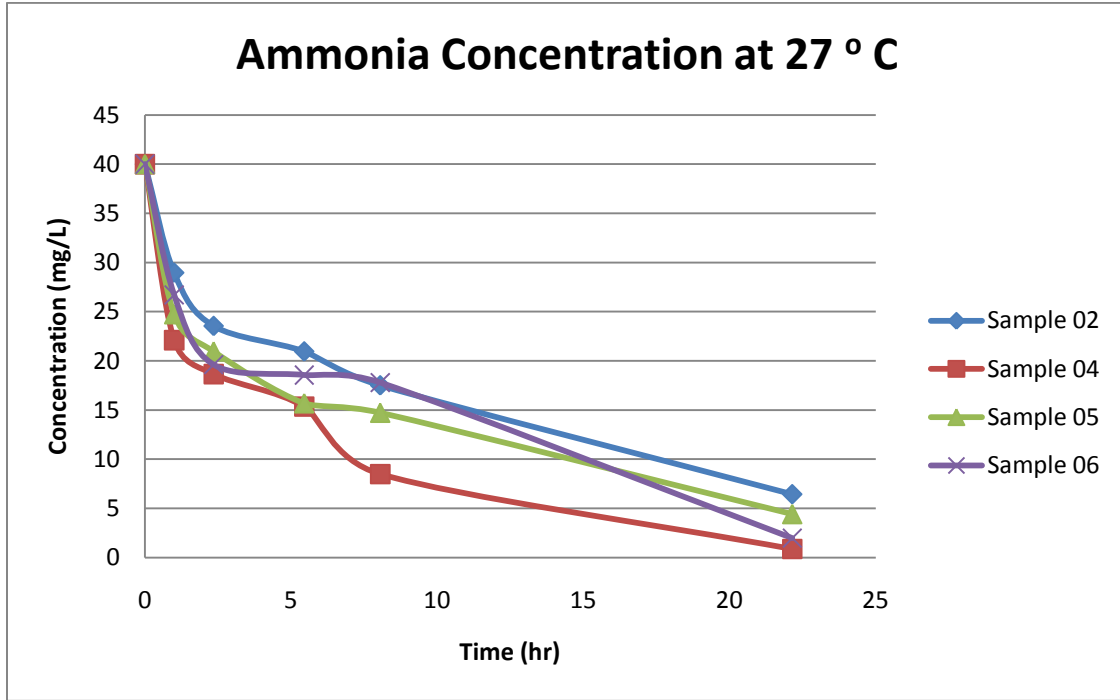
Figure 16 Batch Culture - Ammonia Concentration at 27 C

I conducted the same experiment at a higher temperature to compare and contrast the results and to investigate if modifications in temperature had an effect on nitrification performance. Figure 17 below shows the behavior of ammonia reduction for batch culture method operated at $32\,^{\circ}$C over a period of one day. Comparing with the results at a lower temperature, I observed that there was a greater decrease in ammonia concentration over the same period of time. At $32\,^{\circ}$C sample 05 had an ammonia concentration reduction of 97.5%, while sample 06, sample 02 and sample 04 had a reduction of 98.8 %. The average percentage of NH_4^+- N concentration reduction for these four samples was 98.5%, which is very close to a complete removal of ammonia.

All samples of immobilized activated sludge pellets had better results when the temperature was increased to $32\,^{\circ}$C. The experiment conducted at a lower temperature showed a difference of 8.5% of the ideal percentage of ammonia removal, however, when the temperature was raised to $32\,^{\circ}$C, the difference was only of 1.5% of the ideal percentage. Figure 18 shows the effect of temperature in sample 02. From this graph, one can observe that there is a significant difference in ammonia removal when the temperature is raised.
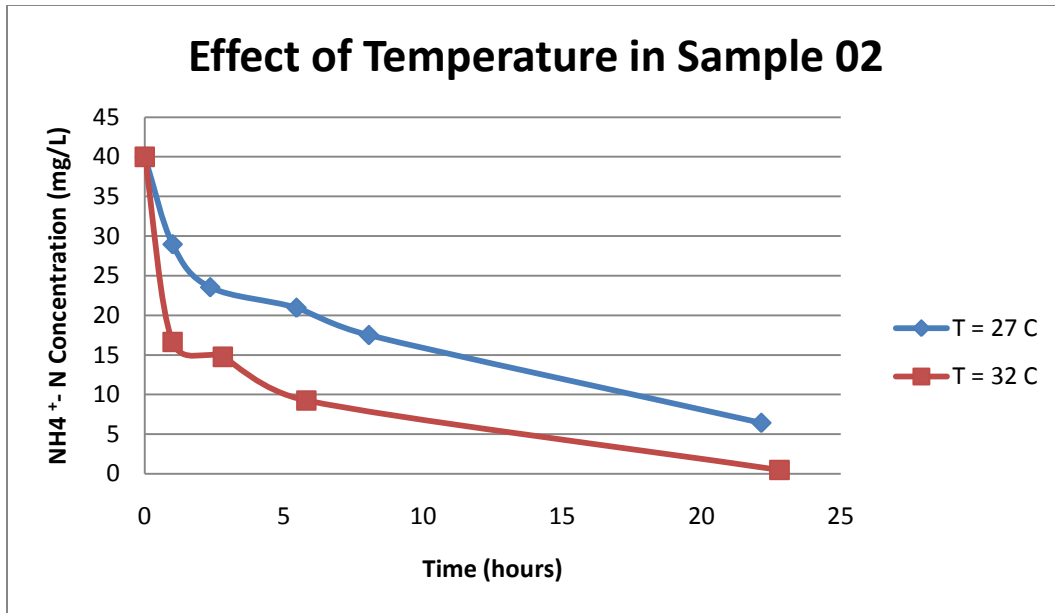
Comparing the four samples of immobilized activated sludge pellets, sample 02 was had a relatively high mechanical strength than the other samples and better outcomes in both temperatures tested. Table 6 below is a condensed table just focusing only on the results of the nitrification performance of immobilized cells acquired from sample 02 at 27 $^{\circ}$C and 32 $^{\circ}$C.

