

College Students' Understanding of the Carbon Cycle: Contrasting Principle-based and Informal Reasoning

LAUREL M. HARTLEY, BROOK J. WILKE, JONATHON W. SCHRAMM, CHARLENE D'AVANZO, AND CHARLES W. ANDERSON

Processes that transform carbon (e.g., photosynthesis) play a prominent role in college biology courses. Our goals were to learn about student reasoning related to these processes and provide faculty with tools for instruction and assessment. We created a framework illustrating how carbon-transforming processes can be related to one another during instruction by explicitly teaching students to employ principle-based reasoning—using, for example, laws of conservation of energy and matter. Frameworks such as ours may improve biology instruction more effectively than a strategy of cataloging alternate conceptions and addressing them individually. We created four sets of diagnostic question clusters to help faculty at 13 US universities assess students' understanding of carbon-transforming processes from atomic-molecular through ecosystem scales. The percentage of students using principle-based reasoning more than doubled from 12% to 27% after instruction, but 50% of students still poorly used principle-based reasoning in their responses, and 16% exhibited informal reasoning with no attempt to trace matter or energy.

Keywords: active teaching, misconceptions, conservation of matter and energy, respiration, photosynthesis

In this article we explain and discuss the implications of our investigation of college students' ability to apply the principles of conservation of matter and energy when reasoning about biological processes such as photosynthesis, respiration, and biosynthesis. Although principle-based scientific reasoning is an essential skill for scientific literacy, it is rarely learned, even by students at the college level (Chi et al. 1981, Lawson 1988, Gilbert 1991, Treagust et al. 2002, Wilson et al. 2006). Instead, most college students rely on mainly informal reasoning derived from their personal experiences when answering biological questions that require synthesis or application. For example, a student reasoning informally about weight loss will not trace matter once it leaves the individual; this student might instead say that fat “melts away” or is “burned off.” In contrast, a student using principle-based reasoning would attempt to account for the matter being oxidized, and is more likely to recognize that matter is exiting the body as carbon dioxide and water.

Principle-based reasoning and misconceptions

Students' alternative conceptions is an important area of biology education research that has produced a lengthy

list of misconceptions related to student understanding of biological processes such as photosynthesis (table 1). Our goal has been to move beyond this research by identifying problematic patterns in students' thinking that extend across content covered in college-level biology courses and are the root cause of many seemingly unrelated misconceptions. In particular, we suggest that students' misconceptions about many biological processes are connected with their failure to understand fundamental principles that constrain all biological models. We focus on principles associated with tracing matter and energy through biological systems at multiple scales.

Although biological models can be detailed and complex, biologists understand that all processes and interactions take place in a hierarchy of systems at multiple scales, constrained by fundamental physical laws. Photosynthesis, digestion, biosynthesis, and respiration involve transformation of inorganic and organic carbon-containing compounds, and biologists recognize that the products and reactants are the same atoms rearranged into new molecules (the principle of conservation of matter). These processes also involve energy transformations that are constrained by the principle of conservation of

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Table 1. Sample of known and hypothesized students' misconceptions related to carbon-cycling processes categorized by whether the root cause is an inability to apply the principles of conservation of matter and energy or a failure to choose the appropriate scale for reasoning.

Task or concept	Misconception	
	K-12	College
Tracing matter	Plants either do not respire, or if they do, they only respire in the dark (Haslam and Treagust 1987).	Oxygen is needed by animals, whereas carbon dioxide is needed by plants (Anderson et al. 1990).
	Gases are not matter or they don't have mass (Benson et al. 1993).	Food that is broken down in respiration leaves an animal's body entirely by urine and feces (Wilson et al. 2006).
Tracing energy	Respiration produces energy, rather than converts energy (Gayford 1986).	Energy can disappear (Wilson et al. 2006).
		Energy is used up during biological processes (Wilson et al. 2006).
Conflating matter and energy	Carbon dioxide contains chemical potential energy that living things can use (hypothesized).	Carbon dioxide contains chemical potential energy that living things can use (hypothesized).
	Water contains chemical potential energy that living things can use (hypothesized).	Water contains chemical potential energy that living things can use (hypothesized).
		Living organisms can convert matter into energy on a measurable scale (Wilson et al. 2006).
		Adenosine triphosphate is energy rather than a molecule with chemical potential energy (Wilson et al. 2006).
Recognizing appropriate scale for reasoning	Cellular respiration and the process of breathing are the same (Haslam and Treagust 1987).	Cellular respiration and the process of breathing are the same (Songer and Mintzes 1994).
	Respiration and combustion have little in common (Mohan et al. 2009).	
	Gases such as carbon dioxide lack sufficient mass to lead to the development of dry biomass in plants. Plants get mass from the soil (Driver et al. 1994).	

energy. In this study, we examined college students' awareness of and ability to use these fundamental principles when reasoning about carbon-transforming processes.

Research objectives

Our work builds upon earlier research with college biology students in the diagnostic question cluster (DQC) project (Wilson et al. 2006), as well as efforts to describe patterns in K-12 student reasoning about carbon-transforming processes (Mohan et al. 2009). These patterns were framed within a learning progression that describes how K-12 students can move from using purely informal patterns of thinking to applying qualitative scientific models and principles, with transitional stages that represent a growing awareness of hidden mechanisms and increasing use of scientific terminology. Although this learning progression was intended to describe transitions during high school, we believe that many college students have not yet developed consistent use of principle-based reasoning across scales (Wilson et al. 2006). A major goal of this project therefore was to identify more effective ways to address this gap in reasoning among college students.

