FEEDING TIME

HOW NUTRIENTS DRIVE PHYTOPLANKTON GROWTH

Brianna Stanley
Virginia Institute of Marine Science

Grade Level
High School

Subject Area
Biology & Environmental Science
The 2019/2020 VA SEA project was made possible through funding from the National Estuarine Research Reserve System Margaret Davidson Fellowship Program which supports graduate students in partnership with research reserves where fieldwork, research, and community engagement come together. VA SEA is currently supported by the Chesapeake Bay National Estuarine Research Reserve, Virginia Sea Grant, and the Virginia Institute of Marine Science Marine Advisory Program.
Title Feeding Time: How Nutrients Drive Phytoplankton Growth

Focus How both nutrient availability and ratios impact levels of phytoplankton

Grade Level HS Biology, HS Environmental Science

VA Science Standards
- BIO.1 d) graphing and arithmetic calculations are used as tools in data analysis;
- BIO.2 a) water chemistry and its impact on life processes;
- BIO.8 c) succession patterns in ecosystems;
- BIO.8 d) the effects of natural events and human activities on ecosystems;

Learning Objectives
- Students will apply a data set to create a graph and to answer a question
- Students will identify limiting nutrients
- Students will recognize that proportions of nutrients are as important as concentration
- Students will distinguish important biological growth factors
- Students will reflect on their own contribution to eutrophication

Total length of time required for the lesson
75 minutes (60 minutes + optional 15 minutes second part)

Key words, vocabulary
Nitrogen – an element needed for biological growth. Phytoplankton can use multiple nitrogen-based nutrients, including ammonium and nitrate.
Phosphorus – an element needed for biological growth. Many phytoplankton use the nutrient phosphate as their source of phosphorous.
Nutrients – substances required for biological growth. This includes nitrogen, phosphorus, carbon, metals, and other complex compounds.
Nutrient limitation – when the nutrient supply suppresses additional biological processes, such as growth.
Redfield ratio – 106 carbon: 16 nitrogen: 1 phosphorus (see PowerPoint and background or more information).
Eutrophication – the increased input of organic matter.
Liebig’s Law of the Minimum – biological growth is not controlled by total resource availability, but rather by the availability of the scarcest resource. This scarce resource is known as the limiting factor.

Background Information
See background in PowerPoint

My name is Brianna Stanley and I am a graduate student at the Virginia Institute of Marine Science studying nitrogen cycling in both the Chesapeake Bay and in the Alaskan Arctic. I am interested in studying nitrogen because it is an important nutrient (food) source for phytoplankton. Phytoplankton form the base of the food web and are important members of our ecosystems. I am particularly interested in which nitrogen compounds phytoplankton prefer to use to grow, which can be important for managing our coastal ecosystems. Phytoplankton growth in our coastal ecosystems is a complex
process that is impacted by the availability of many different nutrients and environmental conditions. An important concept to understanding phytoplankton growth in our coastal waters is nutrient limitation, which can be explained by Redfield ratios. This lesson plan explores limiting factors through Redfield ratios, and provides students an opportunity to practice their graphing skills and their interpretation of ratios!

There are many different nutrients that are important for phytoplankton growth, which can include nitrogen, phosphorus, carbon, and even metals (i.e. nickel, iron etc.). For water quality management, two of the most important nutrients are nitrogen and phosphorus because they are often limiting nutrients. Limiting nutrients limit the growth of phytoplankton in a concept often referred to as Liebig’s law of the minimum. The growth of phytoplankton is believed to be limited by the nutrient that is most scarce (reviewed in Sarmiento & Gruber, 2006). That is, even if one nutrient is plentiful, if there is not enough of another necessary nutrient, the growth will be limited by that scarce nutrient.

This however, is not as simple as looking for the nutrient present in the lowest or highest quantity. Proportions, or ratios, of the nutrients present can also matter. For example, phytoplankton need more nitrogen than phosphorus to grow. In the 1930s, Alfred Redfield, a prominent oceanographer, proposed what is known today as the Redfield ratio. Redfield found that the ratio of the important nutrients of carbon, nitrogen, and phosphorus were found throughout the world in the ratio 106 carbon: 16 nitrogen: 1 phosphorus (Gruber & Deutsch, 2014). This is also the ratio of the nutrients that phytoplankton need! We can then use this ratio to determine if a system is nutrient limited for a particular nutrient. Focusing on nitrogen and phosphorus, if the nutrients measured in an ecosystem are in a ratio >16:1, it is phosphorus-limited and if the ratio is <16:1, it is nitrogen-limited (Howarth 1988). Researchers can use this as a tool to understanding how ecosystems function and to make important management decisions.

