

Impact of Environmental Factors on the Algae Overgrowth in Pond Water

Mingzhu Victoria Huang

Emma Willard School, Troy, NY, USA

ABSTRACT

Algae overgrowth and ecosystem deterioration at the pond located at the Estate of Chadds Ford, Pennsylvania was noticed and potential factors were studied to understand the cause. The environmental factors such as air temperature, sunlight, bacteria, and human activities were considered to be associated with the algae growth. Algae density was monitored through visual observations and water samples were collected and tested for the analysis of bacteria counts and pH of the water according to EPA methods. At the time of each water sample collection, the weather temperature and condition were recorded. The water samples were also observed under a microscope to study the types of algae present. The major species of algae found included *Cyanobacteria* and *Chromerida*. Algae presence was seasonal and related to temperature and bacteria counts. Bacteria counts drastically increased with the increase of temperature which can be explained because algae and bacteria formed a complex ecosystem. The pH of the water was greater than the EPA water limit of 8.5 when Algae abundance is high during summer. Commonly used nutrients containing T-phosphates and/or O-phosphorous in fertilizers were detected in the water samples which also affected the water pH. In this study, important factors associated with algae overgrowth were identified. Global warming would exacerbate to the pond water and ecosystem deterioration. Human activities such as the long-term application of lawn fertilizers and pesticides that were washed into the pond by rainfall promoted the growth of microorganisms which resulted in the pond water and ecosystem deterioration.

Introduction

The community of Estates at Chadds Ford (EACF) was built starting from 2006 and was finished around 2010. There is a natural pond in the community with an area of around 150 feet × 300 feet.

Initially, the pond water was clear during all months of the year, and thriving wildlife was present including fish, turtles, and various species of birds. Throughout the years, there has been a visible decrease in the pond water quality, mainly seen in the overgrowth of algae as well as less wildlife.

With an interest to know what the cause of the changes are, the pond water quality was monitored throughout the seasons. By knowing what is in the water and factors that may have caused the algae overgrowth, we can further understand the impact of the factors not only on the pond water, but also on our living environment in large, therefore find the potential feasibility of solutions to control the algae growth and environmental protection.

Background

Exploration of Water Quality

The first pond water sample was collected on June 21, 2021 and was sent to the ETR Laboratories (Environmental Testing and Research Laboratories, Inc, 60 Elm Hill Ave., Leominster, MA, USA) for a full environmental scan to detect if harmful chemicals were present in the water. The tests included:

- Bacteria: total Coliform, *E. Coli*, and total Bacteria Count
- Mineral Chemistry
- Radiochemistry
- Heavy Metals
- Other Parameters such as PCB (Polychlorinated Biphenyls), MBAS (Methylene Blue Active Substances) H₂S, O-phosphate, and T-phosphorus
- Volatile Organics
- Pesticides
- Herbicides

It is common that bacteria test does not meet EPA environmental criteria. Although it may not pose a near risk to human health in a near term, it raises and an awareness for us to take the preventive measures. The tests for Mineral Chemistry provide us knowledge about the chemical composition in the water.

All other tests listed above such as Radiochemistry, Heavy Metals, Other Parameters (including PCB (Polychlorinated Biphenyls), MBAS (Methylene Blue Active Substances) H₂S, O-phosphate, and T-phosphorus), Volatile Organics, Pesticides, Herbicides are critical to human health. A lot of the chemicals are carcinogens, or may cause a long-term damage in digestion, nerve, brain or reproductive systems. Fail of EPA limits will pose an immediate risk to human health.

The test results in ETR Laboratories Report #624212091 (See Section 8.0, Appendix) show that although most of the results were within the normal range of EPA standards, the sample failed the tests for total Coliform, total Bacteria count, and pH. The water contained Coliform which should not be present and the total bacteria count was greater than 160,000 cfu/100 mL, exceeding the EPA limit of 50,000 cfu/100 mL. The water pH of 8.7 was also beyond the basicity limit of 8.5.

Understanding of Total Coliform, Bacteria, and Algae – Bacteria Interaction

Total Coliform

According to US EPA, total coliforms are a group of related bacteria that are relatively safe to humans. A variety of bacteria, parasites, and viruses, known as pathogens, can potentially cause health problems if humans ingest them [1]. Total coliforms are present throughout the environment and are found in the soil, water, and human or animal waste. Total coliforms and *E. Coli* were tested according to US EPA Method 1604: Total Coliforms and *Escherichia coli* in Water by Membrane Filtration Using a Simultaneous Detection Technique (MI Medium) [2].