We investigated college students' ability to apply the principles of conservation of matter and energy when reasoning about biological processes. We focused on students'

abilities to trace matter and energy across processes (e.g., photosynthesis, biosynthesis, and cellular respiration) and across scales (e.g., atomic-molecular to organismal to ecosystem). We chose to focus on these principles and processes because they underlie much of the content that we teach students in undergraduate biology courses (table 2), and because an understanding of these processes is essential for scientific literacy and good citizenship practices. For example, being able to trace matter and energy into and out of the atmosphere as a result of photosynthesis and respiration is necessary before one can understand the causes and consequences of global climate change.

Assessment development

We examined students' abilities to apply the principles of conservation of matter and energy across scales to questions about carbon cycling using DQCs, which are sets of interrelated questions about core biological concepts and ideas. These clusters are "diagnostic" in that the questions are designed to identify specific problematic reasoning tendencies that students have (e.g., students who think matter can become energy in a biological context). Diagnostic questions are considered "clusters" because answers to multiple questions may identify patterns that are consistent for that student or even for an entire class.

Table 2. Framework relating typical introductory biology curriculum to conservation of matter and conservation of energy, arranged by the appropriate scale for reasoning about the processes.

Scale	Matter	Energy
Atomic-molecular or cellular	Photosynthesis: Calvin cycle, light and dark reactions Transformation: anabolism, catabolism Oxidation: Krebs cycle, glycolysis	Photosynthesis: transmembrane proton pumps
Macroscopic or organismal	Photosynthesis: net photosynthesis Transformation: consumption or eating, biomass allocation and growth Oxidation: heterotrophic respiration, autotrophic respiration, decomposition and decay	Photosynthesis: net photosynthesis Transformation: growth, biosynthesis Oxidation: respiration, movement, temperature regulation, decomposition and decay
Large scale (ecosystems)	Photosynthesis: net primary production Transformation: consumption, herbivory, predation, biomass allocation, sequestration Oxidation: net ecosystem respiration	Photosynthesis: net energy storage in plant biomass Transformation: energy flow through food webs (especially efficiency) Oxidation: electricity generation, atmospheric warming

In addition to being diagnostic and clustered, DQCs also have the following characteristics: (a) They combine questions in different formats, such as open response, multiple choice, multiple true-false, and mixed formats where students choose an answer, then explain; (b) they are brief, fitting on the front and back of a single page and can be completed by most students in 15 minutes; and (c) the questions do not focus on the details of biological processes—instead, they are application questions designed to assess whether students' reasoning is based on scientific principles or informal reasoning (e.g., making inappropriate inferences about biological phenomena based on personal experiences or commonly used informal language). The DQCs, their underlying framework, and other supporting information are on our Web site, *Thinking Like a Biologist* (www.biodqc.org).

Our research framework (table 2) included three primary components pertaining to reasoning about the carbon cycle: principles, processes, and scale. Each question within these DQCs focused on the overarching principles of conservation of matter or conservation of energy or both. Each question explored student understanding of one or more of three basic processes: generation (e.g., photosynthesis, primary production), transformation (e.g., building of biomolecules within an organism, consumption of one organism by another), and oxidation of organic carbon (e.g., autotrophic respiration, heterotrophic respiration, decomposition, combustion of fuels). Most questions required students to make connections across atomic-molecular, organismal, and ecosystem scales.

Two of the DQCs were primarily about conservation of matter and two focused on conservation of energy. The DQCs were designed to be roughly equivalent in difficulty. Some questions appeared in both pre- and postinstruction DQCs, and several appeared in both the matter and the energy DQCs. Questions from the DQCs were taken from published sources (Ebert-May et al. 2003, Wilson et al.

2006) or developed by us. To develop questions, we asked open-ended written questions about a principle or process and interviewed students. We then refined the questions and improved their targeting by using common student answers as distractors in multiple-choice and true-false formats. Questions in the DQCs were sometimes ambiguously worded, allowing us not only to diagnose whether students had a correct or incorrect understanding of a carbon-related process but also to uncover their ways of reasoning about carbon-related processes. Each question was classified according to the principle and process it primarily addressed.

Administering assessments

Assessments were given to 525 students in 13 universities across the United States in fall 2008 and spring 2009. Institutions included three large public universities, three small- to medium-sized public universities, six small- to medium-sized private liberal arts institutions, and one community college. Students assessed were enrolled in courses ranging from introductory biology to upper-level ecology, and ranging in size from 16 to 145 students. Before administering DQCs, faculty participated in a one-day workshop in which they received instruction about the framework, guidance on how to score extended answers, and an introduction to using active teaching strategies to help students trace matter and energy separately through biological processes. Instructors chose whether to use the "Conservation of Matter" or "Conservation of Energy" pair of DQCs, or both. They gave one DQC in a pair as a pretest at the beginning of the semester. For the posttest, half of the students in each class were again given the pretest DQC, and the other half were given the other DQC in the pair. This approach was intended to verify that the relative difficulty of the DQCs was comparable. In between administration of the pre- and posttests, instructors used one to three active-learning lessons designed to teach students to explicitly trace and

conserve matter and energy in carbon-related processes. (Visit www.biodq.org to view lessons.) Instructors were provided with active-learning activities that they could adapt for use in their own classrooms as well as advice and support from the authors and other faculty.