Table 6 Nitrification Performances of Sample 02

Sample 02 at 27° C	All concentrations are in mg/L		
Time (hours)	*NH₄ concentration*	*NO₂ concentration*	*NO₃ concentration*
0	40		
1	28.95	3.16	20.23
2.35	23.54	18.62	10.06
5.45	20.95	29.53	5.59
8.05	17.5	37.7	5.18
22.15	6.43	25.89	57.26

Sample 02 at 32° C **Time (hours)**	*NH₄ concentration*	*NO₂ concentration*	*NO₃ concentration*
0	40		
1	16.64	12.25	0.705
2.8	14.77	28.62	25.52
5.8	9.25	31.34	37.73
22.8	0.483	5.89	58.06

4.2 Continuous Mode

The continuous mode method treats a large amount of wastewater, since water is being constantly fed to the system, however, this process requires a longer time (sometimes it may take several weeks) to treat all the ammonia in the wastewater. The nitrites responded very slowly to this method and samples were taken in a daily basis instead of in an hourly basis as for batch culture. Based on the results obtained using the batch culture method, I also varied temperature to observe nitrification performance and I chose a higher temperature to start the experiment. Unlike the batch culture method, in continuous mode ammonia reduction was very slow. When the temperature was set at 30°C, it took more than a week for the NH_4^+- N absorption to reduce until it was almost 0.005 nm. Due to the limitation of time for the experiment, I stopped collecting samples at 30°C when NH_4^+- N absorption was about 0.1 nm. The same slow response

was obtained when the temperature was set to 32°C and nearly two weeks were needed for the

NH$_4^+$- N absorption to lower, however, ammonia was not completely removed from the water.

Figure 19 is a comparison between the results collected at 30°C and at 32°C.
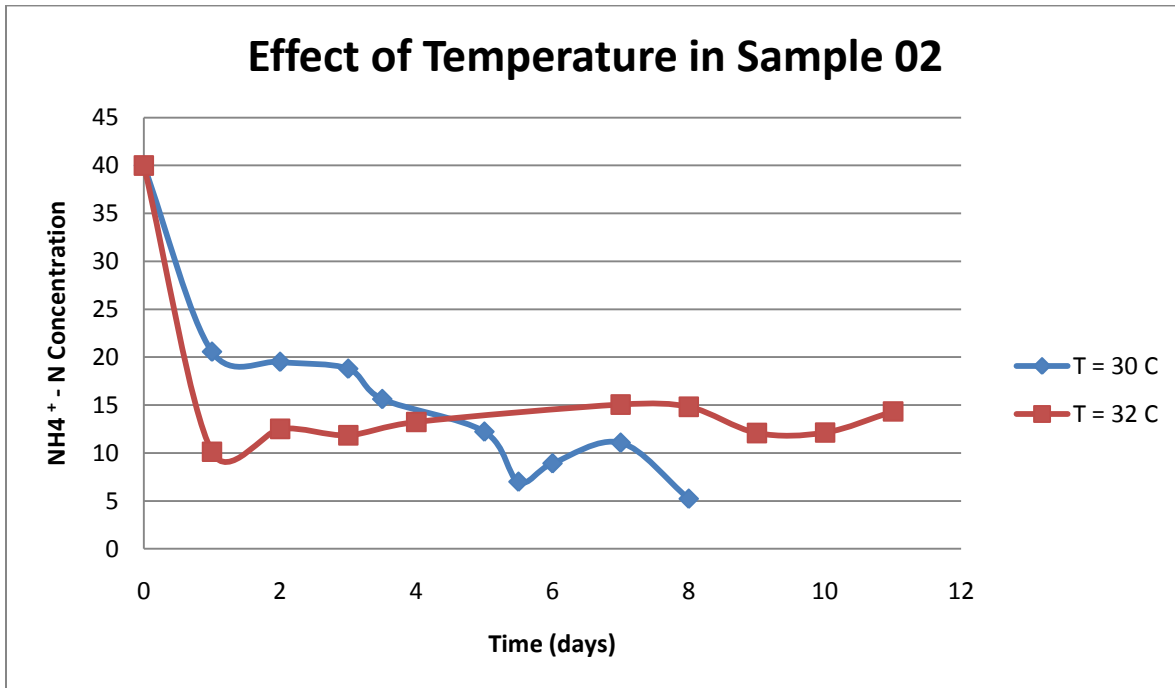
Figure 19 Continuous Mode - Effect of Temperature in Sample 02

As it is seen from Figure 19, ammonia reduction did not follow a trend such as in batch culture. In continuous mode method, ammonia concentration increased and decreased over the period of days. This fluctuation in concentration is expected since this is a long term method and oscillation is predictable until concentration stabilizes and decreases. Unfortunately, stabilization or acclimation was not fully achieved even after 8 days of experiment and unexpectedly ammonia removal was more significant at a lower temperature (30°C) than at a higher temperature (32°C). Even though stabilization was not fully achieved, Figure 19 points out that at 30°C ammonia concentration was perhaps beginning to stabilize, but more time was needed to

verify this hypothesis. For acclimation or stabilization to occur, the activated sludge pellets should be adjusted or adapted to changes in the environment such as the variation of temperature. In this particular experiment, the absorptions for NO_2^- - N, NO_3^- - N and specially NH_4^+- N need to come to a value of 0.005 or less.

The efficiency of ammonia removal from the fed wastewater cannot be proven from these two temperatures, since more time was needed in order to observe the behavior of the immobilized activated sludge pellets given these conditions. The variation of ambient temperature of $\pm 3\ ^oC$ might also have influenced these results since the aeration used is an open system and therefore very sensitive to outside temperature.

