Ain’t No Mountain Pine Enough: A Case Study of How Mountain Pine Beetles are Affecting Ecosystem Processes

Chiara Forrester, Kendi F. Davies, Laura Dee, and Lisa Corwin

Department of Ecology and Evolutionary Biology, University of Colorado Boulder

Abstract
This case study engages students in generating hypotheses and predictions, analyzing and interpreting data, critically evaluating contrasting results in science, and thinking about feedbacks between biota and abiotic ecosystem dynamics. To address these areas, students use primary literature and simulated data about the interactions between mountain pine beetles and terrestrial carbon cycling. Mountain pine beetles are causing unprecedented mortality in forests in the mountain west, serving as a particularly relevant issue for our students in Colorado. The case is designed for three 50-minute class periods or two 75-minute class periods and should be implemented after students have learned the basics of terrestrial carbon cycling. During implementation of this case in a lower-division ecology course, we found that 1) content knowledge increased for most questions regarding student comprehension of carbon cycling, 2) students were better able to identify reasons for contrasting scientific results beyond scientist bias or human error, and 3) including a narrative in the assignment did not increase student perceptions of topic relevance.

Learning Goals
Students will:

• understand how biota can influence ecosystem processes and how subsequent feedbacks can occur.
• understand the mechanisms by which contrasting scientific results can occur.
• value the complexity of research results.

Learning Objectives
Students will be able to:

• describe fluxes of carbon and reservoirs of carbon in the terrestrial carbon cycle.
• describe ways in which biota influence the carbon cycle and vice versa.
• describe factors that influence Mountain Pine Beetle population dynamics.
• generate hypotheses and predictions about how Mountain Pine Beetle might impact the carbon cycle and design experiments to test their hypotheses.
• calculate effect sizes and standard deviations in R, generate figures in R, and interpret the results.
• evaluate the quality of data and experimental design.
• brainstorm experimental factors that contribute to contrasting scientific results.
• summarize complex and context-dependent results in a way that is communicable to public audiences.
INTRODUCTION

Case study teaching allows students to practice application of science skills to complex real-world problems (1). In addition, case studies that focus on socio-scientific issues are often more relevant to students, which can motivate them to engage more fully in their own learning (2,3,4). Case study teaching is also broadly recognized as having the potential to engage students in critiques and analyses that allow them to view an issue from multiple perspectives (4). Research about mountain pine beetle outbreaks is well-suited for case study teaching since it typically addresses complex biotic-abiotic interactions, must consider contextual factors that affect the beetles and trees in that area, and is constrained to and influenced by a specific context (e.g., a single forest or region). Additionally, research about mountain pine beetles often address climate change, a universally recognized socio-scientific issue. Lastly, mountain pine beetles are highly relevant to students in the American West, many of whom live in areas affected by beetle outbreaks. All of these benefits influenced our decision to write the case “Ain’t No Mountain Pine Enough.”

Mountain pine beetles affect large swaths of land across the mountain west, causing massive pine tree mortality. The beetles bore into the phloem of the tree, limiting the tree’s ability to transport photosynthates. If the tree is unable to fight them off with pitch (sap), the beetles lay eggs under pine tree bark and infect the sapwood with a blue stain fungus that restricts the tree’s ability to fight off the beetle with pitch flow and alters water flow throughout the tree. The combination of these effects kills an infected tree (5). While these insects are native to the mountain west, longer growing seasons have allowed them to complete more life cycles each year, causing population surges with dramatic consequences for forests. Additionally, drought and elevated temperature cause increased stress in trees, which impedes their ability to resist outbreaks of pine beetles. Thus, outbreaks are projected to continue to affect an increasing number of forests as the climate changes (6).

Changes in forest structure and function can affect overall carbon cycling, in some cases causing forests and their associated soils to become carbon sinks rather than sinks due to decreased photosynthesis as a result of killed trees (7). It has been posited that mountain pine beetle outbreaks may cause a positive feedback loop; they may increase atmospheric CO₂ by transforming affected forests into carbon sources. This, in turn, would raise temperatures and facilitate increased beetle population sizes and higher drought stress for trees (7). Thus, mountain pine beetle outbreaks offer a useful system to introduce undergraduate students to biotic influences on terrestrial carbon cycling, positive feedback loops, and climate change, all important topics in ecosystem ecology.

A main goal of this case study is to provide students an opportunity to critically evaluate primary literature and to think about how variation in scientific findings can occur. The pine beetle system enables this because, while it is established that mountain pine beetle outbreaks cause widespread tree mortality and thus lower primary production, the degree to which mountain pine beetles cause positive feedbacks with climate change is debated in the literature (7,8). To this effect, we hypothesized that participating in this case study would increase students’ ability to generate content-specific reasons why two studies could have different results, as opposed to citing general issues such as “one scientist was wrong” or “one of the scientists was biased.” In addition to providing an ideal case study of important concepts in ecosystem ecology, we used this topic due to its impacts on areas local to our university. We hypothesized that using a local issue would increase students’ perceptions of topic relevance, and thus increase engagement. In addition to using a local issue, we sought to test whether including a narrative storyline in the case study materials would make students feel that the topic is more relevant to them and/or their communities. To do this, we taught this case study both with and without a narrative storyline (in which fictional characters guided students through the case study) in two different sections of the course and asked each group questions about their perception of topic relevance.

We find that this ecosystem ecology case study increases student knowledge of biotic effects on terrestrial carbon cycling and improves students’ ability to critically evaluate discrepancies between results coming from primary literature. Further, we find that including a storyline in case study narratives did not increase student perception of topic relevance and may make students feel resistant to case study material.