Materials & Supplies
- Projector/screen for introductory PowerPoint
- Worksheets (Appendix 1), 1 group sheet per group and 1 individual sheet per person
  - Multiple versions available
- Key (Appendix 2)
- Optional: Calculator
- Optional: Plastic baggies, split peas, and 2 other bean varieties
  - Water samples
    - 4+ plastic baggies (1+ per sample, prepare enough so each group gets 1 sample)
    - Split peas for phytoplankton cells
    - 2 bean varieties, for nitrogen and phosphorus, respectively.
    - If you are preparing 1 set per sample type, you will need:
      - Split Peas (phytoplankton) - 30
      - Type 1 (nitrogen) – 138 beans
      - Type 2 (phosphorus) – 9 beans

Teacher Preparation
Students will be working on this activity in small groups of 2-3. There is an option for the student groups to collect their own data by providing them with “water sample” baggies to collect data. If this option is taken, group size may be adjusted based on the supplies available to the instructor. Using available
supplies, add the different beans in each plastic bag (water sample). Make sure that the number of split peas (phytoplankton) and beans (nutrients) matches the data in the provided data table. This ensures students will get the correct ratios. There are two versions of the activity sheets to support either the data collection activity or to interact with provided data if time/supplies are limited. Note that the data for the “provided data” worksheet, is slightly different than the counting exercise. Finally, there is an individual activity sheet (Appendix 1D). This can be used if teachers wish to have some students work independently.

Procedure
Introduction (~15 minutes) – PowerPoint may be utilized to introduce the idea of nutrient limitation and eutrophication. It can also be used to introduce the double y-axis graph activity.

Slide 1: Presentation Title
Slide 2: I am a graduate student studying nitrogen and how it moves throughout the environment. I’m particularly interested in how processes vary across the world. The first picture on this slide shows me doing lab work in Virginia. I do most of my work in Virginia, but I also get to travel to do research. The second photo shows me in front of the research vessel Sikuliaq. I got to spend almost a month on the Arctic Ocean, off of Alaska, to study how nitrogen cycling is changing there.

Slide 3: This slide introduces the idea of limiting factors for biological growth. Often referred to as Liebig’s law of the minimum, biological growth and activity is limited by the scarcest factor that is needed. One way to think of this is with how much water a barrel can hold. If the sides or slates are of varying height, it is the lowest that limits how much water the barrel can actually hold.

Slide 4: Environmental conditions can be an example of limiting factors. For example, if an organism requires sunlight for photosynthesis, access to sunlight can be a limiting factor if it is cloudy or winter time. Nutrients are another example of limiting factors and are the focus of today’s activity. Nutrients are like food for phytoplankton and can include nitrogen, phosphorus, carbon, and even metals!

Slide 5: The concept of the Redfield ratio is one way that biologists monitor limiting factors. The Redfield ratio is the ratio of important inorganic nutrients in both the ocean and inside phytoplankton biomass. The “golden” ratio is 106 carbon: 16 nitrogen: 1 phosphorus. This ratio can vary across ecosystems depending on local conditions, but the general ratio is upheld across the world! For now, knowing the difference between inorganic and organic nutrients are not needed. This is just included as a normality of the definition. If students ask, inorganic refers to nutrients that don’t contain carbon and are often thought of as an easier to use food source.

Slide 6-10: These slides introduce the idea of eutrophication. It is classically defined as “the increased supply of organic matter.” One way of thinking about this is that as nutrients are delivered to an ecosystem, more phytoplankton are able to grow, which leads to more biomass in the system. As more phytoplankton grow, certain phytoplankton that can cause harmful algal blooms can also grow. These can negatively impact the ecosystem by releasing toxins, but by also taking up the oxygen needed by fish to live. If you have more nutrients… you get more phytoplankton. And these phytoplankton can cause large blooms. One way scientists are studying these blooms is by focusing on the nutrients. Even with the increase in nutrients, the dependence on Redfield ratios still apply. As is shown in the next slide, this can help inform management decisions.
Slides 11-12: In modern ecology, we use the Redfield ratio to determine if an ecosystem is limiting for one of these critical nutrients. To do this, scientists focus on the nitrogen: phosphorus part of the ratio. By measuring the nutrients in the environment, scientists can then calculate ratios. When the measured N:P is greater than the ideal 16:1 (say 18:1), the system is labeled as phosphorus-limited, because there is less phosphorus than is needed. Another way of saying this is there will still be nitrogen left when all the phosphorus has been used by the phytoplankton. Even if there is nitrogen left, they can’t continue to grow because there is no longer any phosphorus. When the measured N:P is less than the ideal 16:1 (say 15:1), the system is labeled as nitrogen limited, because there is less nitrogen than is needed. In the environment, freshwater systems are usually phosphorus limited and saltwater systems are nitrogen limited. This can be important for management, especially when trying to combat eutrophication. For example, today most laundry detergent does not contain phosphorus because of the impact that nutrient has on our inland waterways (https://www.npr.org/2010/12/15/132072122/it-s-not-your-fault-your-dishes-are-still-dirty). With less phosphorus being imputed into the freshwater environment, fewer blooms are thought to occur there.