4.3 Effect of a Carbon Compound

The synthetic ammonia wastewater prepared was different for batch culture and continuous mode methods. In continuous mode method, glucose was added as a carbon source to observe if carbon plays an important role in the wastewater treatment used in this experiment. In batch culture, there was no glucose in the prepared wastewater and possibly this was the cause for NO_3^- - N curves to increase drastically compared to NH_4^+- N and NO_2^- - N curves for both tested temperatures. This behavior can be seen in both Figures 20 and 21 where sample 06 concentrations at T= 27^oC and at T= 32^oC are shown.
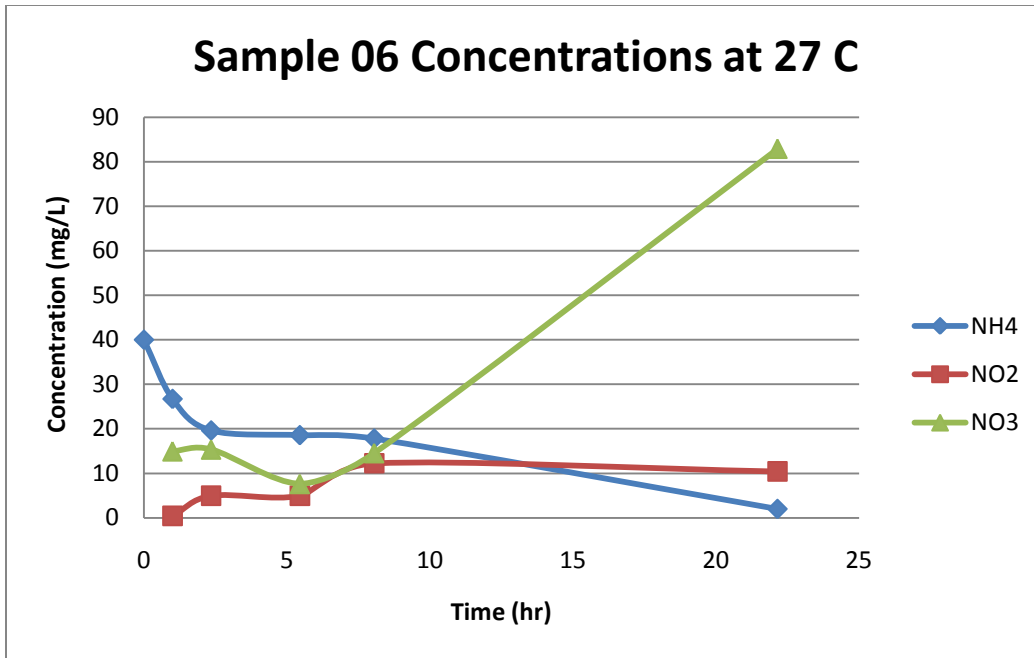
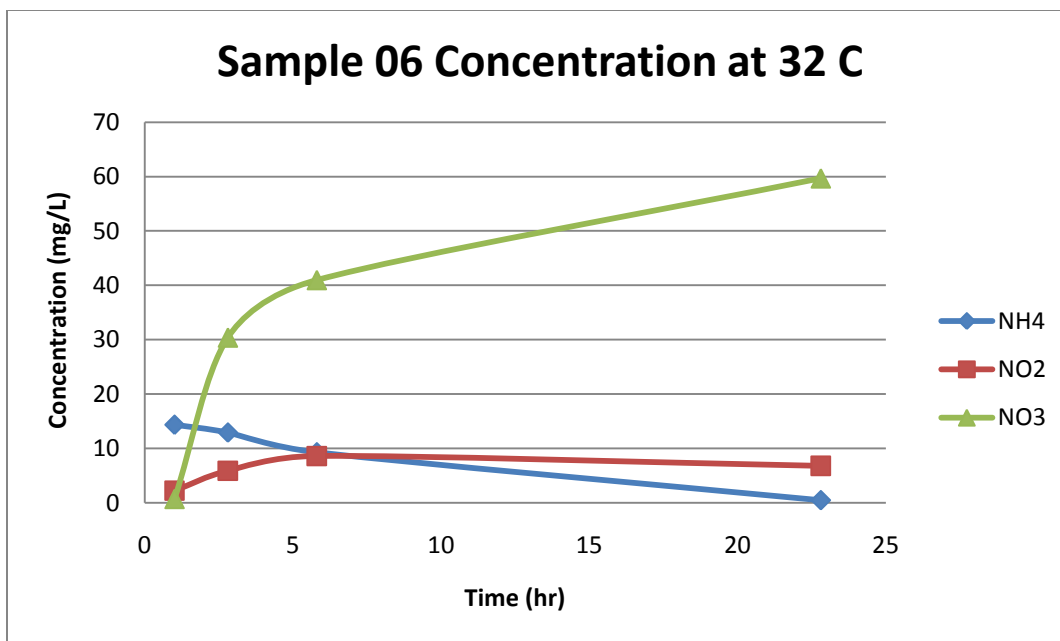**Figure 20 Batch Culture - Sample 06 Concentrations at 27 C**

Figure 21 Batch Culture - Sample 06 Concentration at 32 C

In order to make further conclusions about the effect of carbon in wastewater treatment and compare with the results of continuous mode, glucose needs to be added to the synthetic ammonia wastewater.

48

Glucose was added to the synthetic ammonia wastewater used in the continuous mode method total organic carbon (TOC) was analyzed before and after the treatment to verify the treatment's effectiveness in removing the carbon in the water. TOC was also measured to observe if the treatment used was responding successfully in ensuring a good water quality at the end of the process. Table 7 below corresponds to the data recorded before and after the wastewater was treated.

Table 7 TOC in Continuous Mode

	Synthetic Wastewater	Treated Wastewater
Total Organic Compound (TOC) (mg/L)	20.66	9.63

As it can be noticed from Table 7, the continuous mode method was relatively effective in removing carbon from the wastewater. The synthetic wastewater had a TOC of 20.66 mg/L while the treated wastewater had only 9.63 mg/L. The method used was able to remove nearly 50% of the carbon present in the wastewater supplied to the system. Some amount of TOC was lowered since biological oxygen demand (BOD) oxidizers grown in the beads consumed the glucose, resulting in a reduction of carbon concentration.

4.4 Total Nitrogen (TN)

Total nitrogen (TN) was also measured in addition to TOC in order to examine if denitrification took place in this experiment. Table 8 below corresponds to the data recorded before and after the wastewater was treated. As it is seen from this table, denitrification did not occur in this experiment. The synthetic wastewater had a TN of 50.20 mg/L and the treated wastewater had 51.28 mg/L, which is a very similar TN from the untreated wastewater. It was

expected denitrification not to take place in this experiment environment, since the immobilized

beads prepared had a porous structure. This porous structure provide an oxygen rich environment

for the immobilized activated sludge, where nitrification takes place but not denitrification. For

denitrification to take place, an oxygen poor environment is needed.

Table 8 TN in Continuous Mode

	Synthetic Wastewater	Treated Wastewater
Total Nitrogen (TN) (mg/L)	50.20	51.28

5.0 Conclusions

This section addresses the conclusions about the different methods used in treating ammonia wastewater with immobilized activated sludge and the effect of temperature in this process. This was accomplished through the collection of samples after a designated aeration period and the analysis for NH_4^+- N, NO_2^- - N and NO_3^- - N absorption through the UV spectrophotometer and concentrations calculations through the standard curves of each kind of nitrogen.

Samples collection varied for each method. In the case of the batch culture, samples were collected every 2-3 hours, since ammonia concentration decreased at a very fast pace in a short period of time. In batch culture, four different kinds of activated sludge were analyzed in order to determine which one would be selected to test for continuous mode method, since the pump in this method could only pump one sample at a time. Sample 02 was selected due to its relatively high mechanical strength. Samples in batch culture were analyzed on their performances at two different temperatures: 27^oC and 32^oC. In theory, wastewater treatment has better performances at higher temperatures. "An increased wastewater temperature has a positive influence on biological wastewater treatment methods, since an increase in temperature also increases the activity of microorganisms. On the other hand, any temperature increase results in a lower oxygen input capacity of the aeration system" (Kayser). Further analysis revealed that ammonia wastewater treatment responded better at higher temperatures than at lower temperatures, matching the theory.

Based on the results from batch culture, the initial temperature was raised to 30^oC instead of 27^oC and the higher temperature at 32^oC was kept. Since continuous mode is a long

term experiment and sample 02 did not have enough time to reach acclimation completely, no further conclusions could be done regarding the effect of temperature in the continuous mode method. Although the graph for temperature comparison in continuous mode suggests that at the end of the experiment, sample 02 had a greater reduction in ammonia concentration at 30°C, the level of ammonia concentration decrease at 32°C in the beginning of the experiment was much greater than at 30°C. While at 30°C ammonia reduction represented 50% of the original concentration, at 32°C ammonia reduction was of 75% of the original value in the starting point of the experiment.