Coding student responses

From the student responses, we tabulated how many students at each institution chose each distractor for each question that was in a multiple-choice or true-false format. In addition, faculty scored each student's written explanation for each question (multiple choice, true-false, and short answer) using a rubric that we developed from an initial subset of 25 to 30 actual student responses. The scoring rubric was based on four categorical ratings. A score of 4 indicated principle-based reasoning; a score of 3 indicated mixed reasoning (students apply principles of conservation of energy and matter, although incompletely); a score of 2 indicated informal reasoning (no principle-based reasoning); and a score of 1 ("no data") indicated that the student gave a non-sense answer, said "I don't know," skipped the question, didn't reach the question, or gave an illegible response.

An additional coder coded every 10th student answer to each question. When the discrepancy rate between coders was greater than 10%, we revised the coding rubric to resolve the issue that caused unreliable coding and then recoded all answers for that question.

Exploration of qualitative and quantitative trends

We examined student responses to individual questions for qualitative trends in reasoning that were common across questions. We specifically explored patterns that were related to students' ability to trace matter, trace energy, or recognize the appropriate scale at which to reason about a question. We also looked for characteristics that were prominent in informal and mixed reasoning so that we could better define the nature of informal and mixed reasoning.

We used Pearson's χ^2 test and contingency tables to examine whether the proportionate distribution among categorical responses changed significantly after instruction. All

statistical tests were performed using the statistical package R (R Development Core Team 2005). We did not attempt to make inferences about differences in learning gains as a function of instructor or specific lessons used because faculty had license to adapt lessons, we did not observe classroom instruction, and the classes varied in size and ability.

Contrasting principle-based, mixed, and informal reasoning

Some students applied principle-based reasoning (12% in pretests and 27% in posttests) to problems related to carbon cycling, and some applied purely informal reasoning (22% in pretests and 16% in posttests). However, the majority (58% in pretests and 50% in posttests) of college students used a combination of informal and principle-based reasoning (i.e., mixed reasoning; figure 1). As described above, we created scoring rubrics for each question. On the basis of those scoring rubrics, we identified and summarized general characteristics of principle-based,

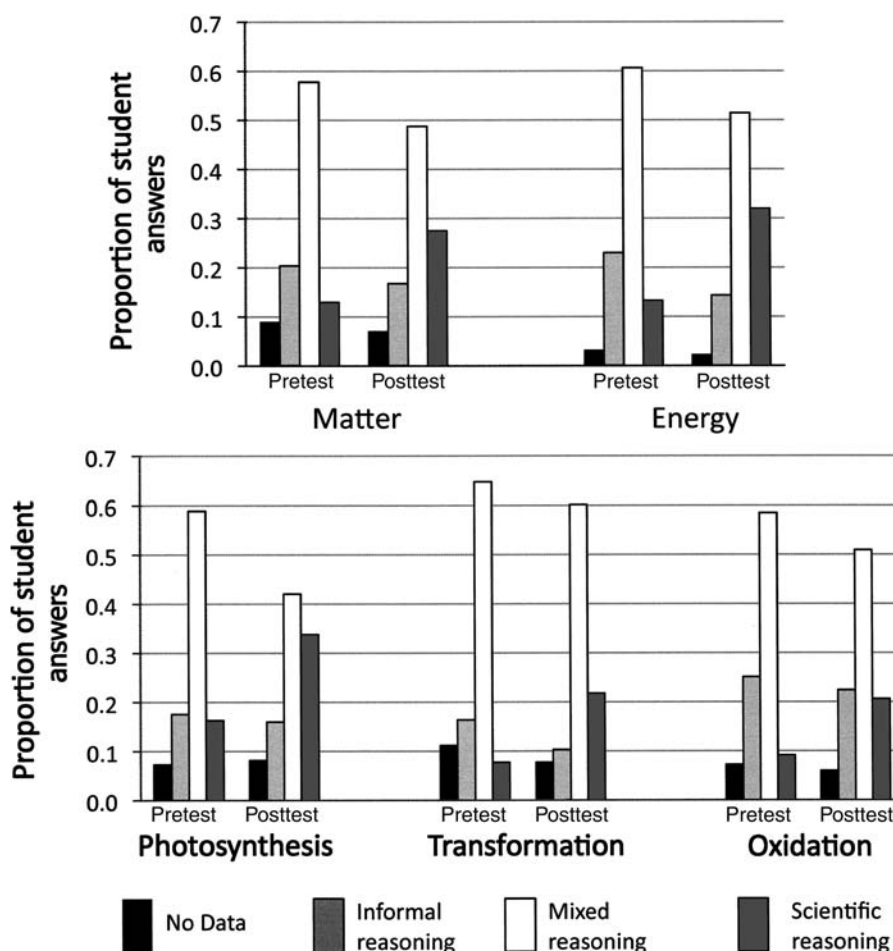


Figure 1. Proportions of student answers indicating various reasoning strategies before and after instruction. We classified each of the 42 questions according to the (a) principle and (b) process it addressed, and compared all pretest responses with all posttest responses in a given category. "No data" indicates that a student either skipped the question or answered "I don't know."

mixed, and informal reasoning responses (see box 1 for exemplar answers for each type of response). Students using principle-based reasoning were able to trace matter and energy across scales. These students did not indicate that matter could be converted to energy or that matter or energy could simply disappear; they were able to account for individual atoms as they were rearranged into different molecules and were able to account for matter when it transformed from gaseous to solid states and vice versa. Some of the most prominent indicators of mixed reasoning were a demonstration of an awareness of “invisible” processes such as movement of gases, but a lack of sufficient knowledge of the details of atomic-molecular transformations to properly account for matter and energy (e.g., incorrect matter-to-energy conversions, incorrect matter-to-matter conversions); a prolific use of scientific terminology as a substitute for properly tracing matter and energy; an oversimplification of the laws of conservation (e.g., “everything is conserved”); and the use of informal language and concepts (e.g., “fat burns off”) in part, but not all, of an answer. Students using informal reasoning did not attempt to trace matter or energy at all and relied heavily on informal language and ideas in their explanations of processes.

