

## **ISSUES : DATA SET**

### **Forest Ecosystem Responses to Chronic Nitrogen Additions**

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#### **THE ECOLOGICAL QUESTION:**

How have pine and hardwood stands in a temperate forest responded to 15 years of nitrogen additions?

#### **ECOLOGICAL CONTENT:**

forest ecosystem, nitrogen (N) deposition, primary productivity, soil respiration, microbial community, replication

#### **WHAT STUDENTS DO:**

Students work with data to ask and address questions about effects of chronic N additions to two forest types. Four Microsoft Excel files on net primary productivity, tree photosynthetic capacity, soil respiration, and soil microbial biomass allow students to consider effects of long-term N enrichment in northeast US forests. Students in a basic ecology class can use part of the data set; more advanced students, in a plant ecology class for example, can work up all the data.

#### **SKILLS:**

Asking and addressing questions with data, critical thinking, graph preparation, data analysis and interpretation, and writing or oral presentation.

#### **ASSESSABLE OUTCOMES:**

Simple statistics (means and standard errors), graphs from spreadsheet data, and written and oral communication of results and conclusions.

#### **SOURCE:**

Bauer, G.A., F.A. Bazzaz, R. Minocha, S. Long, A. Magill, J. Aber, and G.M. Berntson. 2004. Effects of chronic nitrogen additions on tissue chemistry, photosynthetic capacity, and carbon sequestration potential of a red pine (*Pinus resinosa* Ait.) stand in the NE United States. *Forest Ecology and Management* 196:173-186.

Bowden, R.D., E. Davidson, K. Savage, C. Arabia, and P. Steudler. 2004. Chronic nitrogen additions reduce total soil respiration and microbial respiration in temperate forest soils at the Harvard Forest. 2004. *Forest Ecology and Management* 196:43-56.

Frey, S.D., M. Knorr, J. Parrent, and R.T. Simpson. 2004. Chronic nitrogen enrichment affects the structure and function of the soil microbial community in a forest ecosystem. *Forest Ecology and Management* 196:159-171.

Magill, A.H., J.D. Aber, W.S. Currie, K.J. Nadelhoffer, M.E. Martin, W.H. McDowell, J.M. Melillo, and P. Steudler. 2004. Ecosystem response to 15 years of chronic nitrogen additions at the Harvard Forest LTER, Massachusetts, USA. *Forest Ecology and Management* 196:7-28.

## OVERVIEW

*This section is written for faculty who can modify the Overview as appropriate for their students. "Student Instructions" also include introductory material.*

### The research

Inputs to the terrestrial nitrogen (N) cycle have more than doubled in the past century due to anthropogenic activities, particularly fertilizer use and fossil fuel combustion (Vitousek, 1997; Vitousek et al., 1997). Global anthropogenic N inputs now exceed non-anthropogenic sources (e.g., biological nitrogen fixation). One consequence of this human-driven change in the N cycle is the significant increase in N that is being deposited as wet or dry deposition in forest ecosystems. At the local and regional scale, N deposition near industrialized and agricultural areas can greatly exceed those in unpolluted areas of the world (Vitousek et al., 1997). Rates of N deposition are particularly high in the Midwestern and Northeastern U.S. and in Europe. Since most temperate forest ecosystems evolved under low N conditions, chronic N enrichment would be expected to alter ecosystem properties and processes in fundamental ways.

Aber et al. (1989) suggested that the response of forest ecosystems to N enrichment would vary depending on the duration of the additions. They hypothesized that positive responses (e.g., increases in net primary production—NPP) would be observed in the short-term, while negative responses (e.g., increased nitrate leaching, reduced NPP) would be observed over the long-term. They established the Chronic Nitrogen Addition Experiment at Harvard Forest in 1988 to test these hypotheses. Results from the first 15 years of this experiment were recently detailed in a special issue of *Forest Ecology and Management* (Aber and Magill, 2004). The purpose of this activity is to illustrate how chronic N additions at Harvard Forest have impacted a range of ecosystem properties and processes, including both plant and soil responses.