In continuous mode method, glucose was added in the ammonia wastewater to observe for the effect of carbon in wastewater treatment by immobilized activated sludge. After the experiment, TOC was analyzed from the synthetic wastewater and the treated wastewater. TOC revealed that there was approximately 50% of organic compounds removal. The continuous mode method was relatively effective in removing portions of glucose from the synthetic wastewater. TN was also analyzed and the results showed that no denitrification took place since the immobilized beads prepared had a porous structure which provides an oxygen rich environment.

After much study and investigation on batch culture and continuous mode methods, and the effect on variation of temperatures, batch culture was more effective in treating ammonia wastewater by immobilized activated sludge sample 02. A generalized conclusion on the efficiency of each method tested cannot be stated since only sample 02 was analyzed in both methods. For the type of activated sludge with the same properties such as the ones from sample

02, batch culture worked better in the removal of ammonia concentration in wastewater than using continuous mode method.

6.0 Recommendations

Based on the analysis and comparison of batch culture and continuous mode methods, some recommendations were proposed on how to improve the experiment and draw further analysis on the effect of carbon and temperature on both methods used.

6.1 Evaluate TOC in Batch Culture

In order to observe the effect of carbon in the wastewater treatment using batch culture method and to compare the results with continuous mode method, I suggest that glucose should be added to the prepared synthetic ammonia wastewater. The addition of glucose will enable a TOC analysis and therefore a more detailed examination on a batch culture method's performance in treating organic compounds. In addition to this, a thorough determination on the levels of TOC comparison before and after the treatment using both methods could be made.

6.2 Further Continuous Mode Analysis

Due to time constraint, the continuous mode experiment had to be stopped for both temperatures tested before complete acclimation could be reached. The continuous mode method requires a longer time than batch culture method since it treats more wastewater and ammonia concentration removal response is slower when comparing to batch culture. If there was more time for the experiment, further conclusions could be drawn regarding the effect of temperature in continuous mode. Once acclimation is reached, it would be easier to determine which of the two temperatures tested had better outcomes in treating the synthetic ammonia wastewater fed to the system.

6.3 Test All Samples of Activated Sludge Pellets in Both Methods

After the experiment, the results showed that batch culture method was more effective in treating ammonia wastewater by immobilized activated sludge. Since time was limited and the pump used in continuous mode could only pump one kind of activated sludge sample, only sample 02 was tested in both batch culture and continuous mode methods. Therefore, from the results obtained from sample 02, I observed that batch culture method brought better outcomes in treating the wastewater than continuous mode method for this kind of activated sludge. In order to compare and observe more closely the efficiency of both methods, I would test all the four kinds of activated sludge pellets using both methods. This would enable me to make further comparisons and contrasts about the methods used and how these four activated sludge pellets behaves differently in both methods.

7.0 References

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8.0 Appendix

This section contains all the raw data and graphs for batch culture and continuous mode methods tested at 27°C and 32°C for batch culture, analyzed at 30°C and 32°C for continuous mode.

8.1 Appendix A – Batch Culture Data

8.1.1 Batch Culture at 27°C

Table 9 Batch Culture at 27 C Raw Data

Absorption & Concentrations

Samples	NH_4^+-N Absorption 697 nm	NH_4^+-N Concentration	NO_2^--N Absorption 540 nm	NO_2^--N Concentration	NO_3^--N Absorption 220nm	NO_3^--N Absorption 275nm	NO_3^--N Total	NO_3^--N Concentration
Wavelengths (nm)								
1 trial								
Sample 05	0.509	24.736	0.009	8.617	0.224	0.035	0.189	77.597
Sample 06	0.55	26.701	0	0.435	0.035	0	0.035	14.944
Sample 02	0.597	28.954	0.003	3.162	0.049	0.001	0.048	20.233
Sample 04	0.454	22.100	0.01	9.526	0.044	0	0.044	18.606
2 trial								
Sample 05	0.43	20.950	0.026	24.071	0.043	0	0.043	18.199
Sample 06	0.402	19.608	0.005	4.980	0.036	0	0.036	15.351
Sample 02	0.484	23.538	0.02	18.617	0.023	0	0.023	10.062
Sample 04	0.382	18.649	0.02	18.617	0.048	0	0.048	20.233
3 trial								
Sample 05	0.32	15.677	0.05	45.890	0.018	0	0.018	8.028
Sample 06	0.38	18.553	0.005	4.980	0.017	0	0.017	7.621
Sample 02	0.43	20.950	0.032	29.526	0.012	0	0.012	5.587
Sample 04	0.313	15.342	0.019	17.708	0.056	0	0.056	23.488
4 trial								
Sample 05	0.3	14.719	0.043	39.526	0.034	0.002	0.032	13.724
Sample 06	0.364	17.786	0.013	12.253	0.034	0	0.034	14.537
Sample 02	0.358	17.499	0.041	37.708	0.011	0	0.011	5.180
Sample 04	0.17	8.488	0.032	29.526	0.062	0	0.062	25.929
5 trial								
Sample 05	0.085	4.414	0.03	27.708	0.167	0.005	0.162	66.612
Sample 06	0.034	1.970	0.011	10.435	0.202	0	0.202	82.886
Sample 02	0.127	6.427	0.028	25.890	0.143	0.004	0.139	57.255
Sample 04	0.011	0.867	0.006	5.890	0.222	0	0.222	91.022

