

Improved Student Reasoning About Carbon-Transforming Processes Through Inquiry-Based Learning Activities Derived from an Empirically Validated Learning Progression

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Abstract This paper reports on our use of a fine-grained learning progression to assess secondary students' reasoning through carbon-transforming processes (photosynthesis, respiration, biosynthesis). Based on previous studies, we developed a learning progression with four progress variables: explaining mass changes, explaining energy transformations, explaining subsystems, and explaining large-scale systems. For this study, we developed a 2-week teaching module integrating these progress variables. Students were assessed before and after instruction, with the learning progression framework driving data analysis. Our work revealed significant overall learning gains for all students, with the mean post-test person proficiency estimates higher by 0.6 logits than the pre-test proficiency estimates. Further, instructional effects were statistically similar across all grades included in the study (7th-12th) with students in the lowest third of initial proficiency evidencing the largest learning gains. Students showed significant gains in explaining the processes of photosynthesis and respiration and in explaining transformations of mass and energy, areas where prior research has shown that student misconceptions are prevalent. Student gains on items about large-scale systems were higher than with other variables (although absolute proficiency was still lower). Gains across each of the biological processes tested were similar, despite the different levels of emphasis each had in the teaching unit. Together, these results indicate that students can benefit from instruction addressing these

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processes more explicitly. This requires pedagogical design quite different from that usually practiced with students at this level.

Keywords Learning progressions · Carbon cycle · Photosynthesis · Respiration · Misconceptions

Introduction

Carbon-transforming processes, including photosynthesis (which reduces carbon into an organic molecule), biosynthesis (digestive and other metabolic processes that alter carbon molecules without large changes in redox state), and cellular respiration (which oxidizes carbon back to inorganic forms), are an important set of topics in secondary-level biology. Ideas about how matter and energy transform in carbon-transforming processes are emphasized as disciplinary core ideas in the recently released US National Research Council K-12 Science Education framework (Quinn et al. 2012) and the Next Generation Science Standards (NGSS Lead States 2013).

It is not difficult to see why these processes receive such attention in teaching and learning biology. It is impossible to understand life on earth without a basic grasp of the matter cycles and energy flows that link organisms to each other and to their abiotic environment, and these movements underlie everything from the trophic structure of ecosystems to biodiversity across the globe. Further, understanding these biological processes is essential for making responsible decisions as global citizens in this era of climate change and other global conundrums. Public debate about carbon sequestration strategies, for instance, makes little sense without recognition that all such strategies depend on moving carbon into very stable molecules and locations, often beginning with photosynthesis in long-lived, woody plants. It is essential that biology curricula continue to strengthen students' understanding of these vital processes.

In the USA, carbon-transforming processes together account for large portions of the secondary-level curriculum. Students learn about plant growth and photosynthesis at several points in the range of 7–12th grade (also earlier, in primary school), with each iteration adding more sophistication and nuance to the students' understandings (Stern & Roseman 2004). Likewise, generations of high school students have built on their earlier understanding of food and macromolecules to consider how the chemical energy provided by those molecules is released during cell respiration (Assaraf et al. 2013). Although molecular-level biosynthetic processes are typically as often represented in state science standards as photosynthesis and respiration, they tend to receive substantially less attention in most classrooms (Banet & Núñez 1997).

Despite the current strong emphasis in most students' biology learning on carbon-transforming processes, evidence abounds that many students graduate from high school with a limited functional understanding of these processes (Brewer & Smith 2011; Hartley et al. 2011 ; Jin and Anderson, 2012; Mohan et al. 2009; NGSS Lead States 2013; American Association for the Advancement of Science 1989; Stern & Roseman 2004). In particular, students lack the ability to apply fundamental thermodynamic principles (i.e., matter conservation, energy conservation, and energy degradation) to carbon-transforming processes in real-world situations (Hartley et al. 2011). At least in part, students' low achievement could be attributed to current curriculum and instruction. The curriculum evaluation conducted by AAAS shows that current secondary curriculum materials on life sciences seldom provide adequate and clear representations to make the fundamental principles of matter and energy intelligible for students (Stern & Ahlgren 2002).