Slide 13: Introduces the activity for the day. If you are using the data collection version the table is empty.

Slides 14-15: One aspect that may be confusing to students is that the graphing activity includes a graph with 2 y-axes. This is an opportunity to introduce that concept to students. Scientists often use graphs with 2 y-axes because it allows us to visualize more data together and try to piece together relationships. In order to read the axes, you need to determine which measurement goes with which axes (will be guided in worksheet). Give the example of the red nutrient measurement goes with the y-axis labeled nutrient concentration. Green, then goes with phytoplankton cells.

Slide 16: This slide provides instructions for students on the activity. They can be used for more introduction or can be left up while students are doing the activity.

Slide 17(-18): Provides answers for the students for any final class discussions. Slide 17 in the counting version has the filled in table, which can be left up after the class shares their data.

Activity Part 1
Using provided dataset (~30 minutes)
1. Divide the class into groups (preferably 2-3 students each) and pass out one group worksheet (Appendix 1A) per group. Pass out one individual worksheet per student (Appendix 1C).
2. The first part of the activity asks students to graph their data with the 2 y-axes graph. Using graphing question 1 and the PowerPoint, help guide students to use both axes.
3. Once they graph their data, students will use the graph to make some initial observations about the data and the different levels of phytoplankton in their samples.
4. Next, students will individually calculate Redfield ratios. They may optionally use calculators, but to make the activity more challenging you may ask students to use math skills to simplify their ratios.
5. Using the final questions in the individual worksheet, students should reach the conclusion that the ratio is as important as the amount of food/nutrients.
6. As a challenge for advanced students, ask them to think about identifying other examples of limiting factors for growth.
7. Review results with students as a whole class.
With data collection option (~45 minutes)
1. Divide the class into groups (preferably 2-3 students each), ensuring that there are at least 4 groups. Distribute worksheets (Appendix 1B), using the set with an empty data table, and one “water sample” to each group. Pass out one individual worksheet per student (Appendix 1C). (Note: If students are working individually instead of in groups, use the combined worksheet Appendix 1D.)
2. Have groups collect their water sample data and then share their data with the class.
3. Using their collected data, they will then graph in groups. The activity asks students to graph their data with the 2 y-axes graph. Using graphing question 1 and the PowerPoint, help guide students to use both axes.
4. Once they graph their data, students will use the graph to make some initial observations about the data and the different levels of phytoplankton in their samples.
5. Next students will calculate Redfield ratios. They may optionally use calculators, but to make the activity more challenging, you may ask students to use math skills to simplify their ratios.
6. Using the final questions in the individual worksheet, students should reach the conclusion that the ratio is as important as the amount of food/nutrients.
7. As a challenge for advanced students, ask them to think about identifying other examples of limiting factors for growth.
8. Review results with students as a whole class.

Optional Activity Part 2* (~15 minutes)
1. Make sure to emphasize slide 6-10 on the PowerPoint to introduce the concept of eutrophication.
2. Divide the class into groups of 3-4 to answer the two discussion questions given in the group worksheet.
3. Pass out one group worksheet per group.
4. Call on groups to discuss their answers as a class.

*Option: if students finish part 1 faster than the rest of the class, part 2 could be added to make the expansion question section longer. This could also be a take home reflection or research assignment.

Assessment
The main forms of assessment in this activity are the student worksheets and their final discussion of the last two questions. As the instructor works through student answers, they should focus on the student’s ability to create the graph, interpret ratios, and to think critically about their data.