Responses to added N at the Harvard Forest have been dramatic (Magill et al., 2004). Long-term fertilization has led to increased N concentrations in leaf litter and roots. Tree mortality has been high in the high N pine stand (56% by 2002), and biomass accumulation has stopped altogether. The high N hardwood stand shows increased aboveground net primary productivity relative to the control plot, but excess N availability coupled with a severe drought in 1995 led to high mortality of red maple trees in this plot (72% by 2002). Soil respiration was stimulated by N additions during the first year of fertilization in the hardwood stand; however, in the second year, soil respiration was not significantly different between control and fertilized hardwood plots and was suppressed by up to 25% in the pine fertilized plots (Bowden et al., 2004). After 13 years of continuous N fertilization, soil respiration in the high N plots was suppressed by 40% in both stands. Reductions in soil respiration are concomitant with reductions in microbial biomass, due primarily to a significant reduction in the fungal component of the microbial community (Frey et al., 2004). Further details of the results from the first 15 years of this experiment can be found in Aber and Magill (2004).

## References

- Aber, J.D., K.J. Nadelhoffer, P. Steudler, J.M. Melillo. 1989. Nitrogen saturation in northern forest ecosystems. *BioScience* 39:378-386.
- Aber, J.D. and A.H. Magill. 2004. Chronic nitrogen additions at the Harvard Forest (USA): the first 15 years of a nitrogen saturation experiment. *Forest Ecology and Management* 196:1-5.
- Bowden, R.D., E. Davidson, K. Savage, C. Arabia, and P. Steudler. 2004. Chronic nitrogen additions reduce total soil respiration and microbial respiration in temperate forest soils at the Harvard Forest. 2004. *Forest Ecology and Management* 196:43-56.
- Frey, S.D., M. Knorr, J. Parrent, and R.T. Simpson. 2004. Chronic nitrogen enrichment affects the structure and function of the soil microbial community in a forest ecosystem. *Forest Ecology and Management* 196:159-171.
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- Vitousek, P. 1997. Human alteration of the global nitrogen cycle: causes and consequences. *Issues in Ecology* No. 1. Ecological Society of America. <http://www.esa.org/science/Issues/>.
- Vitousek, P.M., J.D. Aber, R.W. Howarth, G.E. Likens, P.A. Matson, D.W. Schindler, W.H. Schlesinger, and D.G. Tilman. 1997. Human alteration of the global nitrogen cycle: sources and consequences. *Ecological Applications* 7:737-750.

## Resources

- <http://www.aber.sr.unh.edu/chronicn/hfintro.html>  
An online description of the Chronic Nitrogen Addition Study at Harvard Forest
- <http://harvardforest.fas.harvard.edu>  
Harvard Forest

## STUDENT INSTRUCTIONS

Over the last century or so, humans have greatly increased the amount of nitrogen (N) released into the atmosphere. For instance, combustion of fossil fuels such as gasoline and coal results in emission of N containing gases such as nitrous oxide, nitric oxide and ammonia. Some of this N returns to earth in rainfall and snow. You are probably familiar with the term "acid rain"; the nitric acid we now see in rain is largely the result of fossil fuel combustion and other human activity.

Both the northeast US and central Europe have been especially impacted by this long-term deposition of N due to wind patterns and population density. While N is a fertilizer that we apply to gardens and agricultural fields, it seems likely that the continuous addition of N over time to temperate forest and grassland ecosystems will have negative consequences because these systems evolved under low N conditions. The Chronic Nitrogen Amendment Study was established in 1988 at the Harvard Forest in central Massachusetts to evaluate how long-term N additions influence soil processes (such as nitrogen cycling), growth of trees and other plants, and the chemical composition of leaves and needles. Over the last 15+ years, two types of common forests — hardwood and pine — have been fertilized with two levels of N. You will be working with data collected in two adjacent forest stands. One, a pine stand, is dominated by mature red pine (*Pinus resinosa*). The other is a hardwood stand of black (*Quercus velutina*) and red (*Quercus rubra*) oak mixed with black birch (*Betula lenta*), red maple (*Acer rubrum*), and American beech (*Fagus grandifolia*).

Three plots measuring 30 x 30 m were established in each stand and have been treated annually with 0, 5, or 15 g N m<sup>-2</sup> (control, low N, and high N, respectively). There is one 30 x 30 m plot per treatment in each stand for a total of six plots. Nitrogen fertilizer is applied at 4-week intervals from May to September as a concentrated solution of ammonium nitrate (NH<sub>4</sub>NO<sub>3</sub>). The amount of N applied to the low N plots is similar to the amount of N deposited in some European ecosystems. The N addition in the high N plots is not intended to be representative of current N deposition levels, but rather to push the system to saturation, the point at which the forest can no longer use and retain the added N. This study method is called a "dose for time substitution." As Alison Magill, the plot manager, notes "We don't have time to put on 12 kilograms per year and wait 60 years to see the results because this type of research is, hopefully, oriented toward getting results that can be used to bring about changes in environmental policy."

