

CENE 486C: Engineering Design Capstone

Nitrification Column

Final Design Report

Table of Contents

Table of Tables

Table of Figures

Table of Equations

Abbreviations

Acknowledgments

The Waste to Nutrients capstone team would like to express appreciation to Lecturer, AlarickReiboldt, for his continuous project support, instruction, and guidance. We could not have asked for a better project mentor.

We would also like to express gratitude to our Academic Advisor, Professor Baxter, for his insightful comments and encouragement, but also for his recommendations which expanded our research.

Our sincere thanks goes to CENE Lab Manager, Gerjen Slim, for his access to materials, the laboratory, and research facilities. Without this support it would not be possible to conduct this research.

1.0 Project Description

1.1 Project Understanding

Urine is an unsterile, nutrient-rich resource that can be used to fertilize plants. Human urine contains high quantities of nutrients such as nitrogen and phosphorous. Though urine is unsterile, it does not contain the same biological hazards that exist in solid waste [1]. Separating urine from solid waste is an effective approach for nutrient recovery. Exploiting the benefits of urine is a good way of 'closing the cycle' in nutrient flows. This system could reduce waste treatment costs, ecological consequences, and improve sanitation in developing areas. The ultimate purpose of this project is to recover nitrogen nutrients from liquid waste in a scale of a single-person use column. The main purpose of the nitrification column is to convert nitrogen compounds in urine (ammonia) to a form that can be used by plants directly. The nitrification column provides suitable growth and living conditions for the bacteria involved in the process of nitrification.

1.2 Project Process Description

Liquid waste rapidly converts to the ammonia species which must be oxidized to nitrate before plants can take it up through their root systems. Nitrogen compounds are reduced and are sequentially oxidized to nitrite and nitrate by nitrifying bacteria through the nitrification process. The following figure illustrates the process of nitrification within this column.

Diluted urine enters the top of the column, saturating the growth media packing. This media allows for the growth of two types of bacteria; nitrosomonaseutropha and nitrobacterwinogradski[2]. This diluted urine rapidly converts from urea to ammonia, the predominate form of nitrogen in human urine. Nitrosomonas oxidizes the ammonia to nitrite, an intermediate step within the column. This is described by the following equation [3,4].

Equation 1: Step 1

 $NH_3 + O_2 \rightarrow NO_2 + 3H^+ + 2e^-$

Nitrobacter oxidizes nitrite to our desired final product nitrate. This process is described by the following equation [3,4].

Equation 2: Step 2

Figure 1: Nitrification Column Illustration

$NO₂ + H₂O \rightarrow NO₃ + 2H⁺ + 2e⁻$

Detailed images of these bacteria are described in the appendix, *8.1 Nitrifying Bacteria Images*.

1.2 Project Tasks and Subtasks

It is critical to identify all tasks required to complete the project. This section describes the complete list of tasks preformed. For more information, refer to the nitrification project proposal.

1.0 Legality and Sanitation Aspects

1.1 **State Local and Federal Regulations**

This included detailed research into the regulations that influence all project elements.

1.2 **Effluent Quality Regulations**

This included detailed research into the regulations in regard to the use of this effluent in house-scale gardens.

1.3 **Hazards to Public Safety**

An analysis of the potential consequences that could possibly effect the public.

2.0 Preliminary Lab Work

2.1 **List of Standard Methods**

A compilation of all standard methods required for the lab sections of this report. Effectively creating laboratory procedures.

2.2 **Standard Method Access**

Acquiring the above list of standard methods.

2.3 **Lab Access**

Acquiring laboratory use approval, keys, and work stations.

2.4 **Material and Equipment Accesses**

Acquiring all required materials and equipment. Items not provided by the laboratory were ordered through lab manager with technical advisor approval.

3.0 Initial Lab Work Considerations

3.1 **Urine Samples**

An analysis of urine alternatives and collection methods. Comparing natural urine to surrogate samples.

3.2 **Urine Dilution**

Determining the proper urine dilution for effective nitrification.

3.3 **Bacteria Acquirement**

Determining what bacteria was needed and the most effective means of obtaining these bacteria species.