Increasingly, curriculum designers are focusing on building deeper conceptual understanding rather than more superficial learning of scientific facts or procedural knowledge (i.e., depth over

breadth; Millar et al. 1994; van Rens et al. 2010). In support of this approach, recently released National Research Council frameworks and the Next Generation Science Standards both call for classroom teaching to emphasize several disciplinary core ideas rather than potentially disjointed coverage of many concepts, principles, and facts (NGSS Lead States 2013; Quinn et al. 2012). These key ideas are crucial not only because they constrain and structure students' thinking about a wide variety of subordinate concepts but also because they are persistently difficult for many students, due to conflicts between scientific principles and our latent "everyday" reasoning patterns (Carey 1986; Chi et al. 2012; Inagaki & Hatano 2002). For example, in contradiction to the law of conservation of matter, students routinely discount the role of gases in mass changes in biological systems due to their apparently insubstantial nature as compared to liquids and solids.

In this study, we identified four such key ideas—mass, energy, subsystems (i.e., microscopic and molecular scales), and large-scale systems. These key ideas are core aspects of scientific explanations about carbon-transforming processes. They are also scientific ideas about which students hold many alternative conceptions. Based on our previous studies (Jin & Anderson 2012; Jin et al 2013), we developed a learning progression that contains four progress variables, with each variable focusing on explaining one key idea (Table 1). Learning progressions are descriptions of successively more sophisticated ways of thinking about a topic which help to make key reasoning transitions clear (Wilson et al. 2008), and they can generate powerful frameworks that align curriculum, instruction, and assessment (Alonzo & Gotwals, 2012; Fortus & Krajcik, 2012). A learning progression usually contains an upper anchor specifying a learning goal that is determined by societal expectations and content standards, a lower anchor that is defined by students' knowledge and informal conceptions when they enter a certain grade level, and intermediate levels that connect the lower anchor to the upper anchor (Mohan et al. 2009; Shea & Duncan 2013). In this study, we used a learning progression (Table 1; see Appendix Table 8 for a fuller description) developed and revised in previous studies (Jin & Anderson 2012; Jin et al 2013; Mohan et al. 2009) to guide the design of a curriculum (Table 2) that supports students in learning carbon-transforming processes, as well as to assess the learning outcomes. We then examined to what extent that curriculum was associated with improved student reasoning about carbon-transforming processes.

The learning progression grounds the learning goal of the curriculum: for each progress variable, we expect students to make the transition from a lower level to the upper anchor. The lower levels are about intuitive ways of reasoning that students develop based on their earlier experiences in school and everyday life, while the upper anchor (Level 4) describes the fundamental scientific reasoning for each key idea. Level 4 understanding would not represent detailed mastery of each idea, but rather represents a conceptual understanding that would be robust enough to build more scientific detail on with further study. Across all key ideas, explanations at Level 1 are based on "force-dynamic" reasoning (Pinker 2007; Talmy 1988), which frames events in terms of actors using enablers to grow or move. Explanations at Level 2 are based on "hidden mechanism" reasoning, which acknowledges processes happening at spatial or temporal scales beyond immediate observation, but treats them as undifferentiated "black boxes." Hidden mechanism reasoning is still force-dynamic in nature, because it describes how materials and energy are used up to power invisible processes such as cell multiplication, carbon dioxide, and oxygen conversion etc. Explanations at Level 3 modify this force-dynamic framework to accommodate increasing knowledge about matter, energy, and systems. For example, instead of tracing matter and energy consistently, students often explain that organic molecules are converted into energy in biochemical reactions. Explanations at Level 4 represent scientific reasoning, which uses fundamental principles (i.e., matter