The objectives of this exercise are for you to 1) increase your understanding of N deposition and its impact on forest ecosystems and 2) learn to ask questions and then make and interpret figures with actual scientific data from a Long-term Ecological Research (LTER) site. The ecosystem properties and processes that you will examine are net primary productivity (net rate of primary production), tree photosynthetic capacity (photosynthesis of leaves and needles), soil respiration, and soil microbial biomass. The data you will work with are published in a special issue of the journal *Forest Ecology and Management* (Aber and Magill 2004). This special issue reports the results from the first 15 years of N additions at Harvard Forest. Throughout the exercise you should keep in mind the following question: How have the pine and hardwood stands responded to long-term N fertilization?

## Reference

- Aber, J.D. and A.H. Magill. 2004. Chronic nitrogen additions at the Harvard Forest (USA): the first 15 years of a nitrogen saturation experiment. *Forest Ecology and Management* 196:1-5.

## Data Set 1

### Methods and Rationale

Forest decline has been seen in Europe as a result of long-term N additions. One goal of the Chronic Nitrogen Amendment Study at Harvard Forest is to determine how tree growth in northeast forests will respond to long-term N fertilization. All trees greater than 5 cm in diameter in the control, low N, and high N plots were tagged at 1.5 m above ground level in 1988 with numbered aluminum tags (Magill et al. 2004). Tree diameters were measured at tag height during fall/winter of 1990, 1993, 1996, 1999, and 2002.



Ring for measuring DBH (diameter at breast height) used for assessing tree growth. The tree diameters were used to estimate year-end aboveground tree biomass. Net primary productivity is the change in total cumulative tree biomass between time periods. The data are expressed as grams of tree biomass per square meter ( $\text{g m}^{-2}$ ). (Photo courtesy Serita Frey, University of New Hampshire. Used with permission.)

### Open-ended Questions

Think about the following questions and how you will go about answering them given the data provided.

1. How did tree productivity change over time in each of the forest stands, and how was productivity altered by N additions? Consider possible reasons to explain the pattern that you see in the data.
2. How is the effect of N additions different for the pine and hardwood stands? Why might there be a difference?

**Detailed Graphing Instructions**

Using Excel (instructions are for Excel 2003), make a figure of tree productivity from 1988 to 2002. To do this:

1. Click on the chart wizard icon in the toolbar.
2. Choose "XY (Scatter)" for the chart type and "Scatter with data points connected by lines" for the chart sub-type.
3. Click next.
4. Select data for the graph by clicking on the "Collapse Dialog Button" at the end of the "Data range" box. You will now be able to select the data from the worksheet. Highlight the entire dataset including headers (i.e., A6 — G11). Click the "Collapse Dialog Button" again to return to the chart wizard.
5. Select "Series in: columns"
6. Click next and label the chart and axes appropriately.
7. Customize the figure and click "Finish" when through.

**Dataset 2****Methods and Rationale**

One of the most consistent responses of forests to N fertilization is increased leaf tissue N (Magill *et al.* 2000). At Harvard Forest, pine needles (pine stand) and oak leaves (hardwood stand) collected from the N fertilized plots showed increased levels of N in both green and senesced (i.e., dead) leaf tissue, especially for the high N plots. You might predict that an increase in leaf N content would stimulate photosynthesis. Bauer *et al.* (2004) measured leaf photosynthesis of pine needles in the control and high N plots. The measurements were carried out in the tree canopies using two 25 m tall walk-up towers. Photosynthesis was measured for needles with ages of 0 to 3 years. Note that there were no 3-year needles in the high N plots. The data are expressed as the amount of CO<sub>2</sub> consumed in nmol per gram of leaf tissue per second (nmol g<sup>-1</sup> s<sup>-1</sup>).

**Open-ended Questions**

Think about the following questions and how you will go about answering them given the data provided.