Qualitative trends

Although we recorded many of the same misconceptions in students’ answers that previous research has found (table 1), it is also clear that deeper problems with principle-based reasoning, particularly at atomic-molecular scales, underlie many of the responses. There were two overarching trends that we will discuss: (1) Students often use energy as a convenient “fudge factor” when they either cannot or do not see the necessity of tracing matter and energy, and (2) principle-based reasoning across scales is hampered by students’ lack of a robust understanding of atoms and molecules.

Energy as a fudge factor. Although it is true that energy and matter often are coupled in organic molecules as they move through biological systems, an inability to separately trace matter and energy becomes problematic when one reasons about the coupling of matter and energy during photosynthesis and the decoupling of matter and energy during oxidation. Students need to be able to separately trace matter and energy in order to reason about processes in the carbon cycle. Answers across multiple questions supported the idea that students often use energy as a convenient fudge factor when they either cannot or do not see the necessity of tracing matter and energy (Wilson et al. 2006, Mohan et al. 2009). We asked questions that explicitly explored whether students thought matter could become energy or vice versa (table 3). An average of 21% of students (including on pre- and post-tests) chose the distractor containing the incorrect matter-energy conversion when questions were posed in a multiple choice format, and 70% of students chose the distractor with the incorrect matter-energy conversion when the questions

were posed in a multiple true-false format. The multiple true-false format allowed faculty to diagnose that their students simultaneously may hold correct and incorrect conceptions. For example, from averaging the pre- and post-test data, 61% of students thought that some of the mass of decomposing leaves would be converted to carbon dioxide and water, but 55% of those students also thought that some of the mass would be converted into heat energy (table 3).

A previously undocumented trend we observed was that students failed to correctly account for the energetic costs of transformation of matter and energy in trophic webs. For example, when asked, “Of the energy gained by a plant (i.e., producer), what percentage is typically transferred to a rabbit that eats the plant?” 65% of students thought more than 20% of the energy gained by a plant would be transferred to the herbivore that consumes it. When asked about energy transfer through a food web, only 44% of students thought the top of a food web would have “less available energy than the trophic levels below it.” When asked about decomposition, 28% of students thought the mass of bread with mold growing on it would stay the same as the mold grows.

Furthermore, students often included phrases in their written explanations that suggested they thought energy could “disappear,” be “used up,” or be “burned up.” For example, when asked whether matter and energy could each be recycled in an ecosystem, one student wrote: “Energy can be used completely and can not be recycled once it is gone.” Phrases such as “used up” may indicate that a student is drawing inappropriately narrow boundaries around systems. For example, a student may think of respiration as affecting only an organism, rather than the organism and the surrounding atmosphere. Once energy (or matter) leaves the boundaries of the system, students no longer are compelled to account for it. Alternatively, the use of phrases like “burned up” and “used up” may indicate that students are using words and ideas from their informal lexicon, not realizing that those meanings are not directly translatable to scientific discourse. Faculty should be aware of the duality of meaning of some phrases used in both informal and scientific discourse (e.g., energy, bonds, decompose) and should explicitly address how informal discourse can impede scientific understanding.

Reasoning across scales. As educators, we would like students to be able to organize systems hierarchically from atomic-molecular to organismal to ecosystem scales, and to be able to move back and forth across scales within the hierarchy. We posed many questions at the macroscopic scale that required students to make connections to submicroscopic atoms and molecules. We also posed questions that required students to scale up microscopic phenomena to large-scale processes such as the growth of an individual plant or an entire forest. We found that students tended toward macroscopic explanations as a default, perhaps because macroscopic, observable phenomena are those with which

Table 3. Sample of diagnostic questions and student data that provide evidence of students using energy as a convenient “fudge factor” when they can’t or don’t see the necessity of tracing matter. The correct answer is bold.