Intended Audience

We designed this lesson for lower-division undergraduate students in an Ecology course at a large research university, but it would suit many environmental studies courses well. In this course, students learn about the basic principles of ecology, spanning from ecophysiology through landscape dynamics. The majority of students are ecology or environmental studies majors, and many intend to pursue jobs in environmental science and policy or with a biological focus. We taught this course to lecture sections of 90-150 students. One instructor and 1-2 learning assistants were adequate instructional staff for parts 1 and 3 of this case study, but using 2-3 learning assistants for part 2 (in which students analyze data in R) would be ideal.

Required Learning Time

We taught this lesson in three 50-minute sessions (150 minutes total) and also in two 75-minute sessions (150 minutes total) over the course of one week. Students were required to complete ~1 hour of work outside of class. For ease of understanding, we have described the case study as though it is being taught over three class sessions. However, we have also included in the supporting materials teaching notes and PowerPoint slides for teaching it in two 75-minute sessions (Supporting File S12. Pine Beetle Case Study – Additional Teaching Notes; Supporting File S14. Pine Beetle Case Study – PowerPoint Slides for Two 75-Minute Classes).

Prerequisite Student Knowledge

Students should have had a brief introduction to the terrestrial carbon cycle and should be able to name processes of carbon flux (i.e., photosynthesis, respiration and decomposition) and carbon sinks. Students should have exposure to the scientific process used by biologists, for example making predictions based on evidence, testing those predictions using experimental approaches, and modifying one’s understanding based on evidence. Further, they should have familiarity with the concept of null and alternative hypotheses. Previous exposure to the open-source statistical program R is highly recommended, specifically the ability to import a script and .csv file into R Studio and run a line of code.
Prerequisite Teacher Knowledge

Instructors should have basic skills in R (e.g., importing datasets, generating and exporting simple figures like bar graphs, running simple statistical analyses like T-tests, and light trouble-shooting). This can be obtained by carefully reading and executing the annotated R script provided. Additionally, instructors should be able to describe the terrestrial carbon cycle, have a basic knowledge of community and ecosystem dynamics, and be able to critically evaluate primary literature articles. Instructors should familiarize themselves with the pine-beetle life cycle and how it relates to tree infection and mortality. We suggest that instructors read both the Kurz and colleagues (2008) and Moore and colleagues (2013) publications in their entirety to be able to answer student questions that may arise (7,8). Instructors do not need in-depth knowledge of insect, plant, or fungal biology to teach this lesson.

SCIENTIFIC TEACHING THEMES

Active Learning

We presented new information to students in an interrupted lecture format (lecture with breaks for active work) in which students frequently engaged in think-pair-shares, followed by whole-class questions and discussions. A majority of time was spent honing student skills with in-class analysis and graphing in the program R and in-class worksheets completed in pairs and/or groups. We reinforced concepts periodically using whole-class discussion and by asking for volunteer groups to report on their findings/interpretation.

Assessment

We assessed students’ skills in R with in-class assignments, wherein students analyzed imitated data (not real data, but showing the same trends as published papers when analyzed) and interpreted the results. In-class assignments also assessed students’ ability to generate hypotheses, predictions, and prediction graphs. These in-class assignments are integral to the case study and should always be used when teaching it.

Additionally, we assessed content knowledge and students’ ability to generate content-specific reasons for variable scientific results with pre-, during- and post-class multiple-choice questions. Pre- and post-class questions were the same and tested students’ knowledge of terrestrial carbon cycling, how biota interact with carbon cycling, and how discrepancies in scientific results can occur (Supporting Files S10. Pine Beetle Case Study – Pre-Assessment; S11. Pine Beetle Case Study – Post-Assessment). These assignments are not required for teaching this case study, as they serve solely to provide information about student learning gains. We note that some of the questions require interpretation of responses that are open-ended, which may not be possible for every instructor due to time constraints.

Finally, to address whether including a narrative increased relevance, we used a post-class open ended question in two paired offerings of the course (Supporting File S11. Pine Beetle Case Study – Post-Assessment). We do not recommend that instructors consistently use this assessment. Rather, this one-time question allowed us to further refine the case for future use.

Inclusive Teaching

We included ample time for completion of worksheets, in and out of class. By pairing students together, we hoped to decrease student anxiety while learning to code in R. We encouraged collaboration so that students may assist each other with data analysis and interpretation, explain their thinking, and modify their understanding based on peer feedback – this also worked with screensharing in an online teaching format. Grading rubrics emphasized general content knowledge and critical thinking skills over rote memorization and helped to ensure impartial grading. During the coding portion of the case study, we provided laptops for students who needed them.

LESSON PLAN

This case study consists of three sections: 1) an introduction to how mountain pine beetles might affect the terrestrial carbon cycle and vice versa; 2) data analysis in R of a simulated data set modelled on results from the primary literature and interpretation of the results; and 3) critical evaluation through comparing and contrasting summaries of two primary literature papers and then synthesizing the results for public communication. We distributed two versions of this case study, one with a storyline included in the case study narrative (students were asked to walk through the case with three fictional characters Riley, Eliyah, and Eddie – Supporting File S13. Pine Beetle Case Study – Narrative with Storyline), and one without (students were presented with data and problems without the fictional characters’ stories – Supporting File S5. Pine Beetle Case Study – Narrative). Because we did not find that including the storyline positively affected student content knowledge or perception of topic relevance, and instead received informal negative feedback about storyline inclusion, we focus here on the lesson plan and materials that do not include the storyline. Lastly, a third instructor also taught this lesson plan on their own, and we incorporated some of their suggestions for improvements.

We taught this case study to several classes across three years in a lower-division undergraduate Ecology course at a large research university with ~130 students per section. Results of pre- and post-assessments reported here are from two sections of the same Ecology course, one taught with the storyline narrative format and one without the storyline narrative. Each of these sections were taught in the same semester and by the same instructor.