Table 10 Batch Culture at 27 C Concentrations

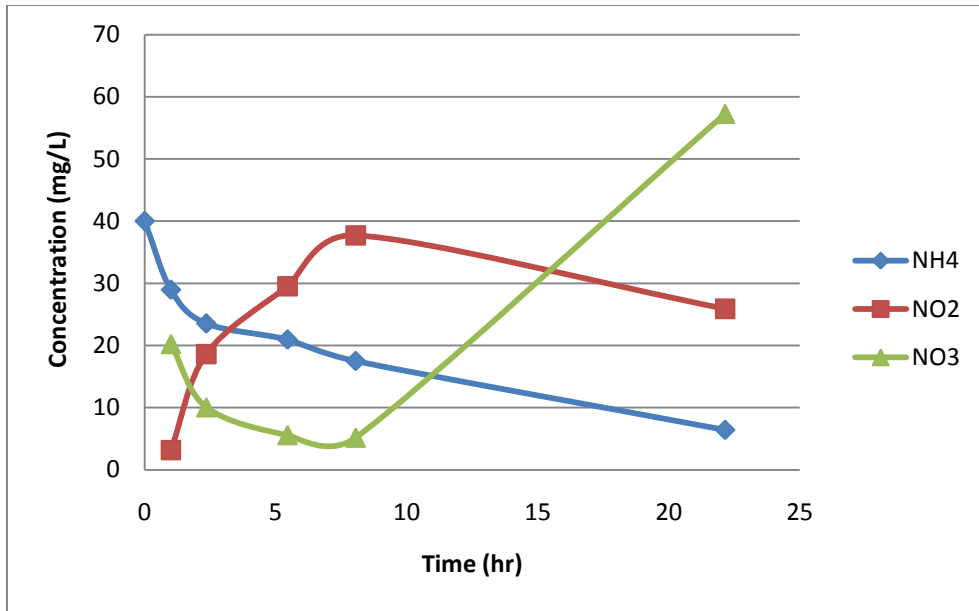| Time (hours)	Sample 02 NH_4^+ -N concentration	Sample 04 NH_4^+ -N concentration	Sample 05 NH_4^+ -N concentration	Sample 06 NH_4^+ -N concentration
0	40	40	40	40
1	28.954	22.100	24.736	26.701
2.35	23.538	18.649	20.950	19.608
5.45	20.950	15.342	15.677	18.553
8.05	17.499	8.488	14.719	17.786
22.15	6.427	0.867	4.414	1.970
	NO_2^- -N concentration	NO_2^- -N concentration	NO_2^- -N concentration	NO_2^- -N concentration
1	3.162	9.526	8.617	0.435
2.35	18.617	18.617	24.071	4.980
5.45	29.526	17.708	45.890	4.980
8.05	37.708	29.526	39.526	12.253
22.15	25.890	5.890	27.708	10.435
	NO_3^- -N concentration	NO_3^- -N concentration	NO_3^- -N concentration	NO_3^- -N concentration
1	20.233	18.606	77.597	14.944
2.35	10.062	20.233	18.199	15.351
5.45	5.587	23.488	8.028	7.621
8.05	5.180	25.929	13.724	14.537
22.15	57.255	91.022	66.612	82.886

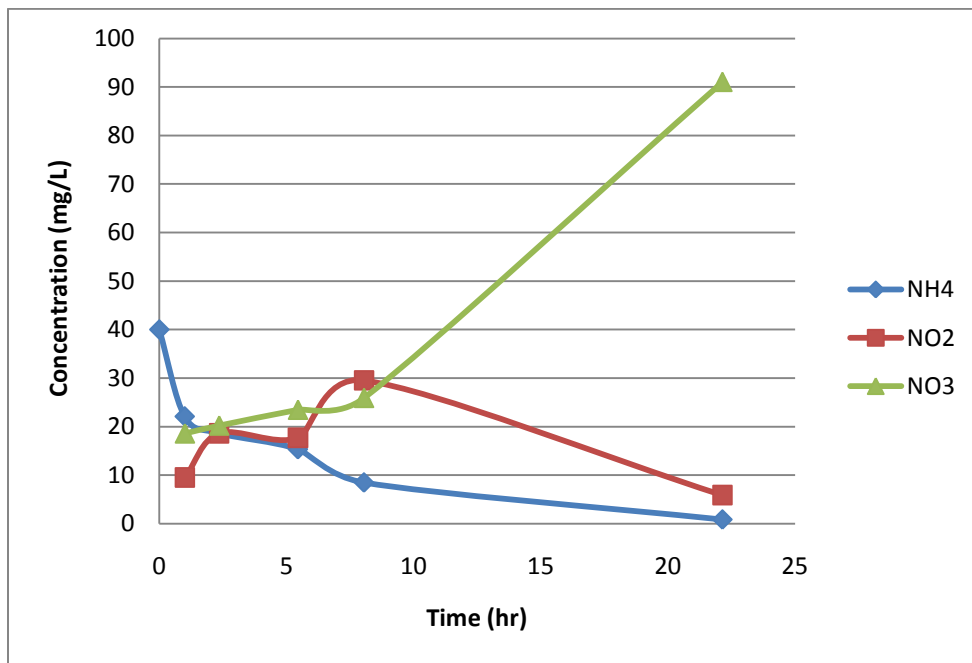Figure 22 Batch Culture - Sample 02 Concentration at 27 C

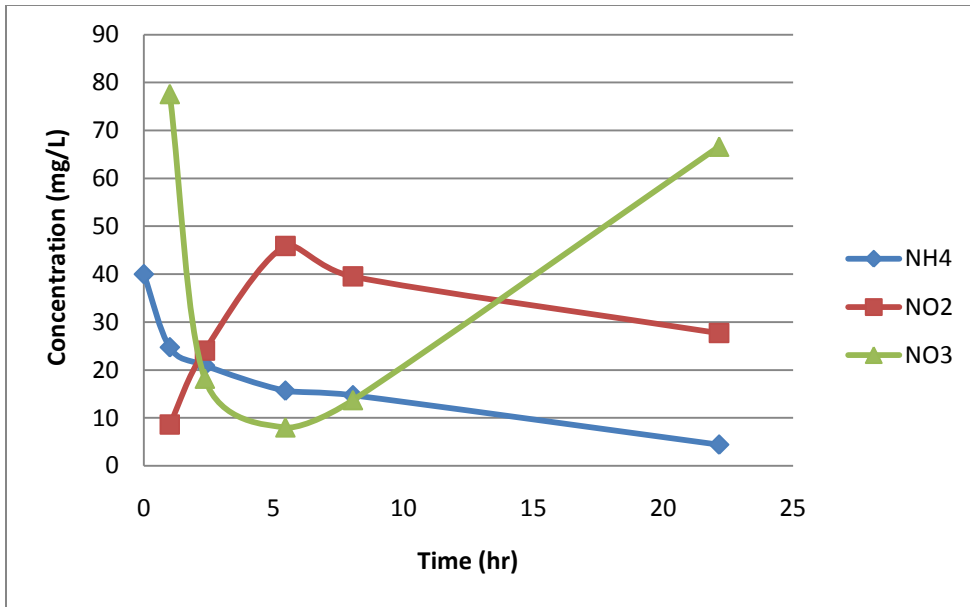Figure 23 Batch Culture - Sample 04 Concentration at 27 C