3.4 **Batch Sample Establishment**

Creating batch bacteria samples that can be directly applied to nitrification column design. Determining which bacteria alternative is optimal for final design selection.

3.5 **Growth Media Selections**

Acquiring potential growth media alternatives. Lab testing of these growth media options. Determining which media alternative is optimal for final design selection.

4.0 Column Cartridge Lab Testing

4.1 **Cartridge Testing Method**

Creating a column cartridge methodology.

4.2 **Cartridge Data Collection**

Completing multiple laboratory trials following the previously created methodology.

4.3 **Cartridge Analysis**

An analysis data collected from above lab testing.

4.4 **Cartridge Conclusion**

Concluding this task. Determining conclusions on hydraulic retention time, oxygen demand, column length, inflow rate, and cartridge efficiency.

5.0 Final Design Considerations

5.1 **Design Concept Generation**

Determining final design alternatives.

5.2 **Design Concept Analysis**

Comparing and analyzing the possible design alternatives.

5.3 **Design Concept Selection**

5.3.1 **Determine Materials**

6.0 Final Design Calculations

6.1 **Determine Desired Efficiency**

This included determining the greatest possible system efficiency from selected design concept.

6.2 **Determine Column Dimensions**

Considering the desired efficiency and final design concept. Analyzing this information to determine optimal column design dimensions.

6.3 **Request Last Minute Feedback**

7.0 Constructions of Final Design

8.0 Final Column Design Testing

8.1 **Column Testing Method**

Creating a final column test methodology.

8.2 **Column Data Collection**

Completing multiple laboratory trials following the previously created methodology.

8.3 **Column Analysis**

An analysis data collected from above lab testing.

- 8.4 **Column Conclusion**
- 8.5 Concluding this task. Determining conclusions on system efficiency.

9.0 Project Management

9.1 **TA Meetings**

Minimum of 4 meeting through project entirety.

9.2 **Client Meetings**

Professional meeting updates every two weeks.

9.3 **Team Meetings**

Professional meeting updates every week.

9.4 **Status Presentations** Professional presentation updates every three weeks

2.0 Technical Section: Getting Started

2.1 Legality and Sanitation Aspects

This technology is relatively new, thus no state, local, or federal regulations of effluent quality exist. However, there are fertilizer restrictions that must be considered in the project process design. This table describes the fertilizer regulations found in the Code of Federal Regulations [5].

Table 1: CFR Fertilizer Regulations

In addition to government regulations, public safety aspects must be considered. Ammonium nitrate fertilizer is a highly explosive compound that could possibly be created by the final column design in the form of organic nitrogen [6]. However, the final column design does not directly create ammonium nitrate fertilizer. This is illustrated in the following figure.

Table 2: Ammonium Vs. Ammonia

To create an explosion a detonator and a fuel source are required, two elements that do not exist within the final column design. Ammonium nitrate compound is stable under most conditions, and is not a direct source of concern.

2.2 Preliminary Lab Work

2.2.1 List of Standard Methods

All standard methods used to design the nitrification column were collected from the HACH website [7]. The following table describes the standard methods that were applied in the laboratory. All interferences are listed in the associated standard method. The scope and application for all of the following methods is water and wastewater.

Table 3: List of Standard Methods

It should be noted that additional standard methods would have been beneficial in testing elements within the nitrification column design. Unfortunately, due to time constraints these could not be applied to this project. These additional standard methods are described in the appendix, *8.2 Additional Standard Methods*.

Temperature and pH

The data collection probe was used to analyze temperature and pH. These elements are necessary to determine the temperature and pH ranges for the nitrifying bacteria while creating the batch samples. These ideal ranges will aid in the survival of the bacteria as well as providing the highest nitrogen removal efficiency.

Alkalinity

Measuring alkalinity was important to determine the urine's ability to neutralize acid inputs. It is critical for the design of the nitrification column to operate in the optimal pH range. Therefore, it was necessary to determine the buffer range that all the nitrifying bacteria can keep alive. It is also necessary to determine any possible pH fluctuations that can occur in the system while creating bacteria batch samples.