Question	Sample size	Interpretation
<p>1. Which of the following are energy sources for plants? Circle ALL correct answers.</p> <p>(A) nutrients (B) sunlight (C) water (D) carbon dioxide (E) others: list sources</p>	<p>Pretest: N = 317 Posttest: N = 166</p>	<p>85% of students in the pretest and 72% of students in the posttest incorrectly chose other answers in addition to B (sunlight), indicating that students view forms of matter used by plants for photosynthesis as sources of energy.</p>
<p>2. A loaf of bread was left uncovered for two weeks. Three different kinds of mold grew on it. Assuming that the bread did not dry out, which of the following is a reasonable prediction of the weight of the bread and mold together?</p> <p>(A) The mass has increased, because the mold has grown. (B) The mass remains the same as the mold converts bread into biomass. (C) The mass decreases as the growing mold converts bread into energy. (D) The mass decreases as the mold converts bread into biomass and gases.</p>	<p>Pretest: N = 701 Posttest: N = 424</p>	<p>13% of students in the pretest and 16% of students in the posttest incorrectly chose C, indicating that matter could be converted to energy.</p>
<p>3. The trees in the rain forest contain molecules of chlorophyll ($C_{55}H_{72}O_5N_4Mg$). Decide whether each of the following statements is true (T) or false (F) about the atoms in those molecules. Some of the atoms in the chlorophyll came from...</p> <p>T F carbon dioxide in the air T F sunlight that provided energy for photosynthesis T F water in the soil T F nutrients in the soil T F glucose produced by photosynthesis T F the seed that the tree grow from</p>	<p>Pretest: N = 316 Posttest: N = 166</p>	<p>68% of students in the pretest and 60% of students in the posttest chose “sunlight” as a possible source of atoms in chlorophyll molecules, indicating that matter could be converted to energy.</p>
<p>4. Once carbon enters a plant, it can be converted to energy for plant growth. True or false?</p>	<p>Pretest: N = 342 Posttest: N = 277</p>	<p>84% of students in pretest and 77% of students in the posttest incorrectly chose “true”, indicating that matter could be converted to energy</p>
<p>5. A potato is left outside and gradually decays. One of the main substances in the potato is the starch amylose ($C_6H_{10}O_5$)_n. What happens to the atoms in amylose molecules as the potato decays? Choose true (T) or false (F) for each option.</p> <p>T F Some of the atoms are converted into nitrogen and phosphorous: soil nutrients. T F Some of the atoms are consumed and used up by decomposers. T F Some of the atoms are incorporated into carbon dioxide. T F Some of the atoms are converted into energy by decomposers. T F Some of the atoms are incorporated into water.</p>	<p>Pretest: N = 329 Posttest: N = 143</p>	<p>86% of students in pretest and 77% of students in the posttest chose true for “atoms are converted into energy,” indicating an incorrect matter-energy conversion.</p> <p>89% of students in pretest and 94% of students in the posttest chose true for “atoms are used up by decomposers” indicating that they were not deterred by the informal term “used up.”</p>
<p>6. A mature maple tree can have a mass of 1 ton or more (dry biomass, after removing the water), yet it starts from a seed that weighs less than 1 gram. Which of the following processes contributes the most to this huge increase in biomass? Circle the correct answer.</p> <p>(A) absorption of mineral substances from the soil via the roots (B) absorption of organic substances from the soil via the roots (C) incorporation of CO₂ gas from the atmosphere into molecules by green leaves (D) incorporation of H₂O from the soil into molecules by green leaves (E) absorption of solar radiation into the leaf</p>	<p>Pretest: N = 323 Posttest: N = 149</p>	<p>Only 8% in the pretest and 4% in the posttest chose the answer indicating the solar radiation could contribute to the mass of a tree. Unlike most questions, students were allowed only one answer choice.</p>
<p>7. You eat a grape. How could a glucose molecule from the grape provide energy to move your little finger?</p> <p>(A) The glucose is digested into simpler molecules having more energy. (B) The glucose reacts to become ATP (adenosine triphosphate). (C) The glucose is converted into energy. (D) The energy of the glucose is transferred to other molecules. (E) The energy of the glucose is transferred to CO₂ and H₂O.</p>	<p>Pretest: N = 97 Posttest: N = 113</p>	<p>40% of students in pretest and 42% of students in the posttest incorrectly chose C, indicating a conception that matter can become energy.</p>
<p>8. When the leaves in a compost pile decay, they lose mass. What do you think happens to the mass of the leaves? Circle true (T) or false (F).</p> <p>T F The mass goes away when the leaves decompose T F The mass is converted into heat energy T F The mass is converted into soil minerals T F The mass is converted into carbon dioxide and water Please explain your answers.</p>	<p>Pretest: N = 328 Posttest: N = 151</p>	<p>52% of students in pretest and 58% of students in the posttest chose true for “mass is converted into heat energy.”</p>

Table 4. Sample of diagnostic questions and student data that address students' understanding of atoms and molecules. For each question, the correct answer is bold.