References
Feeding Time: How Nutrients Drive Phytoplankton Growth

Part 1: Nutrient Limitation
The availability of nutrients is an important environmental factor that can help fuel the growth of phytoplankton in our waters. In order to understand which nutrients are the most important for different waterways, many scientists study which nutrients are limiting. They do this by monitoring nutrient concentrations in water samples. Using the table below, answer the provided questions about nutrient limitation in several water samples.

### Data Collection Table

<table>
<thead>
<tr>
<th>Water Sample</th>
<th>Measurement</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Phytoplankton (x (10^5) cells)</td>
</tr>
<tr>
<td>Sample 1</td>
<td>2</td>
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<td>2</td>
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<tr>
<td>Sample 3</td>
<td>18</td>
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<td>Sample 4</td>
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</table>

Graphing the Data

1. When scientists need to visualize complex data with different types of measurements they will sometimes use graphs with two different y-axes. The bar graph below has a different y-axis for “Nutrient Concentration” and another for “Phytoplankton.” Which axis are you going to use for each of your measurements (Phytoplankton, Nitrogen, and Phosphorus)?

2. Use the following bar graph to visualize your data. Fill in the key with a different color/pattern for each measurement (Phytoplankton, Nitrogen, and Phosphorus).
Key

- Phytoplankton
- Nitrogen
- Phosphorus
Optional Part 2: Eutrophication
Discuss the following questions in your groups. Be prepared to further discuss as a class.

1. In the samples above, we saw how phytoplankton respond to different nutrient conditions. Do you think it is always good to have limitless food? Explain your answers.

2. Do we as a class contribute to eutrophication?
   a. Brainstorm what actions you have taken today that could led to nutrients entering our coastal waters?

   b. List possible ways to reduce your contribution to nutrient pollution.
Feeding Time: How Nutrients Drive Phytoplankton Growth

Part 1: Nutrient Limitation
The availability of nutrients is an important environmental factor that can help fuel the growth of phytoplankton in our waters. In order to understand which nutrients are the most important for different waterways, many scientists study which nutrients are limiting. They do this by monitoring nutrient concentrations in water samples.

1. In your groups you will be collecting data from a water sample. In the table below, record your sample # and your collected measurements. Don’t forget to note what type of bean/bead etc. you are using to represent each measurement.

2. As a class, share your collected data for your different water samples. This will allow your group to graph the data and for you to answer important questions about the data.

Data Collection Table

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Graphing the Data
3. When scientists need to visualize complex data with different types of measurements they will sometimes use graphs with two different y-axes. The bar graph below has a different y-axis for “Nutrient Concentration” and another for “Phytoplankton.” Which axis are you going to use for each of your measurements (Phytoplankton, Nitrogen, and Phosphorus)?
4. Use the following bar graph to visualize your data. Fill in the key with a different color/pattern for each measurement (Phytoplankton, Nitrogen, and Phosphorus).

**Key**

- Phytoplankton
- Nitrogen
- Phosphorus
Optional Part 2: Eutrophication

Discuss the following questions in your groups. Be prepared to further discuss as a class.

3. In the samples above, we saw how phytoplankton respond to different nutrient conditions. Do you think it is always good to have limitless food? Explain your answer.

4. Do we as a class contribute to eutrophication?
   a. Brainstorm what actions you have taken today that could led to nutrients entering our coastal waters?

   b. List possible ways to reduce your contribution to nutrient pollution.
C. Individual handout

Name: ______________________

Date: _______________

Feeding Time: How Nutrients Drive Phytoplankton Growth

Part 1: Nutrient Limitation
Using the data table and your group answers, work individually to answer the following questions about nutrient limitation in several water samples.

Thinking About the Data

1. Looking at the data, which sample had the most phytoplankton? Why do you think the sample you picked had such a high cell count?

2. One of the concepts used to understand biological and nutrient processes in coastal waters is the Redfield ratio. To grow, many phytoplankton need nutrients to be available in the Redfield ratio of 16 units nitrogen: 1 unit phosphorus. Calculating the Redfield ratio for water samples allows researchers to determine if nitrogen or phosphorus is the nutrient limiting phytoplankton growth. Such that when:

   Measured ratio > 16:1 phosphorus is limiting
   Measured ratio < 16:1 nitrogen is limiting
   Measured ratio = 16:1 neither nitrogen nor phosphorus are limiting

   a. Using your data, calculate the Redfield ratios to determine what nutrient conditions are available to the phytoplankton in each of your water samples.