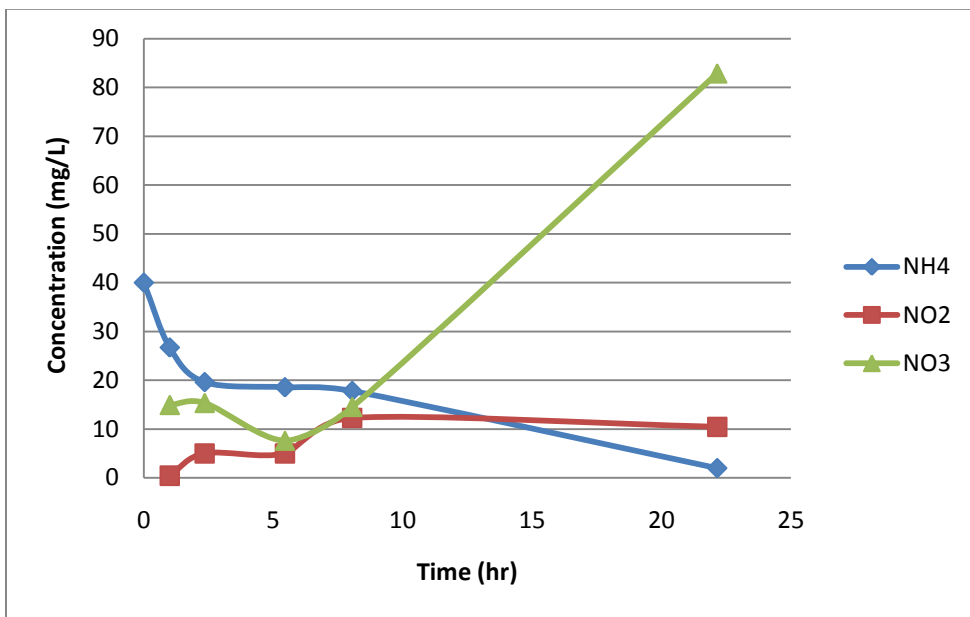Figure 24 Batch Culture - Sample 05 Concentrations at 27 C

Figure 25 Batch Culture - Sample 06 Concentrations at 27 C

8.1.2 Batch Culture at 32ºC

Table 11 Batch Culture at 32 C Raw data

Absorption & Concentrations

Samples	NH_4^+ -N Absorption	NH_4^+ -N Concentration	NO_2^- -N Absorption	NO_2^- -N Concentration	NO_3^- -N Absorption		NO_3^- -N Total	NO_3^- -N Concentration
Wavelengths (nm)	697 nm		540 nm		220nm	275nm		
1 trial								
Sample 05	0.327	16.013	0.008	7.708	0	0	0	0.705
Sample 06	0.293	14.383	0.002	2.253	0	0	0	0.705
Sample 02	0.34	16.636	0.013	12.253	0	0	0	0.705
Sample 04	0.22	10.884	0.013	12.253	0	0	0	0.705
2 trial								
Sample 05	0.291	14.287	0.039	35.890	0.061	0.001	0.06	25.115
Sample 06	0.263	12.945	0.006	5.890	0.074	0.001	0.073	30.404
Sample 02	0.301	14.767	0.031	28.617	0.061	0	0.061	25.522
Sample 04	0.186	9.255	0.03	27.708	0.06	0	0.06	25.115
3 trial								
Sample 05	0.213	10.549	0.047	43.162	0.09	0	0.09	37.320
Sample 06	0.187	9.303	0.009	8.617	0.101	0.002	0.099	40.982
Sample 02	0.186	9.255	0.034	31.344	0.094	0.003	0.091	37.727
Sample 04	0.095	4.893	0.038	34.980	0.132	0.004	0.128	52.780
4 trial								
Sample 05	0.014	1.011	0.033	30.435	0.134	0.001	0.133	54.814
Sample 06	0.003	0.484	0.007	6.799	0.145	0	0.145	59.696
Sample 02	0.003	0.484	0.006	5.890	0.141	0	0.141	58.069
Sample 04	0.003	0.484	0.009	8.617	0.134	0.001	0.133	54.814

Table 12 Batch Culture at 32 C Concentrations

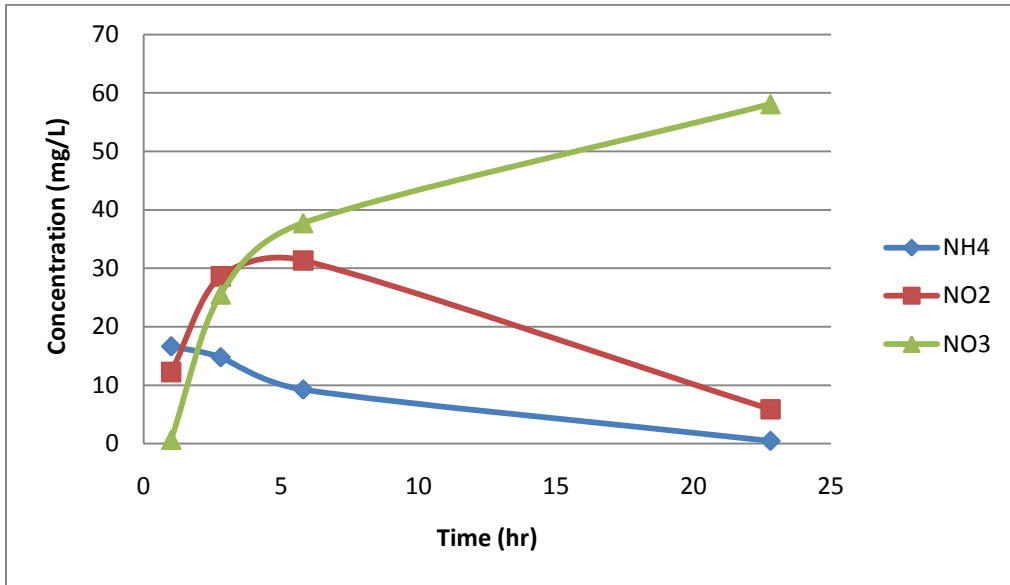| Time (hours)	Sample 05 NH_4^+ -N concentration	Sample 06 NH_4^+ -N concentration	Sample 02 NH_4^+ -N concentration	Sample 04 NH_4^+ -N concentration
0	40	40	40	40
1	16.013	14.383	16.636	10.884
2.8	14.287	12.945	14.767	9.255
5.8	10.549	9.303	9.255	4.893
22.8	1.011	0.484	0.484	0.484
	NO_2^- -N concentration	NO_2^- -N concentration	NO_2^- -N concentration	NO_2^- -N concentration
1	7.708	2.253	12.253	12.253
2.8	35.890	5.890	28.617	27.708
5.8	43.162	8.617	31.344	34.980
22.8	30.435	6.799	5.890	8.617
	NO_3^- -N concentration	NO_3^- -N concentration	NO_3^- -N concentration	NO_3^- -N concentration
1	0.705	0.705	0.705	0.705
2.8	25.115	30.404	25.522	25.115
5.8	37.320	40.982	37.727	52.780
22.8	54.814	59.696	58.069	54.814