Instructor Preparation

Instructors should review the detailed timeline of this lesson (Table 1) and print it for reference. To ensure all students have the necessary materials, instructors should 1) post all case study materials to the online class hub, and/or print out case study materials (one case study packet should be prepared for each student), 2) assign students the pre-class assignment before the first class, and 3) prepare optional pre- and post-assessments in your learning system (e.g., Qualtrics or Canvas Quizzes) if desired. We also recommend that instructors download R (which is free and open source) and practice running through the R script with the data provided. Instructors should read both the Kurz and colleagues (2008) and Moore and colleagues (2013) publications in order to field all student questions that may arise (7,8). If possible, instructors should provide laptops for students who need one during the R analysis portion of this case study.

Pre-Class Carbon Cycle Assignment

A video with accompanying questions guides students through learning about processes of carbon movement and locations of
carbon reservoirs in terrestrial ecosystems. Further, it introduces students to thinking about how biota can influence the terrestrial carbon cycle and vice versa (including positive feedback loops). Students also watch a second video that shows a local researcher studying mountain pine beetle outbreaks in Colorado (Supporting File S3. Pine Beetle Casestudy – Pre-Class Assignment). This second video introduces the pine beetle life cycle, how they impact pine trees and some hypotheses about why pine beetle outbreaks are becoming more extreme with climate change.

Part 1: A Harrowing Hike

Introduce casestudy learning objectives

Using slide 1 from the PowerPoint slides provided (Supporting File S4. Pine Beetle Casestudy – PowerPoint Slides), give a minilecture describing the learning objectives for this casestudy, with the goal of priming students for recognizing and reflecting on what they are learning.

Page 1 of narrative: What killed the trees?

Begin by showing students images of a forest with extensive beetle-kill using the included slides (Supporting File S4. Pine Beetle Casestudy – PowerPoint Slides, photos taken by Dr. Jeff Mitton and reprinted with written permission). Ask them to come up with ideas for what caused the disturbance, and brainstorm on the first page of the case study narrative (Supporting File S5. Pine Beetle Casestudy – Narrative, photo taken by Dr. Jeff Mitton and reprinted with written permission). Then, ask whether they have seen beetle-kill, and what their initial reaction to seeing that forest was. This process helps you to identify the level of student background knowledge in the class and determine how much time to spend describing the pine beetle life cycle. Therefore, be sure to engage many students in the discussion and check-in with all students about their knowledge. For example, you might ask students to raise their hand if they are familiar with pine beetles and the blue stain fungus. Next, depending on students’ level of background knowledge, spend time discussing the beetle’s life cycle using Slide 4 of Supporting File S4. Pine Beetle Casestudy – PowerPoint Slides (figure created by the authors with photos used and reprinted with written permission from Dr. Jeff Mitton). We also recommend posting a fact sheet on an online class portal about the mountain pine beetle (e.g., https://extension.colostate.edu/docs/pubs/insect/05528.pdf) for students to reference as desired throughout the casestudy.

Page 2 of narrative: What abiotic factors affect mountain pine beetle populations?

Students then work in pairs through questions on the second page of the case study narrative (Supporting File S5. Pine Beetle Casestudy – Narrative), proposing abiotic factors that could influence the population dynamics of the Mountain Pine Beetle. Walk around the room and answer questions. This provides students an opportunity to reinforce their knowledge of what causes populations to grow or decline and have opportunities to review elements of community dynamics.

Class discussion 1: Mountain pine beetle interactions with environmental conditions

This whole-class discussion allows you to hear what students have discussed in response to your question regarding factors that influence population growth. During the discussion, highlight particularly interesting or relevant ideas students had by repeating them to the class. Students should respond that longer summers allow the beetles to go through more generations in a year, and that drought and high densities of trees (due to longer durations between fires because of management) make it more difficult for trees to defend themselves. Increases in winter temperature and drought exacerbate these effects.

Questions to pose to students:

- What conditions might support the success of pine beetle populations?
- How might changes in those conditions cause larger outbreaks of this beetle?

Pages 3 and 4 of narrative: Mountain pine beetle interactions with the carbon cycle

During this section, the narrative will guide students in the process of posing a hypothesis for one aspect of how beetles may influence forest carbon cycling or vice versa and predicting what they would find if they tested their hypothesis. During this process, the instructor should help students distinguish predictions from hypotheses. Hypotheses are often general statements based on existing scientific knowledge. A hypothesis must include a mechanism explaining the phenomena and a direction of the hypothesized effect. Alternatively, a prediction includes specific, measurable variables and a direction of the effect. Predictions are often more specific and refer to what you would measure and expect when performing an actual experiment.

Some examples of potential hypotheses include:

- Beetles will increase the release of C from forests due to increases in microbial respiration/decomposition (using beetle-killed trees as substrate).
- Beetles will increase the net release of C from forests due to less conversion of CO2 because there are fewer trees conducting photosynthesis.
- Beetle-killed will decrease the release of C from forests after a short-term increase due to less biomass being produced/decomposed.

In addition to practicing writing hypotheses, this activity is meant to help students think about biotic interactions with abiotic processes and to begin laying the foundation for discussions of positive feedback loops.

Class discussion 2: Hypotheses for pine beetle interactions with the carbon cycle

This discussion serves as a way for students to hear other ideas besides their own, and allows the instructor to interrupt any potentially misleading hypotheses and correct students’ misconceptions. During the discussion, the instructor can highlight particularly interesting or relevant student ideas by repeating them to the class and asking follow-up questions or elaborating as needed.

Question to pose to students:

- What was your hypothesis for one way the mountain pine beetle could impact Carbon cycling?