Oxygen Demand, Chemical

The chemical oxygen demand (COD) test was used to indirectly measure the amount of organic compounds in the urine sample. The mg/L of COD results are defined as mg of O2 consumed per liter of sample. COD commonly indicates the amount of oxygen required to oxidize soluble and particulate organic matter in the water. COD was an important water quality parameter because it provided an index to assess the effect discharged wastewater will have on the receiving environment. Higher COD levels mean a greater amount of organic material in the sample, which will reduce dissolved oxygen (DO) levels and could possibly have negative effects in our system and the environment. For nitrification column project, it is a necessary measurement to evaluate the nitrifying properties of the bacteria batch samples.

Oxygen, Dissolved

Dissolved oxygen refers to the level of free, non-compound oxygen present in water or other liquids. For this project, it is an important parameter in assessing the urine quality because of its influence on the microorganisms in the system. The nitrification process is an aerobic process, both the ammonia oxidation and nitrite oxidation process are in need of oxygen. Therefore, it's necessary to find how much dissolved oxygen is available from the urine and is there any aeration needed to keep the nitrifying bacteria alive.

Nitrate

As we have discussed previously in this document, nitrate makes an excellent fertilizer for plants and agriculture. The purpose of this project is to exploit the benefits of this nutrient as a fertilizer, so the ability to collected nitrate data is critical for the success of this project.

Nitrite

Nitrite is an intermediate effluent that exists for this project. This effluent is oxidized in our sample to our final product, Nitrate. The ability to analyze this intermediate in the system is a critical aspect to the success of the column with regard to concentration and efficiency of our final design.

Nitrogen, Ammonia

The measurement of ammonia nitrogen is the system is important to determine the influent concentrations in the system. Ammonia is realized from the breakdown of urea. Ammonia is oxidized to nitrite, which is then oxidized to nitrate. The ability to analyze this influent in the system is a critical aspect to the success of the column with regard to concentration and efficiency of our final design.

Nitrogen, Total

There are three forms of nitrogen that are measured in this system: ammonia, nitrites, and nitrates. Total nitrogen is the sum of total nitrogen (ammonia, organic and reduced nitrogen) and nitrate-nitrite. Because the design requires the application of nitrification, it was important to determine the sum of the nitrogen in the system. This allowed for determining any total losses within the system.

2.2.2 Lab, Materials, and Equipment Access

The laboratory used for this design is the environmental engineering lab located in the engineering building. The nitrification column and cartridges were constructed tested and stored in this laboratory. This laboratory gave us access to all laboratory equipment required, and most materials. The following list describes the materials that were ordered by the lab manager and approved by the technical advisor.

- Nitrate TNTplus Vial Test, LR (0.2-13.5 mg/L NO3-N)
- Nitrite TNTplus Vial Test, LR (0.015-0.600 mg/L NO2--N)
- Ammonia TNTplus Vial Test, LR (1-12 mg/L NH3-N)

2.3Initial Lab Work Considerations

2.3.1 Urine Samples

It was determined that surrogate samples would have been ideal for this project because they would have a consistent amount of ammonia entering the system for every test, giving more accurate test results. However, due to time constrains and limited resources, it was determined that using natural human urine samples would be more feasible. These urine samples did not contain any antibiotics that could potentially interfere with nitrifying bacteria operations.

2.3.2 Urine Dilution

Nitrifying bacteria work optimally when the concentration of ammonia is below 15 mg/L The average ammonia concentration is 7 mg/L in human urine. Thus, no dilution is required for the physical process of nitrification. The average ammonia concentration in the tested urine samples was determined to be 7.5 mg/L. This is slightly higher than the average human concentration, but dilution is still unnecessary.

However, to keep a stable alkalinity of the system it requires the addition of tap water. The nitrifying bacteria decrease the alkalinity in the system to the point of ineffective operation if this tap water is not added. It was determined that the dilution should be 3-parts-tap-water to 1-part-urine to assure most effective nitrification.