Bacteria

The total bacteria count (TBC) of a substance is an estimate of the number of microorganisms present in a sample. This measurement is represented by the number of colony-forming bacterial units (CFU) per gram (or milliliter) in the sample. Bacteria is one of the factors that can lead to corrosion. The higher the bacterial count, the greater the probability is for the occurrence of microbiological influences of corrosion MIC [3].

Algae – Bacteria Interaction

My observations of algae overgrowth and excessive amount of bacteria counts happened at the same time, which indicated that there was an interactive relationship between the two microorganisms in the pond water. Several studies show that heterotrophic bacteria have an extensive role in algal growth and survival [4-8]. It

was proven that heterotrophic bacteria not only decomposes plant and animal organic matter, but also promotes plant or algae growth by complex communication mechanisms and nutrient exchange [9]. Previous research studies have indicated a close interaction between algae and bacteria regardless of the types of interaction. Therefore, controlling the bacteria population will limit the amount of algae growth and vice versa.

pH Related to the Abundance of Algae and Bacteria

There are many different types of algae that favor different pH ranges, for example, spirulina (*Arthrospira*) prefers a pH of around 10 [10] while other algae cultures well in freshwater that is neutral at pH of around 7.0. Algae growth in fresh water is very common in swimming pools in the state of Florida due to pH and chemical off balance. Algae consume carbon dioxide during photosynthesis, and this consumption is responsible for an increase in pH [11]. Most bacteria thrive around neutral pH values (6.5 - 7.0), however, others can grow in very acidic or basic conditions as low as a pH of 1.0 [12].

Methods and Procedure

Visual Observation of Algae Abundance

Observation and analysis of algae density in the pond water was conducted throughout the year since June 2021 through March 2022 on Jun. 21, 2021, Aug. 28, 2021, Oct. 12, 2021, Nov. 23, 2021, Dec 23., 2021, Jan. 23, 2022, and Mar. 14, 2022. The extent of algae covering the pond water surface was observed visually throughout the duration of the study and photos of the water surface condition were taken periodically to record the condition of the algae growth.

Analytical Analysis of Water Samples

As indicated in Section 2.0, a full environmental scan of the first sample collected on Jun 21, 2021. Since the test of Coliform is not a quantitative test, the later samples were tested for the presence of bacteria and water basicity to determine if there are any relationship with the overgrowth of algae. Further, a long-term monitor of algae growth was conducted. The related factor such as air temperature, water clarity, and water temperature were recorded. In addition, the impact of sunlight on bacteria activity was studied. Moreover, the effect of human activities on algae growth was also considered.

All sampling bottles were provided by the ETR Laboratories. Water samples on June 21, 2021, August 28, 2021, and October 12, 2021, were filtered through an iron mesh before filling into the sampling bottles. Samples collected on November 23, 2021, December 23, 2021 and March 14, 2022 were not filtered since there were no algae observed on the water surface. The ETR sampling bottles were submerged under the water to fill the bottles which were filled completely to avoid any air between the bottle and cap.

All water samples were shipped to the ETR Laboratories on the same day via USPS priority mail to analyze the total bacteria count and pH. The air temperature of the testing day was recorded at the time of sampling using a thermometer (Taylor, Model 1476N). Total bacteria count tests were performed according to the Standard Plate Counts method, also known as Heterotrophic Plate Counts method [13]. The pH of the water samples were tested according to EPA Method 150.3: Determination of pH in Drinking Water [14].

Microscopic Observations of Water Samples

The water sample was also observed under a microscope (OMAX 40X-2500X LED Digital Trinocular Microscope) with a magnification range of 200× to 1000× to study the species of algae present.

Statistical Significance of Results

To study the relationship between the algae growth and the presence of bacteria and pH levels, all raw data including the test results from the ETR reports were entered into a Microsoft spreadsheet. The best fit regression model was used to evaluate the relationships between the different parameters. In this study, both linear and polynomial regression was used for the best fit of data.