Part Two: A Mountain of Data

When taught in three 50-minute sections, this section occurs at the start of the second class-section. When taught in two
75-minute sections, students go through the first part of this section until immediately after the analyses in R (Supporting File S12. Pine Beetle Casestudy – Additional Teaching Notes).

**Mini-lecture, slides 10-12: Learning objectives and carbon terms**

Using the PowerPoint slides provided (Supporting File S4. Pine Beetle Casestudy – PowerPoint Slides), the instructor can re-visit the list of learning objectives, describe how many of them were met in the first class, and highlight the objectives (related to data analysis and interpretation) that will be addressed in the second class. The short lecture provided describes the carbon terms that students will find in the simulated dataset as well as in the homework readings. We also recommend discussing effect sizes and T-tests, as described in the annotations on the R script (Supporting File S6. Pine Beetle Casestudy – R Script).

**Pages 5-6 of narrative: Data analysis of simulated dataset**

After the mini-lecture, students run through the R script (Supporting File S6. Pine Beetle Casestudy – R Script) using data mimicking Moore and colleagues’ study (Supporting File S7. Pine Beetle Casestudy – Dataset), interpret T-tests and linear regressions, and produce figures. This simulated dataset includes data from two sites; students will analyze only data from Fraser Experimental Forest. The data are from replicate study plots in a) “beetle-killed” stands that have been impacted by the mountain pine beetle and b) “control” stands that have not been impacted. Response variables measured in each plot include soil respiration, litter depth, organic soil horizon depth, net ecosystem exchange and gross primary production. While we focus on comparisons between control and “beetle-killed” plots in this case, more analyses could be done using this dataset in advanced classes (see Possible Improvements and Adaptations).

In classes where students are less familiar with R, the instructor can display their computer screen on the projector while opening the script in R, importing the data, running the first statistical analysis, and encouraging students to follow along. We recommend that instructors remind students not to run the entire script at once, but instead to read the annotations and go line-by-line. This is important because there is one line of code that students need to edit, which enables them to practice writing a line of code in a scaffolded way (i.e., rather than generating new code, they edit existing code). Throughout this section, the instructor can answer questions and repeat common questions to the whole class. Students should write down the effect sizes and standard deviations and export their figures for reference as they run through the code. This is especially important if the case study is taught in two 75-minute classes because students write a summary of their results in the following class and will need the figures and statistics for reference. If teaching the case in two 75-minute classes, end here and resume this section at the start of the second class. If teaching it in three 50-minute classes, finish the entirety of this section. During this component of the activity, it is particularly helpful to have assistants who can help students run the R script, allowing troubleshooting and quicker progress. If this is not possible, we recommend walking through example segments of the code with the whole class.

After completing the R script, students write a summary of the results they found. Students should have found decreases in respiration, gross primary production and net ecosystem exchange, along with increases in litter and organic soil horizon depth. These results show that beetles do not necessarily increase the chance that a forest infested by beetles would become a C source due to lowered respiration. Students then diagram the C cycle again and add in any impacts of the beetles they found in their results.

**Class discussion 3 (think-pair-share): Review findings**

Instructors can show the slide (Supporting File S4. Pine Beetle Casestudy – PowerPoint Slides) with discussion questions and have students engage in a think-pair-share with a partner. A think-pair-share allows students a few moments (e.g., 30 seconds) to think quietly and then discuss their thoughts. Lastly, the instructor can ask some pairs to share the highlights of their discussion with the whole class.

**Questions to pose to students:**

- Did the results of your data analysis surprise you?
- How might these alterations in C cycling affect beetle population dynamics?

Students are often surprised that respiration decreased, because many of them predicted increases in respiration. Asking whether the results of their analysis surprised them causes them to reflect on their original hypotheses and start brainstorming why a decrease may have occurred (e.g., due to long-term decrease in substrate for respiration over time). The second question introduces the concept of positive feedback loops between mountain pine beetle population growth and climate change by asking students how the effects of mountain pine beetle populations on forest carbon cycling (e.g., respiration) could then feedback to affect future beetle population dynamics.

**Homework: Paper Summaries**

For homework, students should read one of two primary literature paper summaries (Supporting Files S8. Pine Beetle Casestudy – Kurz Paper Summary; S9. Pine Beetle Casestudy – Moore Paper Summary) that both investigate the impact of beetle infestation on various aspects of C cycling. One of these summaries supports the results they found while conducting their analyses, while the other does not. The purpose of this is to introduce to students one aspect of scientific uncertainty; different studies can find different results even when they are both well executed. The rest of the case study is designed to use the contradictory findings to foster discussion of how and why these two studies on the effects of pine beetles on the C cycle reach different conclusions.

**Part 3: Pining After the Truth**

If teaching this case study in three 50-minute classes, plan to complete this section in the third class. If teaching this case study in two 75-minute classes, start this section in the second class after you complete Part 2.

**Mini-lecture, Slides 17-18: Review carbon terms**

Before delving into part 3 of the case study, the instructor can use the PowerPoint slides provided (Supporting File S4. Pine Beetle Casestudy – PowerPoint Slides) to review the carbon terms used in the paper summaries.

**Pages 7-8 of case study narrative: Understanding scientific complexity**

During the rest of this section, students mostly conduct work with a partner and with a group. During this section, the
instructor can interact with students, asking and answering questions and repeating common or especially interesting thoughts to the rest of the class.