2.3.3 Bacteria Acquirement

Bacteria species were collected through two means: Wildcat Hill Wastewater (WCH) treatment facility and API Quick Start. The pre-treated wastewater collected at the WCH facility contained a plethora of microorganisms that need to be removed. The API Quick Start was an inactivity nitrifying colony that

required activation and growth. The following figure provides and illustration of both types of nitrifying bacteria.

Figure 2:Nitrification Bacteria Image

2.3.4 Batch Sample Establishment

The most difficult project challenge experienced was the growth of a strong bacteria colony. Nitrifying bacteria are fragile organisms, small changes in the surrounding conditions can have a huge impact on the health of the bacterial colony. These bacteria have a small range of acceptable conditions for growth, and these must be maintained to create active nitrifying bacteria batches.

As mentioned above to create a batch bacteria colony for the WCH bacteria, it must be purified and all excess organisms must be exterminated and removed. This was done though initiating nitrification in the system. 500mL of urine was added to the WCH sample and the pH, alkalinity, temperature, ammonia concentration and chemical oxygen demand (COD) were used to monitory the sample. This allowed us to create ideal conditions for the nitrifying bacteria, killing most other microbes off. The data collected from this is described in the appendix, *8.3 WCH Nitrifying Bacteria Batch Monitoring*.

The API bacteria colony was much easier to establish activity. The API Quick-Start solution was added to 2500mL of water to increase sample volume. Every Monday, Wednesday, and Friday 50 mL of diluted urine was added to the batch system. 50 mL of batch sample was removed and those days as well to

prevent nitrate buildup in the container.

Two different nitrifying bacteria sample batches were created. After these colonies were established, 50 mL of diluted urine were added to each container ever Monday, Wednesday, and Friday. 50 mL of the batch sample was removed to avoid nitrate build up in the system. 500 mL of tap water were applied to the two batches daily to avoid evaporation.

Figure 3: Batch Bacteria Samples

These two batch bacteria sample alternatives were as pure as possible, and ready to use in the nitrification column testing.

2.3.5 Growth Media Alternatives

Three types of growth media alternatives were selected. This included pine shavings, cedar shavings, and dry straw. These are illustrated respectively in the following figure. As mentioned previously, this growth media allows for the furnish of the nitrification bacteria within the column. The ideal characteristics of this media include: high absorbability, low decomposability, and high surface area.

Figure 4: Growth Media Alternatives

2.3.6 Bacteria Batch and Growth Media Selections

The bacteria and growth media design components were selected using the media beaker lab test analyzing the optimal characteristics of each system component. The methodology of this beaker lab test was developed. Six 600 mL beakers were collected and filled with bacteria and growth media detailed in the following table.

Table 4: Beaker List Bacteria Batches

250 mL of tap water was then added to each beaker. The beaker tests were then allowed to saturate for a 24-hour period. 25mL of non-diluted were added to each beaker and the initial ammonia, nitrite, nitrate, and COD were tested. After another 24-hour period these values were tested again in the pine and cedar (the straw was thrown out due to excessive decomposition). Observations were recorded throughout the entirety of this process. The following figure illustrates the beaker test.

Figure 5: Batch Bacteria Beaker Test

The data collected from this lab procedure is described in the appendix, *8.4 Beaker Test Raw Data*. Though the Beaker 1 with eth WCH bacteria displayed the most effective nitrification, we selected the API bacteria to use in the final design because it has less interference from residual bacteria. The dry straw was not considered because of its foul odor and rapid decomposition. Pine was selected over cedar because it demonstrated the highest adorability and lowest decomposition. It should be noted that cedar dyed the effluent a reddish color, this was undesirable.

3.0 Technical Section: Column Cartridge Lab Testing

3.1 Column Cartridge Test Set-up

Figure 6: Column Cartridge Lab Testing

A column cartridge is a cylindrical container within the nitrification column that is packed with the growth media. It is the 'bread and butter' of the column. The column cartridge test was used to analyze the system efficiency and hydraulic retention time characteristics. These cartridges were hand constructed, along with a cartridge holding stand. This can be seen in the image at the left.