- How do needles of different ages vary in their photosynthetic capacity?
- How does N fertilization affect photosynthetic capacity?
- Do needles of different ages show different responses to N additions?
- How do these results help explain what you observed for dataset 1?

**Detailed Graphing Instructions**

Prepare a bar graph that shows how photosynthetic capacity differed between the control and high N plots for needles of different ages. Follow the instructions given for Dataset 1 except select "Column" for chart type and "Clustered Column" for chart sub-type. Highlight only the mean values and their headings to start. You will need to label the bars. To do this:

1. Select the "Series" tab in the second dialog box of chart wizard (i.e., the box where you select the data for the graph.)
2. Click on the "Collapse Dialog Button" at the end of the "Category (x) axis labels" box.
3. Select the labels column (A8-A11).

Label the graph and axes appropriately. Looking at the figure, you will notice differences between the control and fertilized treatments, but you have no way of determining whether these differences are significant. To determine whether the control and fertilized plots are significantly different, you need to examine the amount of variability associated with each mean value. This information is provided in the dataset as the **standard error**. Use these data to add y-axis error bars to your graph. To do this:

1. Right click on the first bar in the graph and select "Format Data Series..."
2. Click on the "Y Error Bars" tab.
3. For the Display select "Both" and for the Error Amount select the Custom box.
4. Use the Collapse Dialog Buttons as before to select the standard errors associated with the highlighted bars (e.g., control data). Do not select the heading. You will need to select the same data twice, once for the "+" box and once for the "-" box.
5. Click OK.
6. Repeat these steps for the next set of bars (i.e., high N data).

As a general rule, if the error bars do not overlap, a significant difference exists between the mean values being compared.

### Dataset 3

#### Methods and Rationale

In the first two datasets you explored how N additions have impacted tree growth. The next two datasets focus on belowground (soil) responses. With dataset 3 you will specifically evaluate how soil respiration has responded to N fertilization. Globally, soils contain more carbon stored as organic matter than is contained in aboveground plant biomass and the atmosphere combined. Soil respiration is the primary way that carbon, as carbon dioxide (CO<sub>2</sub>), leaves the soil and enters the atmosphere. The metabolic activity of both roots and soil organisms (e.g., fungi, bacteria) contribute to soil respiration. Small changes in soil respiration may dramatically alter the concentration of CO<sub>2</sub> in the atmosphere. Thus there is great interest in how environmental change, including N deposition, is altering the amount of carbon stored in soil and the amount that enters the atmosphere as CO<sub>2</sub>.

Soil respiration measurements were made from three areas within each control, low N, and high N plot at Harvard Forest using plastic chambers placed over the soil for a 24 hour period (Bowden *et al.* 2004). A sample of air inside the chambers was removed every 4 hours and CO<sub>2</sub> concentrations were determined using a gas chromatograph. These measurements were made once or twice monthly from March to December in 1988, 1989, and 2001. The data in this dataset represent the amount of carbon respired as CO<sub>2</sub> per square meter of forest (g C m<sup>-2</sup>) during the measurement period for each of the three years. Standard errors are also provided.



Collar used for measuring soil respiration.  
(Photo courtesy Serita Frey, University of New Hampshire. Used with permission.)

### Open-ended Questions

Think about the following questions and how you will go about answering them given the data provided.

1. How does soil respiration vary from year to year?
2. Does soil respiration differ between pine and hardwood stands?
3. Is soil respiration affected by nitrogen fertilization? If so, how?

### Graphing Instructions

Using the skills you learned in the first two datasets, prepare a bar graph for each stand (hardwood and pine) that compares the cumulative soil respiration amongst the three treatments (control, low N, high N) for each of the three years that data were collected (1988, 1989, and 2001). Be sure to include error bars and to label the axes appropriately.

### Dataset 4

#### Methods and Rationale

Soil biota in many terrestrial ecosystems evolved under low N conditions and thus one anticipated consequence of elevated N inputs is a change in the soil biotic community as ecosystems move toward N saturation. Until recently, little work had been done at Harvard Forest to determine how the soil microbial community has responded to increased N availability or how changes in the microbial community might be linked to the observed changes in tree growth and soil respiration. Frey *et al.* (2004) collected soil samples in fall 2002 to examine how N enrichment has affected microbial biomass, microbial functional capacity, and the activities of cellulose-degrading and lignin-degrading enzymes produced by soil microorganisms. In this activity, you will analyze the bacterial and fungal biomass data.