First, students discuss the paper they read with a partner. They will then answer questions about the study design and assess the quality of the dataset. Next, students find a group that discussed the other paper and work through the second set of questions to compare the two studies. They will work to understand how the results are different and why. The papers share similar results except for the response of respiration: Moore et al. found a decrease in respiration with beetle-kill while Kurz et al. found an increase. This difference led them to draw different conclusions about whether there is a positive feedback loop between mountain pine beetles and climate change. Key differences include that:

- Kurz et al. found that beetle-caused tree death led to an increase in respiration because there was more dead material available to decomposers when compared to forests where no beetle kill occurred. Because live trees remove carbon from the atmosphere, Kurz et al. posit that the loss of those trees, combined with an increase in respiration, will mean that the forest will not act as a C sink. On a large spatial scale, they argue that this could feed back to climate change.
- After a similar initial spike in substrate for decomposition, and thus respiration, Moore et al. found a decrease in respiration after multiple years, as that material was decomposed. Moore et al. posit that after this long-term decrease in respiration after dead material is decomposed, the loss of trees alone will not be a mechanism for the forest to become a major carbon source.

Because the articles do not directly compare their findings, we cannot say with certainty what caused the differences in their conclusions. We can, however, help students to think about what may have caused the differences, allowing them to practice critically evaluating scientific literature: temporal scale, spatial scale, and method of measurement. Moore et al. used a longer time scale for observations than Kurz et al., which may have allowed their study to include the point at which respiration was no longer increased by the dead trees. Kurz et al. modeled pine beetle impacts over a larger spatial scale, which may have shown more dramatic effects than when looking at pine beetle kill in one forest. Lastly, the differences in their conclusions may have occurred because they measured different types of respiration (Kurz et al. modeled overall forest respiration while Moore measured soil respiration) or because Kurz et al. conducted a modelling study while Moore conducted an experimental study in one forest. At this stage, the instructor can encourage the students to discuss in small groups about what could lead to these different results and thus conclusions (prior to the class discussion 4). This section is the most difficult for students – to facilitate this learning, interacting with students is key. Repeat common themes and questions to the whole class. The instructor can also ask prompting questions to the whole class, such as:

- What were the major differences in their experimental design?
- Did they measure respiration in the same way?

Next, students come up with an experimental design that would reconcile the differences in these two papers. This is a difficult question for students, and so we recommend doing the following in class, but also allowing them 3 days after the completion of the case study to turn in their answers. Guide students in this process, beginning by asking what the main difference was between the two studies and what you would need to know to understand how that difference occurred more thoroughly. For example, one could investigate whether total ecosystem respiration and soil respiration respond in the same way to beetle-kill if measured in tandem at the same site. This question acts as a way for students to address any gaps in the research or discrepancies in the methods. Finally, students come up with a Tweet to practice summarizing the results of both papers together. This reconciliation activity is designed to help students critically evaluate how multiple, sometimes contrasting, pieces of evidence form a larger scientific narrative that enhances our understanding of complex systems. Further, through this exercise we hope that students practice communicating science to non-scientists, and gain appreciation for the many factors that need to go into recommendations for a given management or action plan.

Mini-lecture, slides 20-21: Review paper summaries
After students work independently through pages 7-8 of the narrative, quickly review each paper summary. This is to ensure that they are practicing interpreting information from scientific text, while also giving them the opportunity to correct their responses if they misunderstood the text or needed more time to thoughtfully complete answers. We recommend giving students 3 days after the final class to turn-in the case study.

Class discussion 4: Differences between Kurz et al. and Moore et al.
For the first question below, the instructor can foster a whole-class discussion of the difference in the response of respiration in the two studies, and how this affected the broad conclusions about feedback loop potential each paper made.

Questions to pose to students:

- What was the most important difference in their results?
- Which paper was more convincing to you? Why?

Mini-lecture, slide 23: Why did respiration differ? How did that affect the scientists’ broad conclusions?
This section is to ensure that students have correctly identified the differences between the two studies they read summaries of, and to provide them with a brief description of the mechanisms behind why differences could have occurred in the study conclusions to minimize confusion. See description of “Pages 7-8 of Case Study Narrative” above for more information and follow the slide to discuss (view slide 23 in presenter mode and advance animations in order to see all slide content).

TEACHING DISCUSSION
Through this case study, we taught important concepts in ecosystem ecology by using an active-learning format and a real-world issue that is local to our students. We found after students took part in this case study that 1) their content knowledge increased for most questions regarding student comprehension of carbon cycling, 2) they were better able to identify reasons for contrasting scientific results beyond scientist bias or human error, and that 3) including a narrative in the assignment did not increase student perceptions of topic relevance.
Changes in Student Content Knowledge

In order to address our teaching of the Vision and Change Core Concept, Pathways and Transformations of Energy and Matter (10), we assessed student content knowledge of how biota interact with carbon cycling at the beginning and end of the semester through pre-assessments (n=206) and post-assessments (n=195) (see Supporting Files S10. Pine Beetle Casestudy – Pre-Assessment and S11. Pine Beetle Casestudy – Post-Assessment, and Table 2 for all results). Specifically, we asked students 1) how biota can indirectly remove carbon from the atmosphere, 2) how biota can directly release carbon from the atmosphere, 3) the possible fate of a carbon atom from the trunk of a tree that has died, and 4) to justify their response for potential fate a carbon atom from the trunk of a tree that has died. When asked to provide mechanisms for how “biota can indirectly release carbon to the atmosphere (an animal respiring does not count),” student scores significantly improved from an average score of 49% in the pre-assessment to 63% in the post-assessment (t=2.56, P=0.01). Notably, this question had the lowest average initial score of the content questions and showed the most improvement. While there were no statistically significant increases in student scores for the other questions listed above, we found that their content knowledge improved for most questions, demonstrating an overall increased understanding of biotic controls on terrestrial carbon cycling (see Table 2 for all results).

Changes in Student Ability to Critically Evaluate Discrepancies in Research

Despite uncertainty and complexity being common in scientific endeavors, students rarely get experience dealing with these in traditional “cook-book labs” or lectures. Therefore, one of our goals through this case study was to give students more exposure to scientific uncertainty and complexity in results. Specifically, we wanted to know if students’ understanding of how differences in research findings occur could be improved as a result of participation in the case study. To test this, students were given the following question before and after the case study:

Two scientists studied the same ecological question. Both conducted their studies in the field with the same treatment and control and measured the same independent and dependent variables. One scientist found that the dependent variable increased in response to the treatment, but the other found that it decreased. What are possible reasons why this difference in the results occurred?