	ce-dynamic accounts	Level 2 Hidden processes	Level 3 Matter transmutation (matter-energy conversion)	Level 4 Matter transformation and energy transformation
Mass changes (explaining how Desc materials change in processes and accounting for mass changes)	cribed in terms of feelings/perceptions	Involve "stuff" (phases of matter not well differentiated)	Involve organic molecules (w/ inappropriate conversions)	Explained in terms of atom rearrangement
Look	asuring mass changes in simple systems sons 4–6, 9, 10 ls: balances, matter and \energy process	(carbonated beverages losing CO ₂ ; soil tool	+ growing plant over time)	
Explaining energy transformations Simt (accounting for energy in processes)	ple cause-effect relationships	Energy traced to sources but not sinks	Attempt to trace energy fully but relies on faulty conversions to matter	Consistent and complete transformations of energy
Lear. ca Lessi Tool:	rming structures of basic molecules relev arbon; discussion of photosynthesis and sons 4 & 8 1: matter and energy process tool	ant to plant growth with a focus on rela I respiration	tively high- and low-energy molecular	r bonds involving
Explaining microscopic sub-systems Syste (accounting for processes at the atomic-molecular scale)	tems only described macroscopically	Basic sub-systems identified (e.g., cells)	Acknowledgement that all matter atomic-molecular in nature	Links bonds in organic molecules w/ available energy
Expl br Less Toolk en	loration of atoms and molecules in biold alances w/in transformations (of carbon: sons 2–4, 6, 10, and 11 ls: powers of 10 tool; matter and nergy process tool	ogical and other systems using multimec ated beverage and growing plant)	dia, inquiry to confirm importance of g	gases to mass
Explaining large-scale systems Unm (connecting processes across	nodified sequence of local events	Uses oversimplified processes (e.g., "gas cycle")	Recognition that atoms/molecules are crucial at all scales	Recognition of energy flow and matter cycling
scales, incl. global) Expl ca Less Tool:	loration of matter at vastly different spat arbon cycling as aggregation of many sı sons 2, 3, and 11 1: powers of 10 tool	tial scales (e.g. atoms to solar systems); maller processes	introduction to global scale	

student reasoning in that area, along with the relevant lessons of the intervention and tools used to help develop student reasoning

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Table 2 General structure of the	teaching unit reported o	in in this study, with regard to key idea	s addressed, inquiry approaches, and iterative	e reasoning elements employed
Lesson description	Key idea(s) stressed	Inquiry component	Tools for reasoning	Learning objectives
No. 1—how do plants grow? (Students plant bean and radish seeds; predict mass changes)	M, E	Start of plant growth experiment, incl. predictions about mass changes in soil and plant	Matter and Energy Process tool (describe changes in matter and energy as plants grow)	Experimental design and observational protocol selection; establish competence with basic equipment (scales)
2—Powers of 10 (uses Powers of 10 video by the Eames' to start discussion of systems and scales)	SS, LS		Powers of 10 tool (focus on four broad ranges of scales—large, macroscopic and microscopic, atomic-molecular)	Describe connections between objects at a variety of size or organizational scales; see fundamental similarities b/w types of matter
3—Powers of 10 as a tool (placing a variety of physical entities along a scale of sizes)	SS, LS		Powers of 10 tool (developing a sense for how entities compare in terms of size, qualitatively and quantitatively)	Increased ability to sort objects along spatial scale; understanding of which benchmark scale to discuss particular processes at
4—molecules of air, plants, and soil (reading and discussion of atoms found in each area)	M, E, SS		Powers of 10 tool (considering relative sizes of molecules and implications for biology)	Recognize that soil minerals provide other essential atoms for organic compounds (e.g., P, S, N); clear distinction between forms of matter (solids, liquids, gases made of atoms and molecules) and of energy
5—investigating weight gain and loss (series of simple observations to see mass changes; water as 'temporary' mass source)	M	Predicting and tracking mass change in simple systems (sponge and water, soil, and water)	Matter and Energy Process tool (increasing understanding of locations of mass during physical transformations)	Recognize that physical and chemical changes do not create or destroy atoms; able to measure dry mass accurately and explain rationale for techniques
6—does CO2 have mass? (tracking mass in familiar rxns with CO ₂ ; intro to CO ₂ probe)	M, SS	Tracking mass changes w/ gas in simple systems (exhalation, combustion, baking soda +wa- ter)	Matter and Energy Process tool	Able to use probes to measure CO ₂ concentrations and interpret measurements appropriately
7—plant gas exchange (using CO ₂ probe to track gas movements)	М	Tracking gas in plants under both light and dark conditions		Qualitatively trace mass from CO ₂ and H ₂ O to biomass and back again; trace energy separately from matter