There were 4 cartridges that were created: two 40cm, a 60 cm, and an 80 cm cartridge. All of these cartridges have a diameter of 1.25 inches. Two pumps were collected and match with tubing. It should be noted that 2000mL beakers were used as inflow reservoirs and effluent collection systems.

There are two methods for starting up the system. The first was to pre-saturated the media. We did this by filling a 2000mL beaker with the pine growth media and saturating it with tap water and 300mL API bacteria batch sample. We added 25mL of undiluted urine to this system to start the nitrification process in the beaker.

The second was the 'system start-up fluid'. This system startup fluid was effectively a cartridge 'flush' with the API nitrifying bacteria batch sample. The volumes used to flush

the system are described in the following table.

Table 5: Column Cartridge System Flush Volumes

This completed the set-up for the column cartridge test.

3.2Column Cartridge Test Mythology

1600 mL of urine diluted was created at a 1:3 ration. The initial data characteristics of this sample were collected. This included the dissolved oxygen concentration, ammonia concentration, and total nitrogen concentration. The 1600mL was split evenly into two 1000 mL beakers to act as inflow reservoirs. This sample was pumped through both columns at a rate of 1.6 mL/min. There were three main trials conducted. These are described by the following table.

Table 6: Column Cartridge Trial Schedule

It took approximately 8.5 hours for the influent to be pumped completely through the column. At which point the effluent characteristic data was collected and analyzed. Observations were recorded throughout the entirety of this process.

3.3Column Cartridge Results and Analysis

The raw data from these cartridge trials can be found in the appendix, *8.5 Column Cartridge Test Raw Data.* It should be noted that more data was collect then described in these three trials, this can be found in the same section of the appendix.

The influent and effluent total nitrogen in the system was measured for each test. However, there was only a plus or minus concentration of approximately 1%. Thus, we can conclude that there was no loss of nitrogen within the system. A mass balance based on initial ammonia concentration is described in the appendix.

3.3.1 Trial #1

The percent efficiency of this test is described by the following table.

Table 7: Trial #1 Efficiencies

The following figure gives an illustration of the Trial #1 test results.

Figure 7: Trial #1 Data Illustration

This trial seemed to result in some very odd data. It appears that for the pre-saturated column there was an increase in the initial ammonia concentration and a negative organic nitrogen concentration. This is not true. We believe the reason for this odd data is the residual nitrogen in the growth media that escaped out with the effluent.

3.3.2 Trial #2

The percent efficiency of this test is described by the following table.

Table 8: Trial #2 Efficiencies

The following figure gives an illustration of the Trial #2 test results.

Figure 8: Trial #2 Data Illustration

This trial demonstrates that system efficiency increases with the column length. The 60cm cartridge test showed the greater decrease in ammonia and greater increase in nitrate.

3.3.3 Trial #3

The percent efficiency of this test is described by the following table.

Table 9: Trial #3 Efficiencies

The following figure gives an illustration of the Trial #3 test results.

Figure 9: Trial #3 Data Illustration

This cartridge test has a significant reduction in nitrate being produced. We believe this is because of ammonia concentration entering the column was almost three times stronger. As can be expected the 80 cm column had the greatest system efficiency.

3.2 Column Cartridge Conclusions

From these column cartridge tests we can make six conclusions.

This type of system can effective preform the process of nitrification.

Figure 10: Final Design Image

- The start-up fluid flush would be a better means to starting up the bacteria colonies within the system cartridges.
- Increasing the length of the column increases the column efficiency.

 Cartridges above 80 cm in length will have greater than 75% efficiency.

• Increasing ammonia concentration will decrease the nitrate production.

Cartridges start to loss efficiency after two weeks.

4.0 Technical Section: Final Design Testing

4.1 Final Design Selection and Set-Up

We wanted a minimum of 80% efficiency in this column design. We previously found that 80cm had an efficiency of about 75%. For easy of construction a 120 cm Column height was selected with a 2 cm diameter. Increasing the diameter allowed for an increased loading rate. We hypothesized that this would grant us our desired 80% system efficiency. This system was design with the API system start-up fluid.

This design was constructed with 2 plastic cartridges, the housing unit, and miscellaneous purchases totaling about \$135. This system was then constructed, drilling oxygen intake wholes within the column cartridges. This is illustrated by the figure on the left.