   Sample 1
   Nitrogen : Phosphorus

   Sample 2
   Nitrogen : Phosphorus

   Sample 3
   Nitrogen : Phosphorus

   Sample 4
   Nitrogen : Phosphorus
b. For each of your water samples, indicate if your calculated ratio is greater than or less than 16:1.

c. For each of your water samples, indicate which nutrient is limiting, if any.

3. In Question 1, you found one water sample had more phytoplankton. Now that you have identified your limiting nutrients, write why you think each of your other three water samples did not have as many phytoplankton as your choice in Question 1.

4. Answer the following questions about nutrients.
   a. Based on your water samples which nutrient is needed for phytoplankton growth? Nitrogen? Phosphorus? Both?

   b. Does the quantity of nutrients matter? Explain your answer using the concept of Redfield ratios.

Expansion Question:

Nutrients are just one of many environmental factors that are required for phytoplankton to grow. What are some other environmental factors that may act as a limiting factor for phytoplankton growth?

*Hint: Remember that many phytoplankton are similar to plants.*
D. Activity Compiled into One Handout

Name: ________________________________

Date: __________________

Feeding Time: How Nutrients Drive Phytoplankton Growth

Part 1: Nutrient Limitation

The availability of nutrients is an important environmental factor that can help fuel the growth of phytoplankton in our waters. In order to understand which nutrients are the most important for different waterways, many scientists study which nutrients are limiting. They do this by monitoring nutrient concentrations in water samples.

3. Today you will be collecting data from a water sample. In the table below, record your sample # and your collected measurements. Don’t forget to note what type of bean/bead etc. you are using to represent each measurement.

4. As a class, share your collected data for your different water samples. This will allow you to graph the data and for you to answer important questions about the data.

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Graphing the Data

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6. Use the following bar graph to visualize your data. Fill in the key with a different color/pattern for each measurement (Phytoplankton, Nitrogen, and Phosphorus).

Key

= Phytoplankton      = Nitrogen      = Phosphorus
Thinking About the Data

1. Looking at the data, which sample had the most phytoplankton? Why do you think the sample you picked had such a high cell count?

2. One of the concepts used to understand biological and nutrient processes in coastal waters is the Redfield ratio. To grow, many phytoplankton need nutrients to be available in the Redfield ratio of 16 units nitrogen: 1 unit phosphorus. Calculating the Redfield ratio for water samples allows researchers to determine if nitrogen or phosphorus is the nutrient limiting phytoplankton growth. Such that when:

   Measured ratio > 16:1 phosphorus is limiting

   Measured ratio < 16:1 nitrogen is limiting

   Measured ratio = 16:1 neither nitrogen nor phosphorus are limiting

   a. Using your data, calculate the Redfield ratios to determine what nutrient conditions are available to the phytoplankton in each of your water samples.

   **Sample 1**
   
   Nitrogen : Phosphorus

   **Sample 2**
   
   Nitrogen : Phosphorus

   **Sample 3**
   
   Nitrogen : Phosphorus

   **Sample 4**
   
   Nitrogen : Phosphorus

   b. For each of your water samples, indicate if your calculated ratio is greater than or less than 16:1.

   c. For each of your water samples, indicate which nutrient is limiting, if any.
3. In Question 1 (in “Thinking About the Data”), you found one water sample had more phytoplankton. Now that you have identified your limiting nutrients, write why you think each of your other three water samples did not have as many phytoplankton as your choice in Question 1.

4. Answer the following questions about nutrients.
   a. Based on your water samples which nutrient is needed for phytoplankton growth? Nitrogen? Phosphorus? Both?

   b. Does the quantity of nutrients matter? Explain your answer using the concept of Redfield ratios.

Expansion Question:

Nutrients are just one of many environmental factors that are required for phytoplankton to grow. What are some other environmental factors that may act as a limiting factor for phytoplankton growth?  
*Hint: Remember that many phytoplankton are similar to plants.*
Optional Part 2: Eutrophication
Answer the following questions. Be prepared to further discuss as a class.

5. In the samples above, we saw how phytoplankton respond to different nutrient conditions. Do you think it is always good to have limitless food? Explain your answer.

6. Do we as a class contribute to eutrophication?
   a. Brainstorm what actions you have taken today that could led to nutrients entering our coastal waters?

   b. List possible ways to reduce your contribution to nutrient pollution.
Appendix 2: KEY

Name: ___________KEY______________
Date: ________________

Feeding Time: How Nutrients Drive Phytoplankton Growth

Group

Part 1: Nutrient Limitation

If provided data differs from “collected” data, the provided data is in parenthesis. These differences are to avoid issues with counting 0.5 values.