Question	Sample size	Interpretation
<p>1. When a plant absorbs CO₂ and releases O₂ during photosynthesis:</p> <p>(A) The process increases the mass of the plant.</p> <p>(B) The process decreases the mass of the plant.</p> <p>(C) The process does not affect the mass of the plant.</p> <p>Please explain your answer.</p>	<p>Pretest: N = 48</p> <p>Posttest: N = 141</p>	<p>More than half of the students chose "C" as the answer to both questions 1 and 2, indicating that they viewed molecules of O₂ and CO₂ as equivalent in terms of mass (i.e., "one in + one out = no change"). For the first part of the question, answer choice C was selected by 56% of students in the pretest and 36% in the posttest. For the second part of the question, answer choice C was selected by 76% of students in the pretest and 62% in the posttest.</p>
<p>2. When an animal breathes in O₂ and breathes out CO₂:</p> <p>(A) The process increases the mass of the animal.</p> <p>(B) The process decreases the mass of the animal.</p> <p>(C) The process does not affect the mass of the animal</p> <p>Please explain your answer.</p>		
<p>3. You eat a grape. How could a glucose molecule from the grape provide energy to move your little finger?</p> <p>(A) The glucose is digested into simpler molecules having more energy.</p> <p>(B) The glucose reacts to become ATP (adenosine triphosphate).</p> <p>(C) The glucose is converted into energy.</p> <p>(D) The energy of the glucose is transferred to other molecules.</p> <p>(E) The energy of the glucose is transferred to CO₂ and H₂O.</p>	<p>Pretest: N = 97</p> <p>Posttest: N = 113</p>	<p>Selection of distractor "A" indicates that students do not understand that simpler molecules will not contain more energy than more complex molecules. Distractor A was chosen by 8% of students in the pretest and 5% in the posttest.</p> <p>Selection of distractor B indicates that students are not accounting for the various atoms in molecules because glucose does not contain phosphorus. Distractor B was chosen by 42% of students in the pretest and 35% in the posttest.</p> <p>Selection of distractor "C" is discussed in table 3.</p>
<p>4. A potato is left outside and gradually decays. One of the main substances in the potato is the starch amylose (C₆H₁₀O₅). What happens to the atoms in amylose molecules as the potato decays? Choose true (T) or false (F) for each option.</p> <p>T F Some of the atoms are converted into nitrogen and phosphorus: soil nutrients.</p> <p>T F Some of the atoms are consumed and used up by decomposers.</p> <p>T F Some of the atoms are incorporated into carbon dioxide.</p> <p>T F Some of the atoms are converted into energy by decomposers.</p> <p>T F Some of the atoms are incorporated into water.</p>	<p>Pretest: N = 329</p> <p>Posttest: N = 143</p>	<p>Selection of T for line 1 indicates that students are not accounting for specific atoms in molecules because amylose does not contain nitrogen or phosphorus. 63% of students in the pretest and 55% in the posttest chose true for line 1.</p> <p>Selection of T for line 2 indicates that students are not deterred by the informal term "used up," they believe that atoms can disappear, or they do not choose to follow atoms once they are consumed. 89% of students in the pretest and 94% in the posttest chose true for line 2.</p>
<p>5. A mature maple tree can have a mass of 1 ton or more (dry biomass, after removing the water), yet it starts from a seed that weighs less than 1 gram. Which of the following processes contributes the most to this huge increase in biomass? Circle the correct answer.</p> <p>(A) absorption of mineral substances from the soil via the roots</p> <p>(B) absorption of organic substances from the soil via the roots</p> <p>(C) incorporation of CO₂ gas from the atmosphere into molecules by green leaves</p> <p>(D) incorporation of H₂O from the soil into molecules by green leaves</p> <p>(E) absorption of solar radiation into the leaf</p>	<p>Pretest: N = 323</p> <p>Posttest: N = 149</p>	<p>Students do not account for transformation of matter from gas to solid forms. Choice C was chosen by 29% in pretest and 60% in posttest.</p>
<p>6. Once carbon enters a plant, it can exit the plant as CO₂. Circle true or false.</p>	<p>Pretest: N = 338</p> <p>Posttest: N = 227</p>	<p>Students do not account for transformation of matter from gas to solid forms. "False" was chosen by 60% of students in the pretest and 44% of students in the posttest.</p>
<p>7. When the leaves in a compost pile decay, they lose mass. What do you think happens to the mass of the leaves? Circle True (T) or False (F).</p> <p>T F The mass goes away when the leaves decompose</p> <p>T F The mass is converted into heat energy</p> <p>T F The mass is converted into soil minerals</p> <p>T F The mass is converted into carbon dioxide and water</p> <p>Please explain your answers</p>	<p>Pretest: N = 328</p> <p>Posttest: N = 151</p>	<p>38% of students on the pretest and 24% of students on the posttest chose true for line 1 indicating that they either think matter can disappear or they are drawing inappropriately narrow boundaries around their system.</p>

Box 1. Sample questions and actual student answers from diagnostic questions.

In this box we present sample diagnostic questions administered to students at 13 US universities that illustrate “informal,” “mixed,” and “principle-based scientific” reasoning by students. The correct answer is bold.

Question 1 asks students to reason about conservation of matter and energy at the ecosystem scale: *A tropical rainforest is an example of an ecosystem. Which of the following statements about matter and energy in a tropical rainforest is the most accurate? Please choose ONE answer that you think is best. Please explain why you think that the answer you chose is better than the others.*

- (A) Energy is recycled, but matter is not recycled.
- (B) Matter is recycled, but energy is not recycled.**
- (C) Both matter and energy are recycled.
- (D) Neither matter nor energy are recycled.

Answers demonstrating informal reasoning:

“C, Matter can be recycled that’s why we have recycling bins. Energy can also be recycled and used again.”

“C, This answer is best because every living organism requires energy to live and they also depend on other organisms to gain that energy via food.”

“C, The ecosystem is working together to use its resources to their highest abilities.”

Answers demonstrating mixed reasoning:

“The energy is recycled through the process of photosynthesis in the plants. But, the plants themselves cannot be recycled.”

“I chose C because both cannot be created or destroyed, and so it must be recycled.”

Answer demonstrating principle-based reasoning:

“All things are made up of almost all the same atoms, CHNOP and decompose and become different forms that can be recycled. Energy on the other hand as it moves along through different levels becomes less and less useful and is released as heat.”

Question 2 asks students to trace matter into a plant during photosynthesis and growth; it is posed at the macroscopic scale but requires students to also trace atoms and molecules. *Each spring, farmers plant about 5 to 10 kilograms (kg) of seed corn per acre for commercial corn production. By fall, this same acre of corn will yield approximately 4 to 5 metric tons (4000 to 5000 kg) of dry, harvested corn. What percent of the dry biomass of the harvested corn was once in the following substances and locations? Fill in the blanks with approximate percentages; you may use 0% in your response if you feel it is appropriate.*

Answers demonstrating informal reasoning:

“It takes a little of each substance to allow the plant to grow.”

Answers demonstrating mixed reasoning:

“Solar radiation is not matter. Water is a large part of living organisms.”

“CO₂ does add weight but not a lot as gas and solar energy do not have much mass they do not take up much of the weight.”

Answers demonstrating principle-based reasoning:

“The bulk of the matter comes from water absorbed through the roots and carbon dioxide absorbed through the leaves. It also does get a very small amount of potassium, nitrogen, and phosphorus from the soil.”

Question 3 asks students to trace carbon atoms once they enter a plant during photosynthesis: *Once carbon enters a plant, it can become part of the plant cell walls, protein, fat, and DNA. Circle True or False. Explain.*

Answers demonstrating informal reasoning

“True, CO₂ could possibly seep into the cell walls of a plant.”