Four soil samples were collected to a depth of 10 cm from each of the control, low N, and high N plots in both the hardwood and pine stands. The amount of bacterial and fungal biomass in the samples was estimated by the substrate-induced respiration method. In this method, glucose is added to the soil and the amount of soil respiration due to glucose metabolism is measured over a four hour period. Glucose is a readily usable food source for many soil organisms. Previous studies have shown that this glucose-induced respiration is well correlated to the amount of microbial biomass present in the soil. The data are reported in terms of the amount of CO<sub>2</sub> respired ( $\mu\text{g CO}_2 \text{ g}^{-1} \text{ soil}$ ) in response to glucose addition and thus represent an index of microbial biomass. Bacteria and fungi were differentiated with the use of antibiotics that are inhibitory to one or the other group of organisms. Respiration from soil amended with a bacterial inhibitor is attributed to fungi, and respiration from soil amended with a fungal inhibitor is likewise attributed to bacteria. In this way, the relative contribution of bacteria and fungi to total soil microbial biomass can be determined.

### Open-ended Questions

Think about the following questions and how you will go about answering them given the data provided.

1. Which microbial group, bacteria or fungi, represent the largest proportion of the total microbial biomass?
2. How does bacterial and fungal biomass vary across treatments?
3. Does the microbial response differ between the hardwood and pine stands? If so, how?
4. How do bacteria and fungi respond differently or similarly to N additions?
5. Compare these results to the soil respiration data above. How might changes in bacterial and/or fungal biomass influence soil respiration?

### Detailed Analysis and Graphing Instructions

In this dataset, you are given the raw data (i.e., the individual data points for each sample collected). First orient yourself to how the data are presented. Now get started by calculating the means and standard errors. You should do this in such a way as to make preparing graphs as easy as possible. To do this:

1. Click the cursor on cell H8 of the spreadsheet. In this cell, calculate the mean value for bacterial biomass for the control samples from the hardwood stand using the function "`=average(D8.D11)`."
2. Move to the next cell in that column (H9) and repeat this instruction for the pine stand. The function in this case will be "`=average(D20.D23)`."
3. Now move to the next column (I) and repeat these instructions for the low N treatment, changing the function accordingly.
4. Do the same in column J for the high N treatment.
5. Finally, repeat these instructions in columns K-M to calculate the mean values for fungal biomass for the control, low N, and high N samples.

You are halfway done.

Excel does not have a function for calculating the standard error; however, you can make this calculation by taking the standard deviation and dividing it by the square root of  $n$ , where  $n$  is the number of sample replicates (four in this case). To do this:

1. In cell N8 calculate the standard error for bacterial biomass in the control samples from the hardwood stand using the following function:  $=\text{stdev}(D8:D11) / \text{sqrt}(4)$ .
2. Modify this function as necessary to calculate the standard error associated with each mean value.
3. To make graphing easy, organize the standard errors in the same way as the means. Thus you should have 6 columns of mean values (columns H-M) and 6 columns of standard errors (columns N-S). There should be only two rows of data, one for the hardwood stand and one for the pine stand (rows 8 and 9).

Use the skills you learned previously to prepare two bar graphs, one for bacterial biomass and one for fungal biomass. Each graph should compare the three treatments (control, low N, high N) within each of the hardwood and pine stands. Change the scaling on the Y-axis to be the same for each graph so that you can easily compare the relative amount of bacterial and fungal biomass. To do this, double click on the Y-axis and select the "scale" tab. Input the minimum and maximum value for that axis.

### Synthesis Activity

Choose one of the following:

- a. Prepare a written report that synthesizes the information provided in the above datasets. The report should include an introduction, data summary, conclusion section, and references. The introduction should discuss the problem of N deposition in general terms, clearly state the objectives of the Chronic Nitrogen Amendment Study at Harvard Forest, and give the specific questions addressed by each of the datasets. The data summary should include the graphs that you prepared as well as a written summary of your interpretation of each graph. That is, describe in writing what each graph is depicting and the main conclusion to be drawn from each one. The conclusion section should summarize how chronic N additions have impacted the pine and hardwood stands at Harvard Forest.
- b. Prepare a 15-minute PowerPoint presentation (approximately 15 slides) that synthesizes the information provided in the above datasets. In the presentation, you should introduce the problem of N deposition, clearly state the objectives of the Chronic Nitrogen Amendment Study at Harvard Forest, cite the specific questions addressed by each of the datasets, interpret the graphs you prepared, and give the conclusions you have reached from the data. The last slide should provide a list of any reference materials that you used to prepare the presentation. You will give the presentation to your instructor at a mutually agreed upon time.