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Graphing the Data

7. When scientists need to visualize complex data with different types of measurements they will sometimes use graphs with two different y-axes. The bar graph below has a different y-axis for “Nutrient Concentration” and another for “Phytoplankton.” Which axis are you going to use for each of your measurements (Phytoplankton, Nitrogen, and Phosphorus)?

Phytoplankton – Phytoplankton Cells
Nitrogen – Nutrient Concentration
Phosphorus – Nutrient Concentration

8. Use the following bar graph to visualize the data from your water samples. Fill in the key with a different color/pattern for each measurement (Phytoplankton, Nitrogen, and Phosphorus).
Provided data:

Key

- Phytoplankton
- Nitrogen
- Phosphorus
Collected data:

Key

= Phytoplankton

= Nitrogen

= Phosphorus
Optional Part 2: Eutrophication

Discuss the following questions in small groups and be prepared to further discuss as a class.

7. In the samples above, we saw how phytoplankton respond to different nutrient conditions. Do you think it is always good to have limitless food? Explain your answer.

   Answers will vary depending on comprehension of material in PowerPoint lecture included. Students should indicate an understanding of algae blooms and that these can sometimes hurt the local ecosystem.

8. Do we as a class contribute to eutrophication?
   a. Brainstorm what actions you have taken today that could led to nutrients entering our coastal waters?

   Many answers are possible, but may include eating food grown with fertilizer, flushing the toilet, ridding on the school bus or driving, used school supplies that are manufactured.

   b. List possible ways to reduce your contribution to nutrient pollution.

   Many answers are possible, but may include using low phosphate products at home and reducing consumption.
Individual

Thinking About the Data

1. Looking at the data, which sample had the most phytoplankton? Why do you think the sample you picked had such a high cell count?
   Sample 3, because there was a large quantity of nutrients available. (Answers will vary depending on how the activity is set-up by instructors)

2. One of the concepts used to understand biological and nutrient processes in coastal waters is the Redfield ratio. To grow, many phytoplankton need nutrients available in the Redfield ratio of 16 units nitrogen: 1 unit phosphorus. Calculating the Redfield ratio for water samples allows researchers to determine if nitrogen or phosphorus is the nutrient limiting phytoplankton growth. Such that when:

   Measured ratio > 16:1 phosphorus is limiting

   Measured ratio < 16:1 nitrogen is limiting

   Measured ratio = 16:1 neither nitrogen nor phosphorus are limiting

   a. Using your data, calculate the Redfield ratios to determine the nutrient conditions are available to the phytoplankton in each of your water samples.

   **Sample 1**
   Nitrogen  22:2 (11:1)  Phosphorus  
   - less than, nitrogen limitation

   **Sample 2**
   Nitrogen  27:1.5 or 36: 2 (18:1)  Phosphorus  
   - greater than, phosphorus limitation

   **Sample 3**
   Nitrogen  40:2.5 or 48:3 (16:1)  Phosphorus  
   -equal, no nitrogen or phosphorus limitation

   **Sample 4**
   Nitrogen  24:1.5 or 32:2 (16:1)  Phosphorus  
   -equal, no nitrogen or phosphorus limitation

   b. For each of your water samples, (a) indicate if your calculated ratio is greater than or less than 16:1, and (b) which nutrient is limiting, if any.
   See above

   c. For each of your water samples, indicate which nutrient is limiting, if any.
   See above
3. In Question 1 of this part, you found one water sample had more phytoplankton. Now that you have identified your limiting nutrients, write why you think each of your other three water samples did not have as many phytoplankton as your choice in Question 1.

   Sample 1 – Not enough nitrogen
   Sample 2 – Not enough phosphorus
   Sample 4 – Not enough total nutrients

4. Answer the following questions about nutrients.
   a. Based on your water samples which nutrient is needed for phytoplankton growth? Nitrogen? Phosphorus? Both?

      Both nutrients are needed

   b. Does the quantity of nutrients matter? Explain your answer using the concept of Redfield ratios.

      Yes, generally, the more nutrients the more phytoplankton, but the nutrients need to be present in the Redfield ratios, otherwise their growth becomes limited.

Expansion Question:

Nutrients are just one of many environmental factors that are required for phytoplankton to grow. What are some other environmental factors that may act as a limiting factor phytoplankton growth?

Hint: Remember that many phytoplankton are similar to plants.

Possible answers include: sunlight, oxygen (air okay), temperature, season.