4.2Final Design Test Methodology

1600 mL of urine diluted was created at a 1:3 ration. The initial data characteristics of this sample were collected. This included the dissolved oxygen concentration, ammonia concentration, and total nitrogen concentration. The 1600mL was added to a 2000 mL beaker that acts as inflow reservoirs. This sample was pumped through both columns at a rate of 2 mL/min. Unfortunately, due to time constraints only one trial could be conducted. It took approximately 13.5 hours for the influent to be pumped completely through the column. At which point the effluent characteristic data was collected and analyzed. Observations were recorded throughout the entirety of this process.

4.3Final Design Results and Analysis

The percent efficiency of this test is described by the following table.

Table 10: Final Design Result Efficiencies

The following figure gives an illustration of the final trial result.

The influent and effluent DO and Total Nitrogen values are described by the following table.

Figure 11: Final Design Illustration

Table 11: Final Design Additional Data Collection

There was sufficient oxygen flow in the final design, this is shown with an oxygen increase in the system. It can be concluded that no nitrogen was lost within the system.

4.4Final Design Conclusion

The final design was able to accomplish the goal of 80% efficiency. The final design also significant increased the nitrate production. The design team was expecting a nitrate increase in the 30% 's. The design team believes that the increased nitrate production was due to improved oxygen flow within the system.

5.0 Technical Section: Economic Analysis

5.1 Cost of Implementing the Final Design

The following table describes the costs to implement the final design for a single person use.

Table 12: Final Design Cost

This is a relatively low cost design. However, the above figure does not consider the cost of electricity or water used to operate the system. Considering these values were not within the scope of this project.

5.2 Cost of Services

The following table describes the total projected cost of services of this project.

Table 13: Projected Cost of Services

The following table describes the actual project cost of services.

Table 14: Actual Cost of Services

The following table describes the classifications for each of the above abbreviations

Table 15: Classification Abbreviation

The following table describes the above list of material costs.

Table 16: Cost of Materials

As can be seen from the above tables, the actual project cost was close to twice as much as the projected cost. The only explanation was that the design team seriously underestimated the amount of lab hours that would be required for the completion of this project. This is discussed in more detail in the appendix, *8.7 Project Schedule and 8.8 Staffing*.

6.0 Triple Bottom Line

6.1 Economics Impacts

Because this relatively new technology there is not much information regarding the economic impacts regrading this technology. The design team does believe that this will be low cost because this is a low tech low maintenance technology. The design team also believes that this system is sustainable for home production of fertilizer

6.2 Environmental Impacts

This can be very beneficial to the environment because the effluent can be used to fertilize plants in home gardens and landscapes. The design team believes that this type of system can be scaled up for include fertilizer for agriculture in more rural areas. If this is done however, possible eutrophication in can occur if extreme amounts of nutrients un off into bodies of water. This will createexcessive bacteria and algae growth whom consume all the DO in the water. This will create dead zones for fish and other wildlife [8].

6.3 Social Impacts

Approximately 2.5 billion people have noaccess to adequate waste treatment. These people mostly reside in rural areas. There is also roughly a 115 million tons of nitrogen nutrients used annually, meaning this product will be used [9,10].

The design team does not believe that this technology is very applicable in more developed areas because they don't need this lower less advanced form of waste treatment.The design team does not believe that these areas will change the cartridges regularly, every two weeks.