**References**

- Aber, J.D. (ed.). 2004. Special issue: the Harvard Forest (USA) nitrogen saturation experiment: results from the first 15 years. *Forest Ecology and Management* 196, 1-186.
- Bauer, G.A. F.A. Bazzaz, R. Minocha, S. Long, A. Magill, J. Aber, and G.M. Berntson. 2004. Effects of chronic nitrogen additions on tissue chemistry, photosynthetic capacity, and carbon sequestration potential of a red pine (*Pinus resinosa* Ait.) stand in the NE United States. *Forest Ecology and Management* 196, 173-186.
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## NOTES TO FACULTY

### Teaching Approaches

There are three subsections for each dataset: 1) Methods and rationale, 2) Open-ended questions, and 3) Detailed graphing instructions. The first subsection describes the main methods and why these measurements are of interest. The instructions for making figures are for Excel 2003.

For the second section, a set of open-ended questions are provided. These are designed to give students the opportunity to look at the data with questions in mind, as scientists do. Too often students blindly make graphs without thinking about the types of figures that would be most meaningful. Students could also examine the data and ask their own questions; you can use the ones provided to prompt further thought. In either case, you will likely need to keep asking "how can you use these data to get at that question?" and "what type of graph best presents the data to address that question"? After students have outlined the types of graphs they would like to make, you can provide the detailed instructions for making figures.

Students should work in groups. See "[cooperative groupwork](#)" links in the Teaching section. Beginning students will benefit from first sketching out ideas for figures; this way they will more carefully think about "what goes on what axis," for example. One challenge with this exercise is that students' abilities with Excel vary and therefore some will need more time than others to work with these data. You could have a separate Excel session, perhaps with teaching assistants if you have them, to bring everyone up to the same level.

The Student Instructions begin with a very basic overview; this is written for beginning ecology students. Before providing the specific graphing instructions you might suggest that students hand draw sketches of patterns they might expect to see in this experiment. This would be appropriate for students with little experience working with Excel figures. More advanced students should skip this step and the basic overview.

The time needed depends in part on student experience. Students in a plant ecology class could work with all four data sets in a lab session plus homework assignment. For beginning students, the first two data sets might be sufficient.

### The Data and Experimental Design

The Excel directions are for the 2003 version of the spreadsheet. The resource section below provides directions for older versions.

The data selected for this exercise illustrate how chronic N additions have impacted a range of ecosystem properties and processes, including both plant (tree productivity and photosynthetic capacity) and soil (microbial biomass and soil respiration) responses. There are four datasets with which students can work. Each dataset is located on an individual worksheet page within the Excel spreadsheet (see Datasets section). The faculty dataset includes the figures generated by following the detailed graphing instructions in the student section (see Student Instructions). While each dataset can be used as a stand-alone activity, the students will gain the greatest understanding of how excess N impacts forest ecosystems by working through the activities for all four datasets. The final activity (written report or PowerPoint presentation) is designed to help students synthesize all of the information presented in the datasets and to think more broadly about how northeastern forests are being impacted by N enrichment.

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Teaching Issues and Experiments in Ecology (TIEE) is a project of the Education and Human Resources Committee of the Ecological Society of America (<http://tiee.ecoed.net>).

This experiment provides a great opportunity to discuss the issue of replication and "pseudoreplication" in ecology. Over 20 years ago Hulbert (1984) published his well known paper titled "Pseudoreplication and the Design of Field Experiments" (*Ecological Monographs* 54: 187-211) which stimulated a good deal of discussion among ecologists. Central to the argument is the scale needed for replication — in the case of the Chronic N Amendment Study, you might ask your students whether taking measurements within one N treatment plot is sufficient or whether it is necessary to establish replicate plots in different forest stands. Again, the depth of discussion about this issue will depend on your time constraints and the experience of your students.