“True, Carbon is a building block for all living organisms.”

Answers demonstrating mixed reasoning:

“True, It is used to make glucose.”

“True, As the process of cellular respiration takes place, lots of molecules including carbon, get broken down and used in the body.”

Answers demonstrating principle-based reasoning:

“True, Then biological processes in the cell turn those simple compounds into all the other compounds that the tree needs to live...proteins, lipids, nucleic acids.”

Question 4 asks students to trace energy through a food chain and account for energy costs associated with transformation of organic molecules: *Consider the three diagrams. They represent three situations in which 100 kg of green plants serve as the original source of food for each of the food chains. In situation I, for example, cattle eat 100 kg of green plants and then people eat the beef that is produced by the cattle as a result of having eaten the plants. In which of the three situations is the most energy available to people?*

- (A) I (shows food chain with green plants, insects, fish, people)
- (B) II (shows food chain with green plants, cattle, people)
- (C) III (shows food chain with green plants and people)**
- (D) Situations I and II will roughly tie for the most energy.
- (E) The same amount of energy will be available to people in all three situations.

Answers demonstrating informal reasoning:

“B, Cattle for 100 kg provides more calories for less compared to fish and green plants.”

“B, Meat converts to energy.”

Answers demonstrating mixed reasoning:

“C, No energy is lost when humans eat plants. When other organisms eat plants, some is used for systems to work, some contributes to the organisms total mass and some is excreted as waste.”

“E, Energy can never be created or destroyed. Energy in = energy out. The energy is just recycled.”

Answers demonstrating principle-based reasoning:

“C, The sun provides the most energy we use. The plants use the sun so really all energy comes from the sun. Every chain away from the plants is less energy because each animal uses some of the energy provided from the link before them.”

students have the most experience. In order to scale up or down from the macroscopic, students must have an understanding of atoms and molecules, and they need to realize that they must apply this knowledge in order to reason about larger-scale processes.

A second underlying theme in our data is that students lack a robust understanding of atoms and molecules (table 4). We asked questions that required students to properly account for the rearrangement of atoms into different molecular forms during carbon-transforming processes. Common problems with tracing atoms and molecules included incorrect matter-matter conversions, an indication that students thought atoms could become other atoms, and a lack of understanding that not all molecules contain the same amount of energy (table 4). Additionally, students have difficulty tracing matter from solids to gases and gases to solids—they instead believe in oversimplified gas-gas and solid-solid cycles. Several common misconceptions, such as the beliefs that plants get carbon from the soil, that most carbon returns to the soil during decomposition, and that organisms take in food as solids and lose solid mass rather than gas mass during respiration or weight loss, are indicative that students have difficulty tracing matter from solids to gases and vice versa. Finally, we did not see indications that college students, most of whom had experience with college chemistry, view objects as homogenous, whole entities rather than as associated particles. However, this extreme macroscopic view of matter is common among K–12 students (Mohan et al. 2009). Our results indicate that even if students know that matter and energy must be conserved, they may still be unable to properly trace matter and energy because they lack correct conceptions about the particulate nature of matter. This information may have a bearing on decisions about how to order or align chemistry and biology courses within college curricula.

Quantitative results

For all questions combined, the proportionate distribution among categorical responses changed significantly after instruction ($\chi^2 = 626.8$, degrees of freedom = 3, $p < 0.0001$). The number of responses indicating principle-based reasoning increased from 12% to 27%, but even the highest proportion of students thinking scientifically at the end of the course was just 30% (in the case of photosynthesis and energy-related questions; figure 1). For all questions combined, the number of responses indicating informal reasoning decreased from 22% to 16%, but just over 20% of students were still reasoning informally about oxidation-related questions (figure 1). The most salient finding is that for all principles and all processes, at least half of the students (58% in pretests and 50% in posttests) provided responses that incorporated a mixture of scientific and informal reasoning. Further work is needed to ascertain with statistical reliability whether there are differences in difficulty among processes and across scales. On the basis of the qualitative trends we observed, we hypothesize that all principles are

equally difficult for students to learn. We also hypothesize that of all the scales covered in this study, reasoning about processes at the atomic-molecular scale is the most difficult. Further work using teaching interventions is needed to ascertain and thoroughly describe the nature of specific teaching strategies that are most successful at improving students' use of principle-based reasoning.

Conclusions

Our research shows that some college students correctly apply scientific principles when reasoning about the processes of the carbon cycle, but the majority of students use a mix of principle-based and informal reasoning when asked to answer questions that require application or synthesis. We suggest that one reason students cannot trace matter and energy across processes and scales is that they lack a fundamental understanding of atoms and molecules (e.g., Benson et al. 1993). Another reason is that students often try to reason about large-scale or small-scale phenomena by inappropriately applying cultural models or their own embodied experiences, both of which are situated in the macroscopic world.

Applying fundamental principles such as conservation of matter and energy seems so straightforward to most biologists that they are hardly aware they do it. Their accounts of biological processes are constrained by the conservation laws in ways analogous to the ways their writing and speech are constrained by the rules of English grammar—they follow the rules more or less automatically. Yet even on posttests, the majority of students, even biology majors taking advanced courses, did not follow the rules automatically. So, why is applying these simple principles so hard? We think the answers to this question lie in the deep-seated nature of informal reasoning and in the way we currently teach biology.