7.0 References

- [1] Fumasoli, A., Etter, B., Sterkele, B., Morgenroth, E., &Udert, K. M. (2016). Operating a pilot-scale nitrification/distillation plant for complete nutrient recovery from urine.Water Science & Technology, 73(1), 215-222. doi:10.2166/wst.2015.485
- [2] "Welcome to API Fishcare: QUICK START: Safety Data Sheet", *Apifishcare.com*, 2016. [Online]. Available: http://www.apifishcare.com/product.php?id=618#.WE8hkuYrKHt. [Accessed: 12- Dec- 2016].
- [3] Arp, Daniel J(Sep 2009) Nitrification. In: eLS. John Wiley & Sons Ltd, Chichester. http://www.els.net [doi: 10.1002/9780470015902.a0021154]
- [4] Office of Water, "Nitrification," 15 August 2002. [Online]. Available: http://www.epa.gov/sites/production/files/2015-09/documents/nitrification_1.pdf. [Accessed: 22 Jan 2016].
- [5] "eCFR Code of Federal Regulations", *Ecfr.gov*, 2016. [Online]. Available: http://www.ecfr.gov/cgi-bin/ECFR?page=browse. [Accessed: 12- Dec- 2016].
- [6] L. Science, "What Causes Fertilizer Explosions | Ammonium Nitrate", *Live Science*, 2016. [Online]. Available: http://www.livescience.com/28841-fertilizer-explosions-ammoniumnitrate.html. [Accessed: 05- Dec- 2016].
- [7] "Manufactures Water Quality Testing and Analytical Instruments & Reagents | Hach USA", *Hach.com*, 2016. [Online]. Available: http://www.hach.com/. [Accessed: 05- Dec- 2016].
- [8] "World fertilizer trends and outlook to 2018", *Food and Agriculture Organization*, 2016. [Online]. Available: http://www.fao.org/3/a-i4324e.pdf. [Accessed: 05- Dec- 2016].
- [9] "NOAA's National Ocean Service Education: Estuaries", *Oceanservice.noaa.gov*, 2016. [Online]. Available:

http://oceanservice.noaa.gov/education/kits/estuaries/media/supp_estuar09b_eutro.html. [Accessed: 05- Dec- 2016].

- [10]"Northern Arizona University", Nau.edu, 2016. [Online]. Available: http://nau.edu/. [Accessed: 05- Dec- 2016].
- [11] "File:Zdrs0232.jpg MicrobeWiki", *Microbewiki.kenyon.edu*, 2016. [Online]. Available: https://microbewiki.kenyon.edu/index.php/File:Zdrs0232.jpg. [Accessed: 12- Dec- 2016].
- [12]"NitrobacterWinogradskyi - Blog Coral Wonders", *Blog Coral Wonders*, 2016. [Online]. Available: http://blog.coralwonders.com/en/bacterias-en-el-acuario-marino/nitrobacter-winogradskyi/. [Accessed: 12- Dec- 2016].

8.0 Appendix

8.1 Nitrifying Bacteria Images

The following image illustrates the Nitrosomonas bacteria [11].

Figure 12: Bacteria in Step 1

The following image illustrates the Nitrobacter bacteria [12].

Figure 13: Bacteria in Step 2

8.2Additional Standard Methods

As mentioned previously, due to time constraints these standard methods could not be implemented though they would have been beneficial. The following table describes a list of these standard methods.

Table 17: List of Standard Unused Methods

Conductivity

Electrolytic conductivity is the capacity of ions in a solution to carry electrical current and is the reciprocal of the solution resistivity. In many cases, conductivity is linked directly to the total dissolved solids. The conductivity test is useful for nitrification column to determine the urine quality before and after the nitrification process.

Phosphorus, Total

This standard method is not critical for our project. This would be an optional method that would provide insight into another nutrient, phosphorus that this system will produce.

Salinity

Urine has a lot of dissolved salts in its composition. The exact measure of the concentration of these salts will provide important information in regard to how this salt will build up in the system.

8.3 WCH Nitrifying Bacteria Batch Monitoring

Our initial system had a pH of 7.1, however, the pH declined when the urine was added. This was expected due to the increase of free hydrogen-ions caused by the urea dissolving in the sample. As the system oxidized, converting the ammonia, the pH of our system began to increase steadily. The pH data is displayed in the graph below.

Testing alkalinity was critical in understanding the properties these bacteria. The following equation was used to determine the alkalinity of our sample. Where N=0.017.

Equation 3: Alkalinity

$$
Alkality\left(mg\frac{CaCO3}{L}\right) = \frac{A*N*50,000}{50\ (mL)}
$$

The initial Alkalinity of the sample was very high but with the addition of urine the alkalinity declined as the ions present reacted with the added hydrogen ions. This is a good demonstration as to why it was necessary to dilute the inflowing urine with tap water.