You may be surprised to learn this, but reasons for replication are a major conceptual challenge for many students; therefore discussion about this issue is especially important. The main points to bring out are: why replication is important, the relationship between reliability of conclusions and replication, why replication of large plots (30 x 30 m in this case) is not always feasible (e.g., cost and logistical constraints), and the design of this particular experiment and why scientists might disagree about replication in this case. The goal is not to criticize the Chronic N Addition researchers (some students who have not read the core papers will be very quick to do this!) but to use this dataset to stimulate this important discussion.

**Dataset 1:** contains average values for aboveground net primary productivity for 1988 through 2002 as  $\text{g m}^{-2}$ . NPP for the pine stand was highest in the control plot and decreased 31% and 54% relative to the control for the low N and high N plots, respectively. The hardwood stand responded differently with the fertilized plots showing higher NPP relative to the control.

**Dataset 2:** gives the average values and standard errors for the photosynthetic capacity of red pine needles collected from the control and high N plots. Photosynthetic capacity of needles from the high N treatment was significantly lower than that of the control trees.

**Dataset 3:** contains average values and standard errors for cumulative soil respiration ( $\text{g C m}^{-2}$ ) during the growing seasons of 1988, 1989, and 2001. In the hardwood stand, soil respiration was higher in the fertilized plots compared to the control. However, in the subsequent two years (1989 and 2001), respiration was lower in the high N plot relative to the low N and control plots. In the pine stand, soil respiration was lower in both fertilized plots for 1989 and 2001.

**Dataset 4:** presents the raw data (as opposed to averages as in the first three datasets) for bacterial and fungal biomass in soils collected in fall 2002. Students are first instructed to calculate the means and standard errors before preparing the graphs. This provides them with a better understanding of how data are typically analyzed. Fungal biomass was 27-61% and 42-69% lower in the fertilized compared to control plots in the hardwood and pine stands, respectively. Bacterial biomass was not greatly affected by N additions.

### Synthesis Activity

Students are given a choice of synthesizing the information in the four datasets either in a written report or by making a 15 minute PowerPoint presentation. Some students excel at writing, while others are more comfortable with oral communication. Both skills are of course important, but giving the students a choice will allow them to focus their attention on data comprehension and interpretation rather than on the presentation format. Choice also gives students a sense that they have some control over the evaluation process. Be clear at the outset as to how each of these activities will be evaluated. Below I provide one example of how

the written report can be evaluated. This can be easily modified for the PowerPoint presentation. For either, give students the criteria (rubric) ahead of time that you will use to judge their work. See also "rubrics" in the TIEE Glossary.

<b>The written report will be graded according to the following criteria:</b>	<b>Points</b>
Does the introduction present the problem and objectives clearly?	____(15)
Do the figures present the data clearly?	____(20)
Does the written portion of the results section accurately reflect the data as presented in the figures?	____(25)
Are the interpretations and overall conclusions supported by the data?	____(25)
Is the report well organized and well written?	____(15)
<b>TOTAL</b>	____(100)

### Student Evaluation

Students can be evaluated on a single dataset in terms of the questions they pose and figures they suggest, their understanding of the question being addressed, their ability to properly prepare (e.g., correctly labeled axes) and to accurately interpret the graphs, and their understanding of the main conclusions that can be drawn from that particular dataset. Evaluation can be based on either written or oral communication, or some combination of both. The written report or PowerPoint presentation (synthesis activity) will provide information on all of the above, in addition to evaluating the student's general understanding of N deposition, why the Chronic Nitrogen Addition Experiment was conducted, and his/her ability to synthesize several disparate pieces of information and to draw conclusions about how N enrichment has impacted two different forest stands at Harvard Forest. Students should be presented with the evaluation criteria at the time the assignment is made.

### Resources

#### Excel Tutorials

Please note — there are many versions of Excel; the sites below include tutorials for older versions.

- <http://www.usd.edu/trio/tut/excel/>  
Basic information: how to use functions, filling down, formatting, inserting, graphs and graphing; University of South Dakota
- <http://www.fgcu.edu/support/office2000/excel/>  
Basic — includes visuals; Florida Gulf Coast University
- <http://einstein.cs.uri.edu/tutorials/csc101/pc/excel97/excel.html>  
Has two windows — one with the tutorial and one with the workbook; University of Rhode Island
- <http://www.baycongroup.com/el0.htm>  
Introductory information; BayCon Group — commercial site
- <http://www.extension.iastate.edu/Pages/Excel/homepage.html>  
Basic information; University of Arizona