Results

Algae Density at the Pond Water Surface

The first sampling point was at 1:10 pm on June 21, 2021. The weather was clear and bright and the air temperature was 30.5°C at the time of collection. 95% of the water surface was covered with algae and the water could not be seen through (shown in Figure 1A). There were algae both on and below the surface. The second sampling point was at 1:10 pm on August 28, 2021. The air temperature was 26.1°C at the time of collection which was relatively low for a mid-day in the month of August. 100% of the water surface was covered with algae (shown in Figure 1B) and the algae in water were much denser compared to that in June. The third sampling point was at 7:50 am on October 21, 2021. The algae density decreased at this point as only about 70% of the water surface was covered (shown in Figure 1C). The weather was clear and bright and the air temperature was 19.4°C at the time of collection. Although the algae were much less dense compared to August, the water surface was still covered with algae and could not be seen through. The samples collected on these three days were filtered through a stainless steel mesh before filling into the ERT sampling bottles. The sample collected on Oct. 21 was easier to filter compared with those of the previous two sampling points, which further supported the reduction of algae.

On both November 23, 2021 and December 23, 2021, there were two sampling points on each day, evaluate the effect of solar energy on bacterial activities. Samples on November 23 were collected at 7:10am before sunrise and at 4:40pm before sunset; the air temperature was 1.9°C and 7.2°C respectively while the water temperature was 3.9 °C and 5.5°C. Samples on December 23 were collected at 9:30am after sunrise and at 5:00pm at sunset to monitor the difference in bacteria counts during the day; the air temperature was 4.0 °C and (-1.6) °C respectively, while the water temperature was 3.0 °C and 3.9°C respectively.

On both of these two days, there were no algae on the water surface and the water was transparent, shown in Figures 1D and 1E, respectively. The water was clear and there was no need for water filtration. There were neither algae nor underwater vegetation present and ice had begun to form at the edge of the pond. On December 23, 2021, about 20% of the water surface was covered with ice about ¼ inch thick. By January

23, 2022, the water surface was completely covered with ice of about 3 inches thick (Figure 1F).



Figure 1. The appearance of water surface at the Pond (Photos taken by Mingzhu Victoria Huang)
A: Jun. 21, 2021; B: Aug. 28, 2021; C: Oct. 12, 2021; D: Nov. 23, 2021; E: Dec. 23, 2021; F: Jan. 23, 2022;
G: Mar. 14, 2022

The ice in pond surface started to melt in late February 2022. Samples were taken on March 14, 2022, when it was a clear sunny day, shown in Figure 1G. The air temperature was 15.0 °C and the water temperature was 7.7 °C. The water was clear, no algae were observed on the surface and inside of water. The observation data were tabulated in Table 1.

Impact of Temperature on Algae Growth

During the spring, the algae started to grow and reached its peak density during the summer. During fall when the temperature decreased, the amount of algae decreased as well. By late fall during the month of November when the air temperature dropped to single digits in °C, there was no algae observed on the water surface and the water was completely transparent and aquatic vegetation underneath the surface of the water could be observed. On December 23, the air temperature dropped below 0°C at night. No algae were observed and the water was clearer than it was in November. There was also much less aquatic vegetation in the water.

Table 1. Impact of Temperature on Algae Abundance. Data was taken from <https://www.wunderground.com>

Sampling Date	Sampling Time	Air Temp.(°C)		% of surface area covered by algae	Water Turbidity
		At the time	Average*		
06/21/2021	13:10	30.5	24.6	95	Blurry (cannot see through)

08/28/2021	13:10	26.1	23.9	100	Blurry with thicker algae (cannot see through)
10/12/2021	7:50	19.4	18.6	70	Less blurry (cannot see through)
11/23/2021	7:10	1.9	1.6	0	Transparent (see through and observed green water grass under the surface)
	16:40	7.2			
12/23/2021	9:30	-1.1	0.4	0 (~20% of surface covered by ice)	Clear (transparent and no green water grass under the surface)
	17:00	-1.6			
1/23/2022	17:00	1.0	-1.2	0 (100% of surface covered by ice)	No observation (no sample taken)
3/14/2022	15:30	15.0	8.1	0	Clear (transparent)