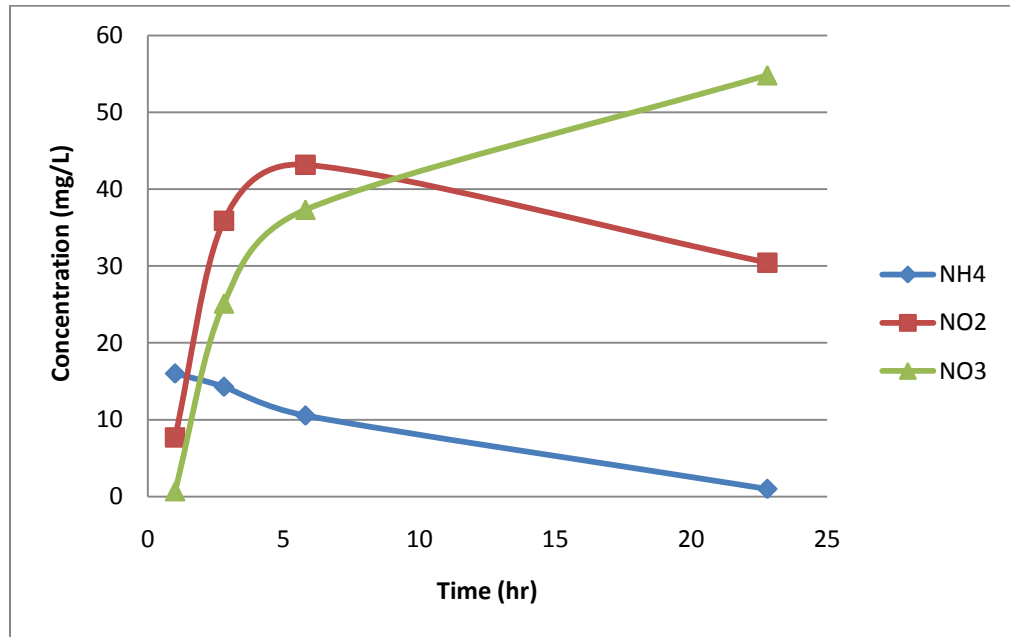Figure 26 Batch Culture - Sample 02 Concentrations at 32 C

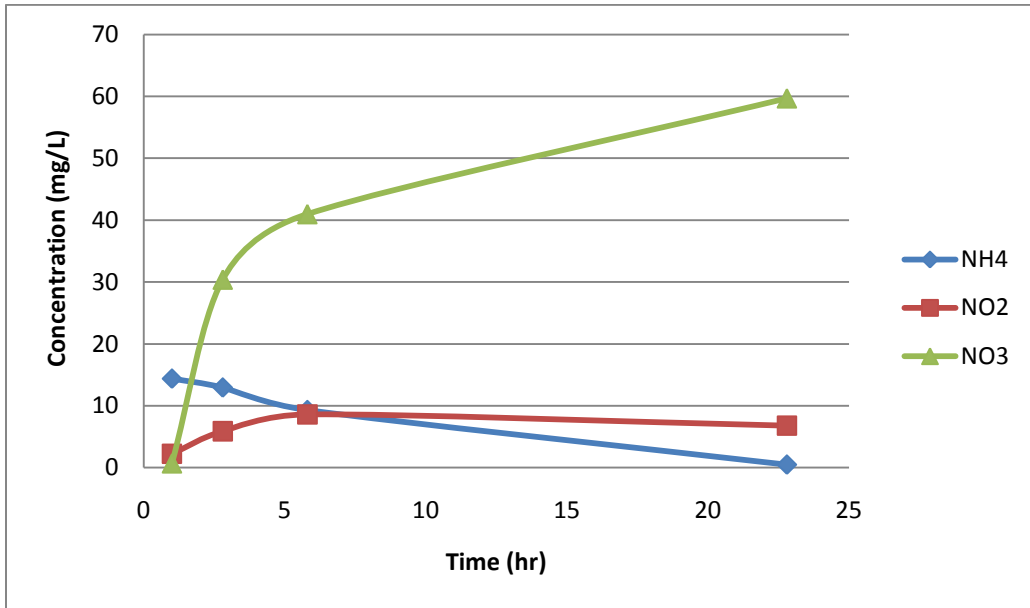Figure 27 Batch Culture - Sample 05 Concentrations at 32 C

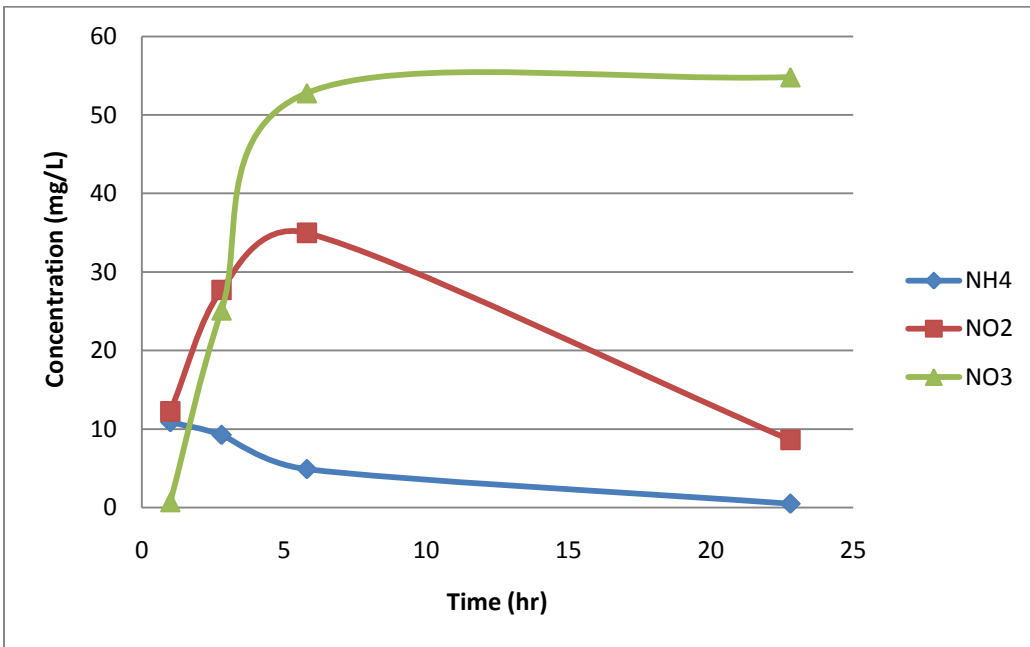**Figure 28 Batch Culture - Sample 06 Concentrations at 32 C**