Table 2 (continued)				
Lesson description	Key idea(s) stressed	Inquiry component	Tools for reasoning	Learning objectives
8—photosynthesis and respiration (detailed discussion of reactants and products of these crucial processes)	M, E		Matter and Energy Process tool (using various iterations to be specific about fates of matter in PS and BS, together w/ locations w/in organisms)	Plant growth as a two-step process: photosynthesis in leaves converts CO ₂ and H ₂ O to glucose and O ₅ ; biosynthesis converts glucose and soil minerals into other organic materials Plant functioning as using energy from cellular respiration that converts glucose and O ₂ to CO ₂ , H ₂ O
9—harvesting plants; measuring changes in soil & biomass (conclusion of plant growth experiment)	м	Tracking mass changes by careful measurement of plant, soil	Matter and Energy Process tool	Recognize soil minerals and water as essential to plant growth but making up a small portion of biomass; able to construct an argument from evidence to support a model
10—explaining changes in mass (discussion built around von Helmont's landmark observations on plant growth)	M, SS		Matter and Energy Process tool (using tools to analyze differences between respiration, photosynthesis, and biosynthesis)	Able to trace C, O, and H atoms through photosynthesis, biosynthesis, and respiration; identify chemical potential energy as contained in organic molecules; recognize differences between high-energy bonds (C–O and C–H) and low energy bonds (C–O and H–O)
11—What's the "matter" with carbon (analyzing carbon cycle diagrams in terms of PS, CR and BS; brief intro to climate change and cnxn to carbon)	SS, LS		Powers of 10 Tool (carbon occurring in different forms across benchmark scales)	Locate photosynthesis, biosynthesis, and cellular respiration within a unified carbon cycle
Also described are the key learni	ng goals for each lesson			

Also described are the key learning goals for each lesson

 ${\cal M}$ mass, ${\cal E}$ energy, SS sub-systems, and LS large-scale systems

conservation, energy conservation, and energy degradation) to explain changes in matter and energy across hierarchical systems.

Effective science teaching often pairs new content with specific scientific practices that reinforce both the meaning and significance of the content (Quinn et al. 2012; NGSS Lead States 2013). Another approach to curricular design that has been shown to improve student learning is simply to return to several key concepts repeatedly and in varied ways across a unit (Fortus & Krajcik 2012; van Rens et al. 2010). Across our teaching intervention in this study, we incorporated both of these strategies. The teaching intervention first familiarised students with the key ideas and then deepened and strengthen their ability to apply and use those ideas in other contexts. In our curriculum, this took the form of scaffolded inquiry activities followed by application exercises (Anderson, 2003; Minner et al. 2010). In inquiry activities, students carry out experiments, collect data, identify patterns from the data, and construct explanations for the identified patterns (inductive reasoning). Teachers provide scaffolding for students to develop scientific reasoning about the four key ideas as students are constructing explanations. In application activities, students use scientific reasoning, about the key ideas, learned in inquiry activities to explain phenomena in new contexts (deductive reasoning).

We also designed two specific tools for reasoning to help students visualize the scientific reasoning connecting all of our key ideas (Harrison & Treagust 1998; Louca & Zacharia 2011). One tool, which we call the "Matter and Energy Process Tool", helps students trace matter and energy in individual carbon-transforming processes, so they begin to visualize how atoms, molecules, and forms of energy change in chemical reactions (Fig. 1a). We called the other tool for reasoning the "Powers of 10 Tool." By placing a wide variety of objects along a logarithmic size scale, this tool helps students make connections among different scales: atomic-molecular, microscopic, macroscopic, and large scale (Fig. 1b). In particular, this helps students better understand subsystems and large-scale systems by relating them to more familiar scales of perception (Kozma 2003; Treagust et al. 2003).

In short, we assessed the potential of a short curriculum built around a learning progression and incorporating inquiry, application, and tools for reasoning, to improve student reasoning about three crucial carbon-transforming processes (photosynthesis, digestion/biosynthesis, and cell respiration). Given the importance of these processes to developing biologically accurate understandings, and their difficulty for many students, we hoped to assess whether instruction explicitly built around a relevant learning progression would improve learning outcomes. Since the curriculum directly addressed not only typical alternative conceptions held by students but also the fundamental patterns of reasoning underlying those conceptions, we expected that learning gains would be demonstrated by all students in the study, regardless of school level or setting. Further, we expected that student reasoning around photosynthesis and respiration would improve the most, given the time spent on these processes in the unit, but that reasoning about biosynthesis would still improve as students reorganized their conceptions of these processes. Finally, we hypothesized that students would demonstrate the greatest learning gains on reasoning about tracing matter and energy, due to their prominence within the unit.