Open-ended responses to this question were coded through the identification of common response types, and a chi-squared test was used to analyze differences in response-type (code) frequency before and after the semester (see Table 3). The frequency of our various codes was significantly different between pre- and post-assessments (X-squared = 65.77, P<0.0001, df=7). The frequency of responses in which students cited “confounding factors,” “scientist bias,” and “scientist error” decreased after the case study (with 18, 6, and 27 fewer instances respectively). Notably, all of these responses placed the onus for different results solely on the scientist, implying that the scientists’ bias, error, or improper design of the experiment resulted in different results. None of these responses considered how different study approaches or different contexts might impact results. Alternatively, responses which cited “season or duration of study” and “spatial study” increased after the case study (with 39 and 21 more instances respectively). These codes reflect that students have changed their understanding to include more reasons why scientists’ results may differ.

Importance of a Narrative Storyline in Student Perception of Topic Relevancy

We hypothesized that including a narrative storyline (i.e., worksheets that guide students through the material using fictional characters and a storyline) would increase student perception of topic relevancy by making students connect the science content to human communities more directly. To test this, the same instructor taught two sections of the same general ecology course, using materials with a narrative storyline in one and without the narrative storyline in the other. After the completion of the case study, students in each section were asked, “In what ways did the structure of this case study convince you (or not convince you) that pine beetle kill is relevant (or not relevant) to you and your community?” Open-ended responses to this question were coded through the identification of common response types, and a chi-squared test was used to analyze differences in the proportion of students out of the class who responded with a given response-type (code) (see Table 4).

The proportion of student responses in each category was not different between students who had materials with a narrative storyline and those without (X-squared = 0.16, P=1, df=10). Thus, we found that including a narrative storyline in student learning materials did not increase student perception of relevancy. Likewise, we received informal feedback from students each semester that they did not like having a narrative storyline included. Reasons provided included that they felt as if they were being treated like younger students (descended to), and that it did not seem helpful enough to outweigh the extra time the materials took students to read. Thus, for the purposes of this case study we have presented materials without the narrative storyline (Supporting File S5. Pine Beetle Casestudy – Narrative) but included the narrative with the storyline in the supplementary materials (Supporting File S13. Pine Beetle Casestudy – Narrative with Storyline).

Possible Student Outcomes Not Directly Tested

This case study used the societally important issue of mountain pine beetle kill to provide students with exposure to experimentation (reading the Moore et al. summary and generating their own experimental ideas), model generation (by reading the Kurz. et al. summary), hypothesis testing (questions in the case study narrative), the use of quantitative skills in biology (using R to conduct statistical analyses), and communication to broad audiences about complex topics (writing plain language summaries and a Tweet). By addressing these topics, we feel this case study addresses multiple core competencies outlined by Vision and Change (10):
Ability to apply the process of science  
Ability to use quantitative reasoning  
Ability to use modeling and simulation  
Ability to communicate and collaborate with other disciplines  
Ability to understand the relationship between science and society  

While we did not directly test these learning outcomes with pre- and post-assessments, future assessments conducted alongside this case study could focus on these outcomes that apply to a wide variety of systems and fields.

Possible Improvements and Adaptations  
Some students expressed feeling rushed during this case study. We would recommend allowing as much time as possible for students to finish assignments outside of class (we typically allowed 3 days after the conclusion of the case study). Further, because coding in a large group setting can be difficult, we recommend walking through opening the R script, importing data, and performing the first statistical analysis in front of students so they can follow along. Instructors should point out how to find the variable names for later editing (lines 10-13 in the R script), and clearly notify students when there is a line of code that they must edit in order for the R script to run successfully. Ideally, students should be provided with supervised experience in R prior to the case study. The R101 tutorial found at the following URL covers the skills necessary for this case study and is also appropriate for introductory classes that use R in general: https://chiaraforrester.weebly.com/r-tutorials.html. For more advanced classes, students could be asked how they would analyze the datasets in this case study and write the code themselves. Lastly, depending on the instructor’s course goals, students could test a greater number of hypotheses by incorporating the full dataset (e.g., students could compare results from the experiment involving girdled trees to the beetle-killed trees in the second research site). While we used R in our teaching of this content, instructors that do not use R or wish to teach students a different program could use Excel or Google Sheets with their students to conduct analyses and generate figures. Numerous tutorials and resources are available to help instructors adapt this to Excel or Google Sheets if desired (e.g., https://www.excel-easy.com/examples/t-test.html; https://www.excel-easy.com/examples/regression.html).

SUPPORTING MATERIALS  
• S1. Pine Beetle Case Study – Student Rubric.  
• S2. Pine Beetle Case Study – Grader Rubric.  
• S3. Pine Beetle Case Study – Pre-Class Assignment.  
• S4. Pine Beetle Case Study – PowerPoint Slides.  
• S5. Pine Beetle Case Study – Narrative.  
• S6. Pine Beetle Case Study – Dataset.  
• S7. Pine Beetle Case Study – R Script.  
• S8. Pine Beetle Case Study – Kurz Paper Summary.  
• S9. Pine Beetle Case Study – Moore Paper Summary.  
• S10. Pine Beetle Case Study – Pre-Assessment.  
• S11. Pine Beetle Case Study – Post-Assessment.  
• S12. Pine Beetle Case Study – Additional Teaching Notes.  
• S13. Pine Beetle Case Study – Narrative with Storyline.  
• S14. Pine Beetle Case Study – PowerPoint Slides for Two 75-Minute Classes.