Informal reasoning is deep seated. Theories about language and informal reasoning are useful in interpreting why students have difficulty when trying to move from informal to scientific discourse (Mohan et al. 2009). When using informal reasoning, students look for “actors” that drive “events” and are aided by “enablers.” For example, a tree grows if it has water, sunlight, and nutrients. We believe students can generally identify actors at the organismal scale. At the cellular and molecular scales, students have trouble thinking of atoms, molecules, and cells as actors. When students think about organisms as actors, they are precluded from thinking about the components and cells within the actor, making movement across scales difficult.

Many students' ideas are based on pervasive informal models of living systems that are rooted in the nature of language (Kempton et al. 1995, Gee 2004). Our everyday language and our personal experiences lead us to believe, for instance, that we lose weight by “burning off” fat. Students have had a lifetime of experience using informal reasoning to generate predictions and explanations of phenomena they observe. For the most part, this has been a

successful strategy because it has allowed them to generate plausible explanations of macroscopic phenomena (Poza and Gomez-Crespo 2005). However, informal reasoning can lead to and reinforce misconceptions when applied to domains outside of students' macroscopic experiences, such as processes occurring on molecular, cellular, ecosystem, or global scales. These habits of mind have served them well in many situations, and students often are unaware of how informal reasoning conflicts with biochemical accounts of processes in living systems.

Biology textbooks and teaching do not adequately teach principle-based reasoning. Expert scientists recognize automatically that physical and chemical laws constrain biological processes (Chi et al. 1981); they therefore don't think to explain their reasoning to students, or they may assume that students are already applying the same models. Biology courses typically emphasize scientific models of biological processes at multiple scales. For example, most introductory biology textbooks in the United States (e.g., Campbell and Reece 2007, Brooker et al. 2008) provide very detailed accounts of the atomic-molecular pathways and cellular structures involved in photosynthesis, respiration, and biosynthesis. These books also include related models of structure and function at other scales, such as organismal physiology or carbon cycling and energy flow at the ecosystem scale. Though the textbooks imply that these models are connected, we suggest that students often fail to see the connections through principles and across scales, and therefore don't organize their knowledge using principles.

Most college-level instruction presents students with complicated narratives about the details of key processes (e.g., cellular respiration), but does not explicitly reinforce the use of key principles to connect those processes. Therefore, students are understandably occupied with memorizing details of processes without focusing on the principles that govern and connect the processes. Faculty may erroneously assume that because students can incorporate scientific vocabulary into their explanations of phenomena and state important constraining principles (such as conservation of matter and energy) they can and do apply those principles when reasoning about biological processes.

When students memorize details while continuing to reason informally about processes, the consequences can be serious. These students can fail to connect related processes such as photosynthesis and respiration or fail to connect the same process across different scales, such as photosynthesis and gross primary production. As a result, students may leave an introductory biology course with the ability to recite the reactions in the Calvin cycle but still believing that plants obtain most of their mass from the soil rather than from the atmosphere, that plants photosynthesize but do not respire, or that the mass of a decomposing organism will primarily return to the soil.

Faculty may be so accustomed to using principle-based reasoning skills that they do not realize their students are more inclined to use informal reasoning skills. Thus, faculty are unknowingly speaking a different language from their students. We define principle-based reasoning as a "hidden curriculum" because it is so familiar to biologists that they are hardly aware they use it; biologists assume students understand it, even when they do not.

Implications for college biology teaching. Faculty who use DQCs in their courses may be discouraged by limited improvements in students' reasoning skills in posttests, even when they use active-learning strategies designed to target identified problems (see www.biodqc.org). However, given the intractable nature of student beliefs about processes such as photosynthesis and respiration (e.g., Anderson et al. 1990), students' very limited experience applying scientific reasoning, and the challenges of reforming introductory biology courses, it may be unreasonable to expect rapid progress.

Science instructors at both precollege and college levels need to help students see the necessity of using principle-based reasoning and help them understand that the application of everyday, informal models in scientific situations interferes with principle-based reasoning. Instructors should also acknowledge that the practice of rote memorization is counterproductive to building knowledge successively across the biology curricula. This transition won't be easy; ideas and misconceptions associated with informal reasoning are powerful because they are based in students' worldviews and longstanding practices as biology learners. For students to see the power of principle-based reasoning, we suggest they will need sustained, active learning over multiple courses that challenge informal language and reasoning. Instructors need tools that enable them to connect the details of course content and student responses to the principles behind them. Thus, instruction should explicitly help students connect processes across scales using principle-based reasoning. Instructors who highlight underlying principles during instruction can simultaneously address multiple misconceptions (e.g., table 1), rather than cataloging them and addressing them individually. DQCs are one tool for helping faculty see where their students are having difficulties, knowledge that can help instructors strengthen their instruction.

The focus of this article is on student understanding of conservation of matter and energy, but we look forward to more research on student understanding of other fundamental principles in biology and on how principles can be used to connect content across scientific disciplines. One goal should be to move beyond misconception-focused research by identifying the problematic patterns in students' thinking that extend across content covered in biology and other science courses, and that are the root cause of multiple seemingly unrelated misconceptions. Another goal should be to develop and test the efficacy of teaching approaches to help students develop principle-based reasoning skills.

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Laurel M. Hartley (laurel.hartley@ucdenver.edu) is an assistant professor of integrative biology at the University of Colorado, Denver. Brook Wilke is a graduate student studying agroecosystems, Jonathon Schramm is an ecologist and postdoctoral researcher studying science education, and Charles Anderson is a science education researcher and professor, at Michigan State University, in East Lansing. Charlene D'Avanzo is a professor of ecology in the School of Natural Sciences at Hampshire College, in Amherst, Massachusetts.