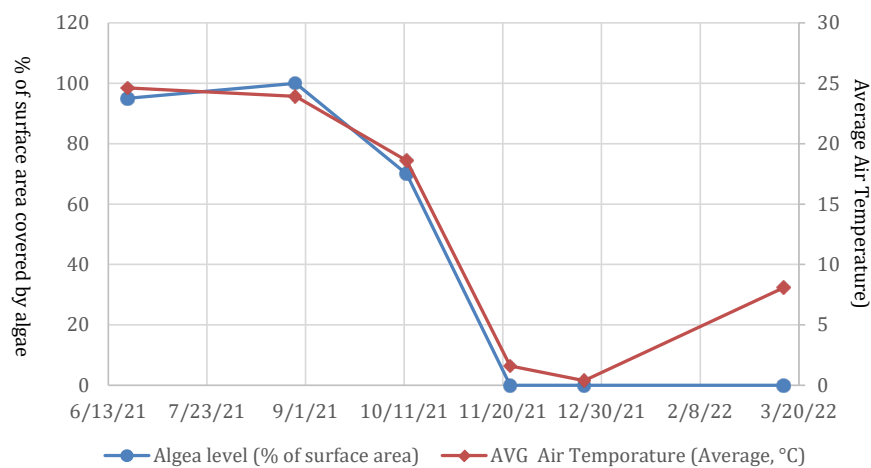


Figure 2. Algae Growth as the Functions of Air Temperature and Seasons

Observation of Algae under Microscope

The water sample collected on October 12, 2021, was observed under the microscope with 400× magnification shown in Figure 3. A large amount of *Cyanobacteria* was present in the water sample. *Chromerida*, a group of unicellular alveolates includes the photosynthetic species *Chromera velia* and *Vitrella brassicaformis* [15] was also found in the water sample.

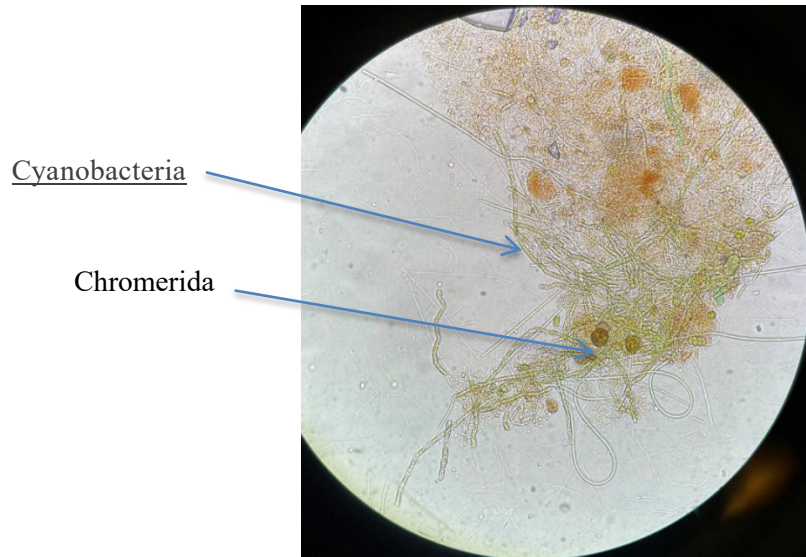


Figure 3. Microscopic observation of sample collected on Oct 12, 2021. Pond water under microscope with 400× magnification

Impact of Sunlight on Bacteria Count

In order to evaluate the impact of sunlight on the bacterial activity, two samples – one in the morning and the other in the afternoon were taken on November 23, and December 23, 2021. All data is summarized in Table 2.

Table 2. Impact of Sunlight on Bacteria Activities. Data were from ERT Laboratory Analytical Reports.

Date of Sampling	Sampling time	Air Temp. (°C)	Water Temp. (°C)	Algae level (% of surface area)	Bacteria Count ^a (×1000 cfu / 100mL)
11/23/2021	7:10 (at sunrise)	1.9	3.9	0	5.8
11/23/2021	16:40 (at sunset)	7.2	5.5	0	20.3
12/23/2021	9:30 (after sunrise)	4.0	3.0	0	13.2
12/23/2021	17:00 (after sunset)	-1.6	3.9	0	4.2

Algae Abundance Associated with Bacteria Count

The bacteria count for each sample is listed in Table 3 below. High bacteria counts were detected during the summer since the month of June and the testing results of the bacteria counts in June and August exceeded the EPA limit of 50,000 cfu/100mL, correlating well to the trend of algae growth. On October 12, 2021, around 70% of the water surface was covered by algae and the bacteria count was also drastically decreased. By early winter in late November, there was no algae observed on the water surface and the bacteria count was also within the EPA limit of 50,000 cfu/100mL. The average bacteria count of the samples taken on November 23, and December 23, 2021, along with data at other sampling points were used to complete the plot in Figure 4.