Methods

This research was conducted as part of a National Science Foundation-funded Math and Science Partnership (MSP) studying learning progressions and environmental literacy in the context of carbon cycling. As with many learning progression approaches, this project operates out of a constructivist framework to describe the development of scientific reasoning patterns, seeking to elucidate typical patterns of reasoning transitions as scientific understandings are



Fig. 1 Graphic organizing tools were used to scaffold student reasoning in the curriculum. The "Matter and Energy Process Tool" (a) forced students to be explicit about separating forms of matter and energy and clear about the transformations undergone in biochemical reactions. For any given biochemical transformation, students would identify the specific forms of energy and amounts of various states of matter present before and after the reaction. The "Powers of 10 Tool" (b) uses a logarithmic scale to help students visualize how items with vastly different sizes relate to each other. Students were asked to place particular objects (e.g., glucose molecule, tree, farm field, etc.) at the most appropriate scale of size and organization

built in the context of latent ideas about the way the natural world works. The learning progression (and analyses used in this study) were constructed semi-quantitatively through inductive coding of actual student responses and subsequent statistical analysis.

Participants and Study Context

During the 2010–2011 academic year, 25 science teachers (9 middle school, 16 high school) in 5 US states (CA, CO, MI, NY, MD) engaged their students (216 middle school and 347 high school students) in a teaching sequence about carbon-transforming processes. Schools varied greatly in their social setting and ranged from rural to suburban to urban. The teachers and students represented a wide range of classes containing life science emphases, including several Advanced Placement courses, but most were general science courses for their respective grade levels, and thus included the typical range of student abilities.

All participating teachers attended a week-long professional development workshop hosted by researchers by project researchers, in the summer before implementation of the curriculum in their classrooms. During the workshop, teachers explored the mechanics and structure of the curriculum and the patterns of student reasoning that typified different levels of the learning progressions. Special attention was given to supporting teachers in diagnosing their own students' levels of reasoning, via pre-tests and other formative assessments, and then to differentiate their teaching based on assessment results and the learning progressions.

During the year after completion of the professional development, participating teachers implemented the curriculum in their classrooms. The curriculum contains 11 lessons, taking approximately 2 weeks of instructional time. Due to time constraints and specific requirements in some school districts, some teachers taught some but not all lessons. The same curriculum was used for all grades in the study by targeting the key ideas that are required from students at any of those grades, but it also provided suggestions for adapting the lessons to fit students at different levels. For example, middle school teachers taught their students how to identify inputs and outputs in processes at the level of substances (e.g., carbon dioxide, water, oxygen, and biomass), while high school teachers focused on identifying inputs and outputs at the level of specific molecules.

Data Collection

In each teacher's classroom, a pre-test was administrated before and a post-test was administrated after the teaching intervention. Two forms of a student assessment were created ("A" and "B"), each with 9 multi-part items. All items involved brief objective responses (multiple-choice, Y/N, multiple-select) followed by space for subjective explanations of their choices. A total of 15 content items were spread across both test forms so that each form included items about the 3 carbon-transforming processes. Items on each form also targeted all 4 key ideas (Table 3). This segregation allowed us to base conclusions on a wider range of items without testing every student on every item (thus saving substantial instructional time for teachers). Two items ("Energy for Plants" and "Carbon in Plants") were used in both forms, to verify that student populations taking each each form of the test were statistically equivalent (i.e., student proficiencies on these two items covered a similar range for both sub-populations (Forms A and B)), They were found to be

	Photosynthesis [PS]	Biosynthesis [BS]	Cell respiration [CR]
Mass	Tree growth Plant mass experiment	Infant growth Digestion and growth <i>Maple mass gain</i>	Animal mass experiment
Energy	Energy for plants (both forms) Energy in plants <i>Fate of energy</i>		Body heat
Subsystems	Plant growth experiment	Carbon in plants (both forms) Locations of carbon	
Large-scale systems		Quantitative carbon cycle	Keeling curve Carbon cycle rationale

Table 3 Classification of items on student assessments by the primary biological process and key idea covered

Items on form A are in plain font, items on form B are italicized

equivalent. Teachers administered the tests either with paper copies (not necessarily a 50/50 split in student numbers between forms A and B) or through a secure online testing system (form distribution was computer-controlled and thus exactly 50% of students took each form).