Figure 29 Batch Culture - Sample 04 Concentrations at 32 C

8.2 Appendix B – Continuous Mode Data

8.2.1 Continuous Mode at 30ºC

Table 13 Continuous Mode Sample 02 at 30 C Raw Data

Absorption & Concentrations

| Time (days)	Samples	NH_4^+ -N Absorption	NH_4^+ -N Concentration	NO_2^- -N Absorption	NO_2^- -N Concentration	NO_3^- -N Absorption		NO_3^- -N Total	NO_3^- -N Concentration
Wavelengths (nm)		697 nm		540 nm		220nm	275nm		
0			40						
1	1	0.422	20.566	0.025	23.162	0.04	0.002	0.038	16.165
2	2	0.4	19.512	0.026	24.071	0.058	0.001	0.057	23.895
3	3	0.385	18.793	0.075	68.617	0.083	0.009	0.074	30.811
3.5	4	0.319	15.629	0.08	73.162	0.066	0.003	0.063	26.336
5	5	0.248	12.227	0.173	157.708	0.045	0.002	0.043	18.199
5.5	6	0.139	7.002	0.188	171.344	0.023	0.006	0.017	7.621
6	7	0.179	8.919	0.19	173.162	0.071	0.003	0.068	28.370
7	8	0.224	11.076	0.181	164.980	0.05	0.003	0.047	19.826
8	9	0.102	5.229	0.28	254.980	0.048	0	0.048	20.233
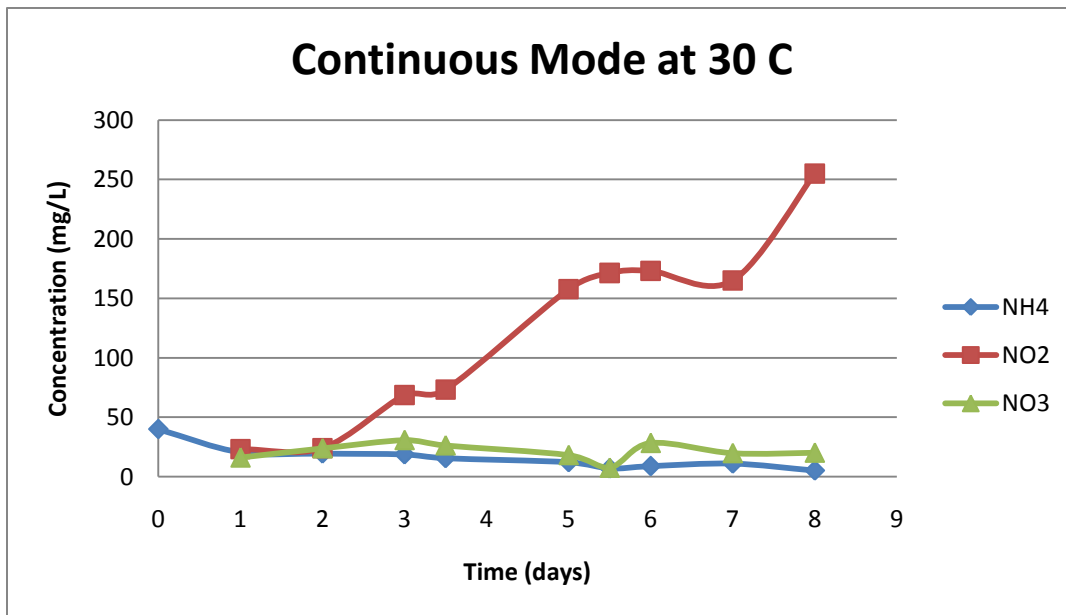
Figure 30 Continuous Mode - Sample 02 Concentrations at 30 C

8.2.2 Continuous Mode at 32°C

Table 14 Continuous Mode Sample 02 at 32 C Raw Data

Absorption & Concentrations

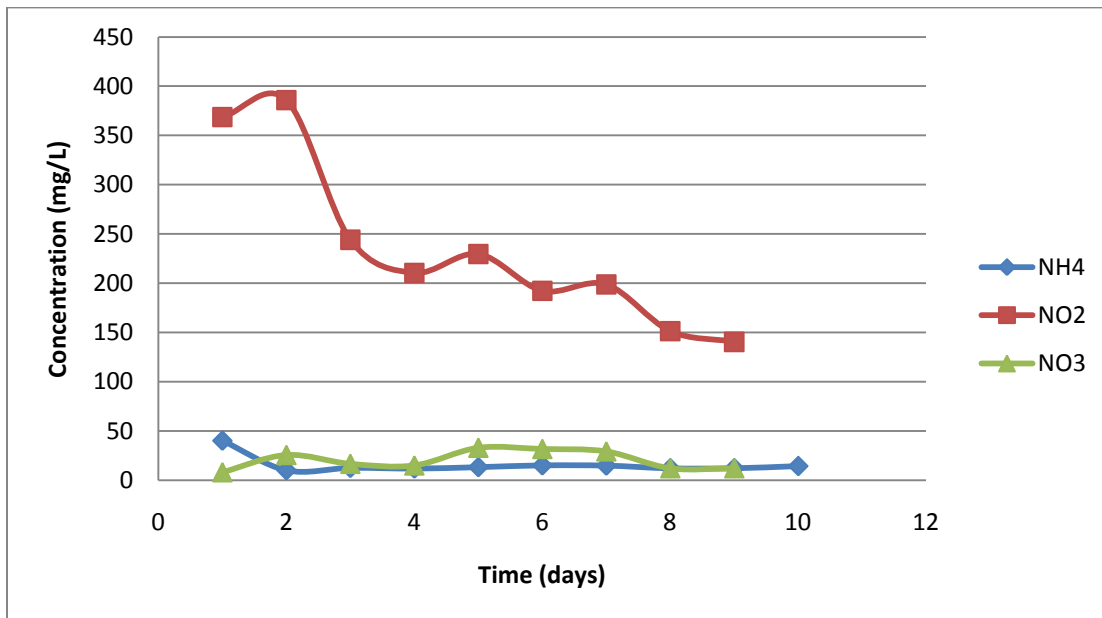| Trials	NH_4^+ -N Absorption 697 nm	NH_4^+ -N Concentration	NO_2^- -N Absorption 540 nm	NO_2^- -N Concentration	NO_3^- -N Absorption 220nm	275nm	NO_3^- -N Total	NO_3^- -N Concentration
Wavelengths (nm)		40						
1	0.204	10.118	0.405	368.617	0.018	0	0.018	8.028
2	0.254	12.514	0.424	385.890	0.062	0.001	0.061	25.522
3	0.24	11.843	0.268	244.071	0.042	0.003	0.039	16.572
4	0.269	13.233	0.231	210.435	0.038	0.003	0.035	14.944
5	0.307	15.054	0.252	229.526	0.086	0.007	0.079	32.845
6	0.302	14.815	0.211	192.253	0.077	0.001	0.076	31.624
7	0.245	12.083	0.218	198.617	0.071	0.001	0.07	29.183
8	0.246	12.131	0.166	151.344	0.029	0.001	0.028	12.096
9	0.292	14.335	0.154	140.435	0.028	0	0.028	12.096

Figure 31 Continuous Mode - Sample 02 Concentrations at 32 C