Bacteria Count Affected by Temperature

In order to evaluate the impact of temperature on bacteria count on the day, the average temperature of the sampling day was considered. In addition, the average value of the bacteria counts for the two sampling points

on both November 23 and December 23, 2021, was used. The available data from June 2021 to March 2022 was summarized in Table 4 and the graph of bacteria count vs. air temperature is shown in Figure 5.

Table 3. Impact of Bacteria Count on Algae Abundance

Date of Sampling	Sampling time	Algae level (% of surface area)	Bacteria Count ^c (×1000 cfu / 100mL)
06/21/2021	13:10	95	> 160 ^a
08/28/2021	13:10	100	> 160 ^a
10/12/2021	7:50	70	67.3
11/23/2021	7:10/16:40	0	13.1 ^b
12/23/2021	9:30/17:00	0	8.7 ^b
03/14/22	15:30	0	14.8

^a Result exceeded the detection limit.

^b Average of two values of the samples taken on the same day

^c Data were from ERT Laboratory Analytical Reports.

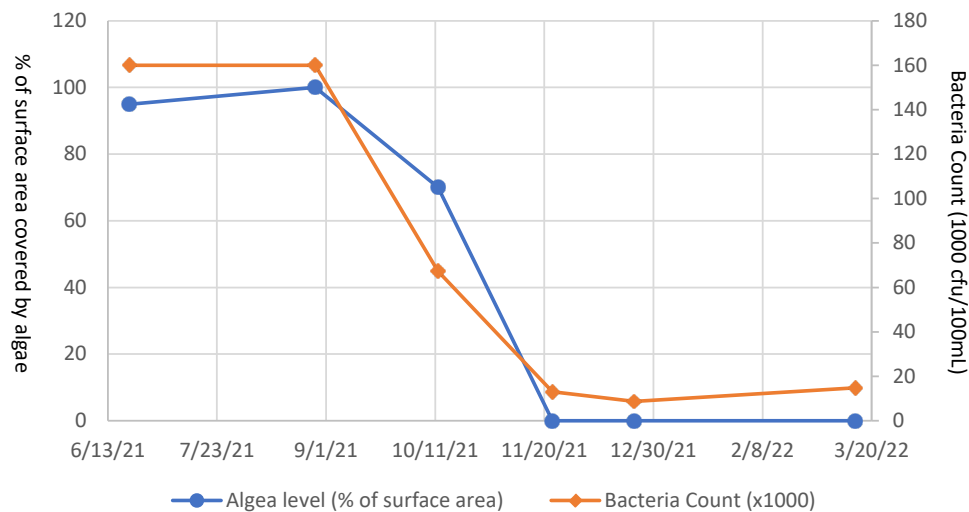


Figure 4. Algae Abundance as the Function of Bacteria and Seasons

Table 4. Bacteria counts change with temperature

Date of Sampling	AVG Daily Air Temperature (°C) ^a	Bacteria Count (x1000 cfu/100mL)
06/21/2021	24.6	160 ^b
08/28/2021	23.9	160 ^b
10/12/2021	18.6	67.3
11/23/2021	1.6	13.1 ^c = (5.8+20.3)/2
12/23/2021	0.4	8.7 ^c = (13.2+4.2)/2
03/14/2022	8.1	14.8

^a <https://www.wunderground.com>

^b Result exceeded the detection limit.

^c Average of two values

Bacteria count drastically increased with the increase of temperature with a polynomial relationship. According to the regression of the data, the polynomial function of bacteria count (B) dependent upon average daily air temperature (T) can be found as Equation 4. The correlation coefficient *r* value of the polynomial regression is 0.9977.

Equation 1:

$$B = 0.0179T^3 - 0.2375T^2 + 1.4378T + 9.4086$$

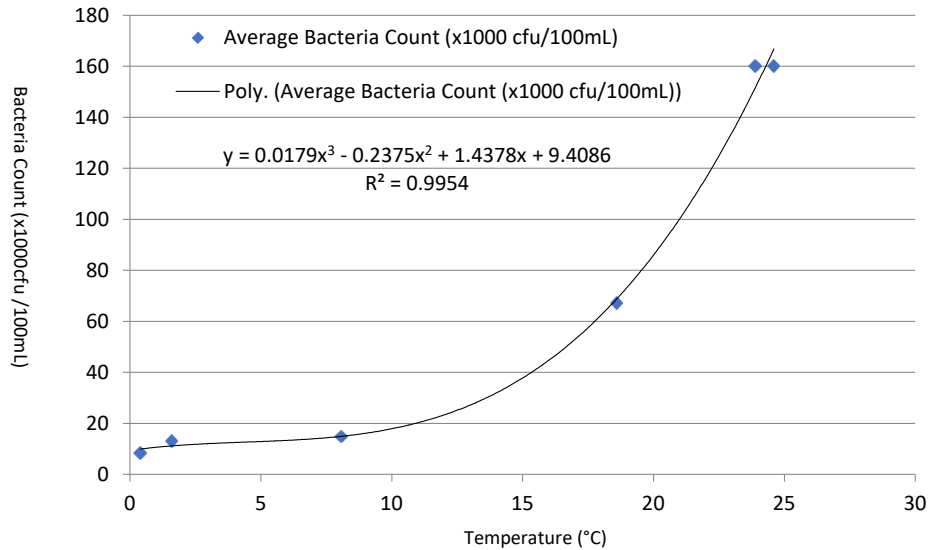


Figure 5. Bacteria Counts Change with Temperature

Water pH Associated with Algae and Bacteria Count

Testing results of pH values from ETR Laboratory Reports along with the algae observation results are tabulated in Table 5. The data indicated that pH of the pond water had a connection with the presence and the number of algae and bacteria. During the late fall and winter season when the number of algae was drastically decreased and bacteria counts were below 100k cfu/100mL, water pH values were much closer to neutral as shown in Table 5.

Table 5. pH Related to Algae Growth and Bacteria Count

Date of Sampling	Time of sampling	Algae density (% of surface area)	Bacteria Count (×1000 /100mL)	pH ^a
06/21/2021	13:10	95	>160 ^b	8.70
08/28/2021	13:10	100	>160 ^b	8.75
10/12/2021	7:50	70	67.3	6.74
11/23/2021	7:10 (at sunrise)	0	5.8	7.00
11/23/2021	16:40 (at sunset)	0	20.3	6.85
12/23/2021	9:30 (after sunrise)	0	13.2	6.91
12/23/2021	17:00 (after sunrise)	0	4.2	6.76
03/14/2022	15:30 (sunny)	0	14.8	7.09

^a Data from ETR Laboratory Analytical Reports

^b Result exceeded the method maximum measurement range.

Discussion

Impact of Temperature on Algae Growth

From the data in Table 1 and the chart shown in Figure 2, it was found that the amount of algae was dependent upon the change of seasons (dates) and air temperature (weather). The profile of algae abundance was almost identical to that of the temperature, although the algae level dependence on temperature on August 28 does not follow the same trend as the other data points. The abundance of algae on August 28 was greater than that on June 21, but the temperature on August 28 was lower than that on June 21. This is because the algae growth is affected not by the weather in just one day, but the duration of a period.

Therefore, the algae in the pond water were mainly due to seasonal changes. During the spring, the algae started to grow and reached its maximum density during summer. During the fall, the algae growth decreased along with the air temperature and during late fall transitioning into winter; the algae began to die as well other aquatic vegetation below the water. As shown in Figure 2, algae abundance correlated well with the air temperature as the function of seasons during summer, fall, and winter. The algae growth did not start in early spring when the temperature was just rise up.

Impact of Sunlight on Bacteria Count

The water temperature did not change as much as the air temperature did as shown in Table 2. However, the bacteria count was more than three times higher at the time when there was hours of sunlight compared to that when there was no sunlight for hours. The data suggests that bacterial activities are very active during the day when there was direct sunlight.

Algae Abundance Associated with Bacteria Count

The data in Table 3 demonstrates a reasonable correlation between the bacteria counts and algae abundance. When there were higher amounts of algae on the water surface during the summer, the detected bacteria counts were higher as well. In late fall and winter when the air temperature was lower, both algae abundance and bacteria counts dropped.

Bacteria Count Affected by Temperature

When temperatures were higher during the summer, and the bacteria count significantly increased, far exceeding the EPA limit. During the same time, there was significant amount of algae in water, which suggested that algal abundance played a ubiquitous role in the increase of bacteria counts. As the algae abundance changed throughout the seasons, the bacteria count changed as well. Temperature had a large impact on both algae and bacteria. When no algae were observed on the water surface during November and December, bacteria counts were also reduced to a level within the EPA limit of 50,000 cfu/100mL. Algae and bacteria were affected by each other through complex communication mechanisms and nutrient exchange, and were interdependent and benefit from each other [8].