Data Analysis

We analyzed students' pre- and post-assessment data and computed students' learning gains. As students' learning gains likely have multiple causes, we cannot claim that the teaching intervention is solely responsible for any gains. But, we can examine areas of overlap between the curriculum and student learning gains. Cognizant of the fact that not all teachers taught all lessons of the curriculum, we also analyzed teachers' coverage of the curriculum to ook for correlations between the degree of coverage and student outcomes.

Analysis of Student Learning Gains All student responses were transcribed (if on paper forms) or pulled from the online system and compiled into spreadsheets that displayed all student responses, from all teachers and grade levels, and for all questions. This format allowed our research team to qualitatively code every student's responses using a coding scheme that had been generated in previous studies (Jin & Anderson, 2012; Jin et al., 2013) This was revised through several rounds of developmental coding in the present study. The coding scheme contains 15 rubrics. Each rubric is used for coding responses for one item; it contains level descriptions that are aligned with the learning progression, keys that help raters to differentiate between two adjacent levels, and exemplar responses selected from the dataset. Appendix 2 shows an item about energy in photosynthesis and its corresponding rubric. In addition to codes 1–4 (aligned with levels 1–4 of the learning progression), several other codes were used to describe answers that were outside the Level framework for various reasons (e.g., no response, student did not reach this portion of test, "non-sense" or off-topic responses etc.). For all items, a subsample of around 30 student responses was first selected, with which to train coding teams in their use of the rubrics for the various items. On items with a long legacy, and for which we knew the level characterizations were well-founded, this training subset was simply used to ensure that new coders understood and applied the rubric consistently. Newer items went through a more rigorous developmental coding process to both describe specific indicators in student responses for each of the four levels and to align those with other indicators for the same level from other items. (This involved several rounds of creating draft indicators, coding a subset of responses using those indicators, discussing inter-rater agreement or lack thereof, and revision of the indicators). In other words, we developed our rubrics by both emergent pattern-finding in the student responses and through consideration of the process-and-key idea framework, upon which our assessments and instruction were based.

Once inter-rater reliability had been unanimously established on the developmental coding subset of responses, primary coders were assigned to code all student responses for a few of the items, ranging in sample size from several hundred responses to nearly 2000. This reduced the extent of coding variation due to differences between raters. Sample sizes were larger for items that appeared on both test forms, and some variation also existed due to paper test forms being handed out to unequal numbers of students. Each item was also assigned one or more reliability check coders, who

independently coded 10% of that item's responses for comparison to the primary coder's judgments. Coding agreement of less than 90% triggered a consultation between the 2 coders to determine the source of the discrepancy, either coder misinterpretation of the rubric or a lack of clarity in the rubric given the actual range of student responses. In especially difficult cases, these discussions were also brought up to the larger research group until resolution had been achieved. In cases where coder interpretation was found to be the issue, that coder adjusted their coding work appropriately. If deeper issues in the rubric had been at fault, both coders revisited their coding, again checking for agreement in excess of 90%, and revising once again if necessary.

Final reliability-checked codes (i.e., verified primary coders' interpretations) were submitted to the Berkeley Evaluation and Assessment Research (BEAR) Center for subsequent analysis. For the analyses reported here, only data from students for whom we have both pre- and post-test responses were included (n = 563). Multidimensional item response models (IRMs) for the polytomous (student \times process \times key idea) data and multidimensional latent regression models were fitted to investigate the learning gains in general and learning gains by key ideas (Shin & Draney, 2014). ConQuest software that implements the marginal maximum likelihood estimation with an expectation-maximization (EM) algorithm was used throughout the analyses (Wu et al. 2007). These tools generate estimates of both student proficiencies (i.e., achievement on the learning progression) and of the relative difficulty of the various items for this student population. For person proficiency estimates, we used expecteda-posteriori (EAP) estimates. This method of quantification allowed us to compare individual student abilities to item difficulty levels using the same numeric scale. These can be read as a relative measure of proficiency where absolute position on the scale is less important than comparisons on that scale (i.e., on the logit scale derived from this estimate, higher numbers reflect both greater proficiency and greater item difficulty). This scale is essentially unitless or can be read as "logits". In order to compare students' proficiencies between pre- and post-test and across the practices, item parameters were estimated first and then were fixed when necessary.