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Table 1. Detailed recommended timeline for this lesson, over multiple class periods and including homework assignments.

| Activity                          | Description                                                                 | Estimated Time                  |
|-----------------------------------|-----------------------------------------------------------------------------|---------------------------------|
| **Preparation for Class**         |                                                                             |                                 |
| Print out case study narrative    | 1. Make one copy of case study narrative for each student.                  | ~30 minutes for 120 students    |
| and post class materials online   | 2. Post student rubric, pre-class assignment, paper summaries, R script, and dataset online for students. |                                 |
| for students                      |                                                                             |                                 |
| Supporting Files:                 |                                                                             |                                 |
| S1. Pine Beetle Case Study –      |                                                                             |                                 |
| Student Rubric;                   |                                                                             |                                 |
| S3. Pine Beetle Case Study –      |                                                                             |                                 |
| Pre-Class Assignment;             |                                                                             |                                 |
| S6. Pine Beetle Case Study – R    |                                                                             |                                 |
| Script;                           |                                                                             |                                 |
| S7. Pine Beetle Case Study –      |                                                                             |                                 |
| Dataset;                          |                                                                             |                                 |
| S8. Pine Beetle Case Study –      |                                                                             |                                 |
| Kurz Paper Summary;               |                                                                             |                                 |
| S9. Pine Beetle Case Study –      |                                                                             |                                 |
| Moore Paper Summary.              |                                                                             |                                 |
| **Pre-Class Assignment**          |                                                                             |                                 |
| Pre-class assignment.             | Watch two videos on the 1) terrestrial carbon cycle and 2) mountain pine beetles, and answer questions. | ~20 minutes                     |
| S3. Pine Beetle Case Study – Pre-Class Assignment. |                                                                             |                                 |
| **Class One – Part One: A Harrowing Hike** |                                                                             |                                 |
| Introduce case study learning     | Review bulleted list of case study learning objectives for students.        | 3 minutes                       |
| objectives.                       |                                                                             |                                 |
| Slide 2 in S4. Pine Beetle Case   |                                                                             |                                 |
| Study – PowerPoint Slides.        |                                                                             |                                 |
| Students work on Page 1 of S5.    | Students brainstorm what they already know about mountain pine beetles.      | 1 minute                        |
| Pine Beetle Case Study – Narrative.|                                                                             |                                 |
| Slide 3 in S4. Pine Beetle Case   |                                                                             |                                 |
| Study – PowerPoint Slides.        |                                                                             |                                 |
| Students work on page 2 of S5.    | Students generate hypotheses about abiotic effects on mountain pine beetle populations. | 10 minutes                      |
| Pine Beetle Case Study – Narrative.|                                                                             |                                 |
| Slide 4 in S4. Pine Beetle Case   |                                                                             |                                 |
| Study – PowerPoint Slides.        |                                                                             |                                 |
| Class discussion.                 | Whole class discussion 1: Students share-out hypotheses for how abiotic conditions affect mountain pine beetles. | 4 minutes                       |
| Slide 5 in S4. Pine Beetle Case   |                                                                             |                                 |
| Study – PowerPoint Slides.        |                                                                             |                                 |
| Students work on pages 3-4 of S5. | Students diagram the terrestrial carbon cycle, generate hypotheses for how mountain pine beetles could alter each process (flux) step of the carbon cycle, and draw prediction graphs. | 25 minutes                      |
| Pine Beetle Case Study – Narrative.|                                                                             |                                 |
| Slide 6 in S4. Pine Beetle Case   |                                                                             |                                 |
| Study – PowerPoint Slides.        |                                                                             |                                 |
| Class discussion.                 | Whole class discussion 2: Students share out hypotheses for how mountain pine beetles could affect carbon cycling. | 4 minutes                       |
| Slide 7 in S4. Pine Beetle Case   |                                                                             |                                 |
| Study – PowerPoint Slides.        |                                                                             |                                 |
| Class close.                      | Give students homework information, any other announcements.                | 3 minutes                       |
| Slide 8 in S4. Pine Beetle Case   |                                                                             |                                 |
| Study – PowerPoint Slides.        |                                                                             |                                 |
| **Class Two – Part Two: A Mountain of Data** |                                                                             |                                 |
| Class opening.                    | Slide gives students information about laptops and working in pairs.        | N/A                             |
| Slide 9 in S4. Pine Beetle Case   |                                                                             |                                 |
| Study – PowerPoint Slides.        |                                                                             |                                 |
| Mini-lecture.                     | 1) Revisit learning goals: describe goals met in first class and discuss goals for this second class. 2) Introduce important carbon terms. | 13 minutes                      |
| Slides 10-12 in S4. Pine Beetle   |                                                                             |                                 |
| Case Study – PowerPoint Slides.   |                                                                             |                                 |
| Students work on pages 5-6 of S5.| Students work through R script, analyzing simulated data set on how mountain pine beetles affect forest carbon cycling, interpreting the results, and comparing to initial hypotheses/predictions. | 30 minutes                      |
| Pine Beetle Case Study – Narrative.|                                                                             |                                 |
| Slide 13 in S4. Pine Beetle Case  |                                                                             |                                 |
| Study – PowerPoint Slides.        |                                                                             |                                 |
## Activity

| Activity                                      | Description                                                                                                                                                                                                 | Estimated Time |
|----------------------------------------------|-------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|----------------|
| Class discussion.                            | Think-pair-share – Whole class discussion 3: Students discuss whether results were surprising to them and discuss what their findings mean for positive feedback loops to climate warming.                                     | 5 minutes      |
| Class close.                                 | Give students homework information, any other announcements.                                                                                                                                                  | 2 minutes      |