Impact of Human Activities on Algae Growth

Besides the environmental factors such as the changes of season and weather, the drastic growth of algae during the spring and summer may also be due to chemicals such as fertilizers and pesticides used by residents. Human activities such as applying lawn fertilizers by residents may have the impact on the overgrowth of algae. Commonly used nutrients containing T-phosphate and/or O-phosphorous were detected in the water sample (Appendix: ETR Laboratories Report #6242122091), although the amounts of the nutrients detected were below the EPA limits. Rainfall brought the fertilizers into the pond which fed both algae and bacteria and promoted the growth of microorganisms. The actual amounts of these chemicals could have been much more than what were detected in the water sample since large amount of these chemicals might have been absorbed by the microorganisms and quickly decomposed. A natural process that results from the accumulation of nutrients that may occur from lawn fertilizers, or eutrophication phenomenon, could also play a significant role in algae – bacteria interrelation. The eutrophication process in the pond resulted from fertilizer runoff from residential properties into the water, excessive nutrients, and algae death which is decomposed by bacteria. The decomposition of algae increased the oxygen demand in the pond which resulted in the death of various species of aquatic life. These activities would also lead to the surface of the pond being covered by algae worse and worse year after year.

Water pH Associated with Algae and Bacteria Count

On Nov. 23 and Dec. 23, 2021, there was no algae observed on the water surface, and the water pH at later afternoon was lower than that in the morning. This was further confirmed that the formation of CO_2 by active bacterial activities during the day resulted in a lower pH in the absence of algae conducting photosynthesis. Therefore, water pH was associated to the amount of both algae and bacteria. When no algae were present on the water surface during late fall and winter, the pH of the water was relatively neutral.

When there were higher amounts of algae or bacteria present during the late spring and summer, the pH levels exceeded the EPA limit of 8.5. As shown in Figure 6, on one hand, the activities of both algae and bacteria produced CO_2 which would make the water more acidic. On the other hand, photosynthesis by algae consumed CO_2 which would make the water less acidic or more neutral. In addition, during eutrophication process [16], the algae died or were decomposed by bacteria. The metabolites and decomposed organic sediment made the water more basic. Furthermore, lawn fertilizers used by residents contain T-phosphates and/or O-phosphorous salts are usually basic which led to high algae abundance and basic water environment that is toxic to aquatic life.