Teachers' Coverage of the Curriculum

In order to better understand the potential role of class-specific instructional effects (as opposed to effects due to the structure of the unit itself on student learning), we included the exact lesson sequence used by 18 of the 25 teachers whose students contributed pre-post data, as a calculation of "curriculum coverage" for each process and key idea (we could not retrieve this level of data from the other 7 teachers). The curriculum coverage for any one variable equals the proportion of all lessons that included that variable (i.e., by design) multiplied by the average number of teachers that actually taught that lesson (i.e., due to teacher autonomy). For instance, photosynthesis was featured in 6 of the 11 lessons, and each of those lessons was taught by an average of 14 teachers. Summing a similar calculation for the other two process variables, allowed us to estimate the proportion of process coverage by teachers devoted to each of the processes. Assessment emphasis was simpler, merely merely requiring the percentage of assessment items that incorporated each process or key idea.

Together with the occurrence of each of these variables in the curriculum, this metric allowed us to compare the degree to which each process or key idea was directly taught and then to compare that with changes in student reasoning about these variables.

Results

Overall

In general, students in this study demonstrated substantial learning gains about carbontransforming processes as a result of this instructional unit. The mean proficiency of the students on the pre-test was -0.925 on a logit scale (s.e. = 0.093). This indicates that most of these students found the pre-test difficult (EAP values lower than 0 indicate lower proficiency compared to the overall item difficulty). However, the average learning gain across all students was statistically significant at 0.622 logit (s.e. = 0.059; effect size = 0.638), meaning that the average proficiency of students after instruction was -0.303. Students at all grade levels gained in proficiency to a similar extent across the course of this unit (Fig. 2). Middle school students started and finished at lower proficiencies than did ninth-graders, but they did finish the unit with a stronger understanding of the material than their older counterparts (who had not received instruction designed in this way during their middle school years had going into the unit) had going into the unit. All high school students demonstrated similar proficiency both pre- and post-instruction, an effect skewed partly by the small sample size of upperclassmen (see below).

A closer look at the data, however, reveals that student gains are more strongly linked to initial proficiency than simply to their current grade level. Students from 7th to 12th grade were involved in this study, but the single largest grade was 9th, which represented 52.4% of the total sample. Middle school students (7th and 8th grades)



Fig. 2 Pre- and post-test expected-a-posteriori (EAP) scores for middle school ("middle," 7th–8th grades), lower high school ("lower," 9th), and upper high school ("upper," 10th–12th) students, together with the effect sizes of the learning gains. All effect sizes were significantly different than zero, but none were different from each other

and upper high school students (10–12th grades) represented 38.4 and 9.2% of the total sample, respectively. In addition to age and developmental differences among these students, each of these grade bands approaches biology in the curriculum differently. Each grade also accompanied this unit with different types and complexities of additional teaching material, depending on the teachers' preferences. Because of this wide variation in prior knowledge, age, and related experiences, for analysis, we divided the entire sample into low-, medium-, and high-performing tritiles, based post facto on students' performances on the pre-test. Three divisions were chosen rather than two (high vs. low) to account for the transitional gains in knowledge about these processes, as seen in learning progression levels 2 and 3. This approach seems valuable when considering where high- vs. low-performing students fell in terms of current grade level: although there is a trend toward fewer low-performing students in the upper grades than in middle school, for instance, the performance tritiles were fairly evenly distributed across the grade bands (Table 4).

Subdivided apart in this way, it becomes clear that students with the lowest initial proficiencies benefited the most from this teaching intervention. Students in the lowest third initially gained two to four times more in their understanding than did students in the upper third, with low-performing upperclassmen gaining the most of any single group (Table 5). Further, ninth graders of any proficiency level consistently gained a good deal from this unit, with gains of at least 0.5 on the logit scale. Across the entire sample, starting proficiency did not explain much of the variability in learning gains (\sim 10%, see Fig. 3), but it did underline the trend that initially low-proficiency students stood to gain the most from this unit.