### Homework Between Classes Two and Three

| Activity                                      | Description                                                                                                                                                                                                 | Estimated Time |
|----------------------------------------------|-------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|----------------|
| Read summaries of primary literature article | Each student should read either the Kurz or Moore paper summary – have student pairs/groups decide who reads which in class two so that each group consists of students who read both. | ~20 minutes    |

### Class Three – Part Three: Pining After the Truth

| Activity                                      | Description                                                                                                                                                                                                 | Estimated Time |
|----------------------------------------------|-------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|----------------|
| Mini-lecture.                                | Review important carbon terms.                                                                                                                                                                               | 4 minutes      |
| Students work on pages 7-8 of S5.            | Students work together on pages 7-8 of narrative: Comparing, critically evaluating and contrasting primary literatures, discussing how similar research can come to different conclusions, and synthesizing results for public communication. | 30 minutes     |
| Mini-lecture.                                | Review paper summaries.                                                                                                                                                                                     | 6 minutes      |
| Class discussion.                            | Whole class discussion 4: students discuss differences in findings between Kurz and Moore (respiration).                                                                                                   | 4 minutes      |
| Mini-lecture.                                | Discuss similarities and differences between the two papers – why did they draw different conclusions?                                                                                                     | 4 minutes      |
| Class close.                                 | Tell students when/how they should turn in their case study packet, any other announcements.                                                                                                                   | 2 minutes      |
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Table 2. Table showing the average score (percent) students (n=184) received on quantitatively analyzed questions that students were asked before and after the case study to assess student content gains. The last row shows the average score students gave for topic relevancy (out of 5). Changes in the average score for a given question after students took part in the case study are shown, with increases in average score demonstrated with green text and up-arrows and decreases in average score demonstrated by red text and down-arrows. Lastly, the table reports T-values and P-values from Student T-Tests on differences between student scores before and after the case study.

| Question                                                                 | Pre  | Post | Change | T    | p    |
|--------------------------------------------------------------------------|------|------|--------|------|------|
| List one way that a living organism could indirectly remove carbon from the atmosphere (plants going through photosynthesis does not count) | 64.2%| 61.6%| -2.6   | -0.47| 0.63 |
| List one way that a living organism could indirectly release carbon to the atmosphere (an animal respiring does not count) | 49.4%| 63.4%| 14.1   | 2.56 | 0.01 |
| Choose all the possible fates of a carbon atom in the trunk of a tree that has died | 72.7%| 73%  | 0.3    | 0.19 | 0.85 |
| Justify your choices for possible fates of a carbon atom in the trunk of a tree that has died | 77.6%| 81.2%| 3.6    | 1.85 | 0.06 |
| How relevant of an issue is pine beetle kill to you and your community? | 2.7/5| 2.9/5| 0.2    | -1.74| 0.08 |

Table 3. Table showing the number of students whose responses included a given code (n=220 pre-assessment and 233 post-assessment) when answering the question; “two scientists studied the same ecological question. Both conducted their studies in the field with the same treatment and control and measured the same independent and dependent variables. One scientist found that the dependent variable increased in response to the treatment, but the other found that it decreased. What are possible reasons why this difference in the results occurred?” before and after the case study. The frequency of our various codes was significantly different between pre- and post-assessments (X-squared = 65.77, P<0.0001, df=7). Darker green indicates more students whose response included a given code. Changes in the number of responses with a given code after students took part in the case study are shown, with increases demonstrated by green text and up-arrows and decreases demonstrated by red text and down-arrows.

| Code               | Pre | Post | Change |
|--------------------|-----|------|--------|
| Study System       | 64  | 69   | 5      |
| Season or Duration | 39  | 78   | 39     |
| Spatial Scale      | 1   | 22   | 21     |
| Data Type          | 24  | 28   | 4      |
| Confounding Factors| 46  | 28   | -18    |
| Scientist Bias     | 8   | 2    | -6     |
| Scientist Error    | 29  | 2    | -27    |
| Miscellaneous      | 9   | 4    | -5     |
Table 4. Table showing the proportion of students given materials with a narrative storyline (“Narrative”) and those without (“NonNarrative”) whose response was given a particular code when answering the question; “in what ways did the structure of this case study convince you (or not convince you) that pine beetle kill is relevant (or not relevant) to you and your community?”. The proportion of students whose responses fell into different categories was not different between students who had materials with a narrative storyline and those without (X-squared = 0.16, P=1, df=10). Darker green indicates more students whose response included a given code. Differences in the proportion of responses with a given code between students given materials with a narrative storyline and those without are reported, with positive differences (i.e. storyline caused higher proportion of a given response as compared to non-narrative storyline) demonstrated by green text and up-arrows and negative differences demonstrated by red text and down-arrows.

| Code                                      | NonNarrative | Narrative | Difference |
|-------------------------------------------|--------------|-----------|------------|
| Beetles Change C Cycle                    | 0.26         | 0.40      | ↑ 0.14     |
| Overall Content Knowledge                 | 0.11         | 0.02      | -0.09      |
| Evidence was Convincing                   | 0.11         | 0.09      | -0.02      |
| Local Scale Makes Relevant                | 0.21         | 0.18      | -0.02      |
| National Scale Makes Relevant             | 0.06         | 0.02      | -0.04      |
| Non-Specific Statement of Relevance       | 0.06         | 0.07      | ↑ 0.01     |
| Student is Not Local, Still Relevant      | 0.02         | 0.02      | 0          |
| Contrasting Results Strengthen Relevance  | 0.02         | 0.00      | -0.02      |
| Unclear/Not Convinced                     | 0.05         | 0.07      | ↑ 0.02     |
| Issue is Irrelevant                      | 0.04         | 0.04      | 0          |
| Miscellaneous                             | 0.07         | 0.02      | -0.05      |