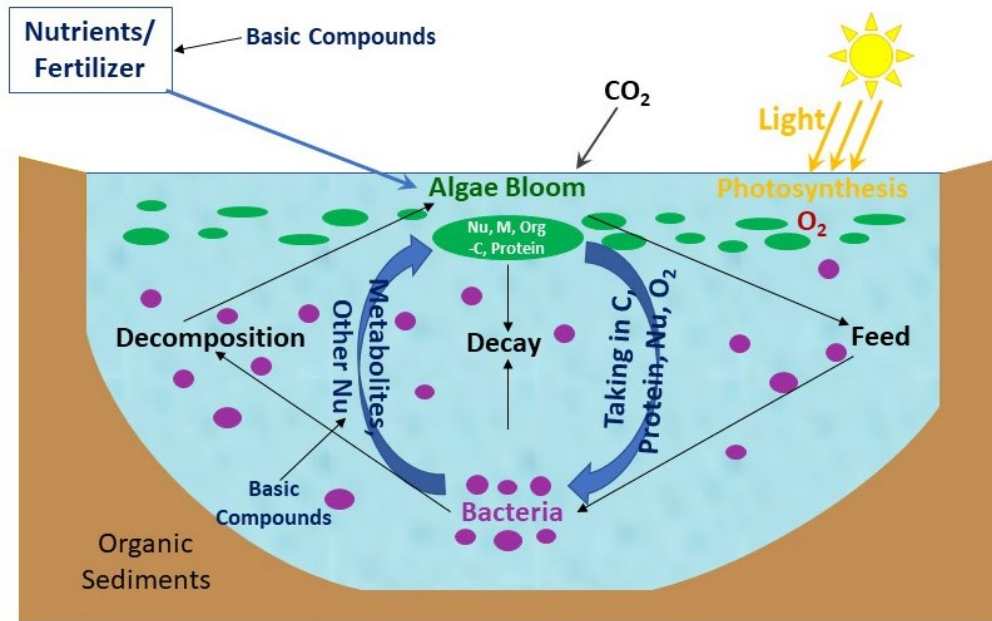


Figure 6. Respiration and Metabolite Cycle of Aquatic Microorganisms

Conclusion

In this study, important factors associated with algae overgrowth in the pond were identified which includes air temperature and bacteria in the water. As the season changed, the water transparency changed as well, becoming more transparent when temperatures dropped. The basic water pH of above 8.5 was caused by the decomposition of microorganisms in the water and chemicals in lawn fertilizers used by residents. Global warming or increasing air temperatures would exacerbate the pond water and ecosystem deterioration. Human activities such as applying lawn fertilizers and pesticides also have significant impacts on the pond environment. These chemicals washed into the pond by rainfall promoted the growth of microorganisms, and resulted in the basic pond water and ecosystem deterioration through eutrophication phenomenon. A long-term study on the water quality should be continued to better understand the impacts of human activities on the pond water and environment, and therefore, bring forth more effective solutions to preserve the environment.

Acknowledgments

The authors would like to thank Emma Willard School for providing the laboratory and equipment to study the bacteria. Additionally, we would like to thank the residents at Estate of Chadds Ford community for the financial support on this project.

References

1. Revised Total Coliform Rule And Total Coliform Rule | US EPA, reviewed in Aug. 2021
2. Method 1604: Total Coliforms and Escherichia coli in Water by Membrane Filtration Using a Simultaneous Detection Technique (MI Medium). US EPA, Sep. 2002

3. What is a Total Bacteria Count (TBC)? - Definition from Corrosionpedia, <https://www.corrosionpedia.com > definition>
4. Amin, S.A., Hmelo, L.R., van Tol, H.M., Durham, B.P., Carlson, L.T., Heal, K.R., et al., 2015. Interaction and signaling between a cosmopolitan phytoplankton and associated bacteria. *Nature* 522, 98–101.
5. Gonzalez, L.E., Bashan, Y., 2000. Increased growth of the microalga *Chlorella vulgaris* when coimmobilized and cocultured in alginate beads with the plant-growth-promoting bacterium *Azospirillum brasilense*. *Appl. Environ. Microbiol.* 66, 1527–1531.
6. Kim, B.-H., Ramanan, R., Cho, D.-H., Oh, H.-M., Kim, H.-S., 2014a. Role of *Rhizobium*, a plant growth promoting bacterium, in enhancing algal biomass through mutualistic interaction. *Biomass Bioenergy* 69, 95–105.
7. Seyedsayamdost, M.R., Case, R.J., Kolter, R., Clardy, J., 2011. The Jekyll-and-Hyde chemistry of *Phaobacter gallaeciensis*. *Nat. Chem.* 3, 331–335.
8. Ramanan, R., Kim, B.-H., Cho, D.-H., Oh H.-M., Kim, H.-S. Algae–bacteria interactions: Evolution, ecology and emerging applications. *Biotechnology Advances* 34 (2016) 14–29
9. Philippot, L., Raaijmakers, J.M., Lemanceau, P., van der Putten, W.H., 2013. Going back to the roots: the microbial ecology of the rhizosphere. *Nat. Rev. Microbiol.* 11, 789–799.
10. <https://algaeresearchsupply.com/pages/algae-culture-and-ph>
11. Michael H. Gerardi, Brittany Lytle, *The Biology and Troubleshooting of Facultative Lagoons*. John Wiley & Sons, Inc. 2015
12. <http://www.brooklyn.cuny.edu/bc/ahp/CellBio/Growth/MGpH.html>
13. 9215 HETEROTROPHIC PLATE COUNT - Standard Methods For the Examination of Water and Wastewater, <https://www.standardmethods.org/doi/10.2105/SMWW.2882.188>
14. EPA Method 150.3: Determination of pH in Drinking Water, <https://nepis.epa.gov/Exe/ZyPDF.cgi/P100SFNI.PDF?Dockey=P100SFNI.PDF>
15. About: Chromerida (dbpedia.org), <https://dbpedia.org > page > Chromerida>
16. How Does Eutrophication Work? Causes, Process and Examples - Earth How, <https://earthhow.com/eutrophication-causes-process-examples>