Like the pH, the alkalinity recovered as the system approached equilibrium. However, the rate in which alkalinity increases in the system is dependent on the content of the water added to the system. The addition of tap water provided the necessary ions when added on a consistent basis. This is illustrated in the following graph.

Figure 15: WCH Alkalinity Chart

The system ammonia change is significant due to the urine additive. The maximum concentration in the system is 78 mg/L. After 10 days the ammonia concentration decreased back down to 7.5 mg/L. This shows the nitrification process is functional. Over the course of these experiments, the system was kept consistently at 25C. The Chemical Oxygen Demand of the system was consistently above 2000mg/L. This is described by the following graph.

Figure 16: WCH Ammonia Chart

8.4 Beaker Test Raw Data

The following table illustrates the data that was collected from the beaker test to select desired growth media and nitrifying bacteria.

Table 18: Beaker Test Raw Data

Test#	Day	COD (mg/L)	$NH3$ (mg/L NH ₃ -N)	$NO2$ (mg/L NO ₂ -N)	$NO3$ (mg/L NO ₃ -N)
Beaker #1	$\mathbf{1}$	2145	5.9	0.15	9.22
	$\overline{2}$	5320	12.3	0.63	16.5
Beaker #2	$\mathbf{1}$	2144	6.1	0.10	9.79
	$\overline{2}$	5400	10.6	0.54	15.3
Beaker #3	$\mathbf{1}$	2169	6.1	0.13	11.2
	$\overline{2}$	2640	13.9	0.29	14.6
Beaker #4	$\mathbf{1}$	2131	6.3	0.12	10.0
	$\overline{2}$	3170	17.2		20.5
Beaker #5	$\mathbf{1}$	3402	20.9	0.98	20.7
Beaker#6	$\mathbf{1}$	4459	18.7	1.19	24.2

The table above shows alternatives for growth medias and bacterial source. The test pine and cedar, show the greatest increase in present Nitrate. The API source of nitrifying bacteria appear to come with less interference from other bacterial activity. The concentration for ammonia in each of the systems was found to be higher than the previous test, even though there was an increase in the nitrate concentration. This discrepancy is due to the presence of ammonia Chloride in the system breaking down into ammonia due to the increased presence of hydrogen ions. Any loss of nitrate is due to the formation of nitrogen based gases. These gases form naturally in water based systems due to mixing and the air-surface interface of water. In the next stage of testing, these values will potentially become negligible over time as the system stabilizes.

8.5 Column Cartridge Test Raw Data

All of the following tables have four compounds that are measured. These all have individual units, however, these values are directly comparable. This is because these units are only measuring the nitrogen in the compound. For example, the units of ammonia, (mg/L NH3-N), are measuring the amount of nitrogen in the form of ammonia.

The organic nitrogen of the system was determined from a mass balance not of total nitrogen in the system, but rather from the ammonia in the system. This was done with the following equation.

Equation 4: Organic Nitrogen Equation

Ammonia Converted to Organic Nitrogen

= Initial Ammonia − SUM(Effluent Ammonia + Effluent Nitrite + Effluent Nitrite)

8.5.1 Trial #1 Data

The following table describes the raw data collected from trail #1. The Total Nitrogen for this test was 1565 mg/L.

Table 19: Trial #1 Raw Data

8.5.2 Trial #2 Data

The following table describes the raw data collected from trail #2. The Total Nitrogen for this test was 2260 mg/L.

Table 20: Trial #2 Raw Data

8.5.3 Trial #3 Data

The following table describes the raw data collected from trail #3. The Total Nitrogen for this test was 3400 mg/L.

Table 21: Trial #3 Raw Data

8.6 Final Design Raw Data

Table 22: Final Design Raw Data

8.7 Project Schedule

The following table compared predicted task completion to actual task completion.

Table 23: Project Schedule

8.8 Staffing

The table below represents the hour load for each completed task and sub task for each team member.

Table 24: Staffing Table

The following table represents the hours for each position.

Table 25: Hours Classification Table