Results Carbon-Transforming Process

Student performance on items dealing with different carbon-transforming processes varied widely. Although the students overall demonstrated similar proficiency across the three processes by the post-test, they had started with a much weaker understanding of photosynthesis than of biosynthesis (i.e., digestion), and, to a lesser extent, cell respiration (See Table 6). Learning gains were statistically significant for all biological process dimensions. The largest learning gain was in photosynthesis, although learning gain differences between processes were not statistically significant. For example, a ninth grader from Michigan substantially improved their reasoning:

On Plant Growth Experiment (photosynthesis and respiration) on the pre-test, when describing
what is happening during photosynthesis in terms of gas exchange between plant and air—

Pre-test performance	Middle school (7th and 8th, $n = 216$)	Lower high school $(9th, n = 295)$	Upper high school $(10$ th -12 th, $n = 52)$
Lowest third	36.1%	31.9%	28.8%
Middle third	35.6%	31.5%	36.5%
Highest third	28.2%	36.6%	34.6%

Table 4Percentages of each of three grade categories whose students were grouped into three pre-test EAPcategories:low-, medium-, and high-performing

	Lowes	t third			Middle	third			Highes	t third		
	MS	LH	UH	Total	MS	LH	UH	Total	MS	LH	UH	Total
Mean gain	0.815	0.902	0.988	0.873	0.628	0.532	0.611	0.579	0.326	0.494	0.260	0.417

 Table 5
 Mean gain in EAP (i.e., after post-test; units are 'logit' scores) across students from the pre-test tritiles, subdivided by grade category (MS middle school, LH lower high school, UH upper high school)

Comparing the total values in each third, shows that students who were already fairly proficient on the pretest seemed to gain the least from the instructional intervention

"Each plant is taking in CO_2 and letting out O_2 ." By the post-test, this fundamental process was much more fully described—"In the dark, photosynthesis cannot occur because there is not light. The amount of CO_2 will increase because this process is not occurring."

 On Locations of Carbon (photosynthesis and biosynthesis) pre-test, when asked whether the leaves, wood, and roots of a plant would contain carbon and how it would get there simply "yes, because carbon is in every living thing." On the post-test, they answered with more nuance—"Carbon is in everything, but carbon gets to its leaves from the air. The leaves take in carbon through its stomata."

For all of the processes, and in keeping with the overall patterns, students with lower initial proficiencies gained the most in their understanding and students with the highest ability gained the least (Fig. 4). This effect was especially pronounced on items about biosynthesis, where high-performing students saw almost no improvement in their ability to explain biosynthesis, while low-performing students were much more proficient explaining that phenomenon than they had been.



Fig. 3 Comparing students' net performance gains as a function of their pre-test scores on the assessment. Although wide variation is present in this population of students, those least proficient on the pre-test typically made greater improvements by the time of the post-test

	Pre-test EAP	Post-test EAP	Learning gain	Standard error	Effect size
Photosynthesis	-1.046	-0.343	0.703	0.058	0.729
Biosynthesis	-0.560	-0.215	0.345	0.074	0.282
Cell Respiration	-0.794	-0.227	0.567	0.053	0.640
Mass	-1.060	-0.353	0.707	0.058	0.740
Energy	-0.870	-0.290	0.580	0.062	0.568
Subsystems	-0.687	-0.234	0.453	0.067	0.411
Large	-1.699	-0.502	1.197	0.045	1.608

 Table 6
 Overall pre- and post-test EAPs (units = logit scores) for all students for each process and key idea, along with their respective effect sizes

Effect sizes among the three processes are not significantly different, while among the key ideas, learning gains for large-scale thinking (bold) are significantly higher than those for the other key ideas

Key idea analysis

As with the carbon-transforming processes, student performances on items organized by the four key ideas also varied substantially. Average initial performances on mass and large-scale systems, in particular, were very low (Table 6), suggesting that students found these items more difficult than the items in other key ideas. For example, even many higher proficiency students struggled on the pre-test with large-scale reasoning:

• On a question about the fate of solar energy in photosynthesis (Energy in Plants), a ninth grade student from Colorado correctly stated that "it is changed into a



Fig. 4 Average proficiency gains among initially low-, medium-, and high-performing students on items about photosynthesis (PS), digestion and biosynthesis (BS), and cell respiration (CR). *Solid bars* show average gain by all students for each process

chemical energy". The same student dismissed the possible effect on atmospheric carbon concentrations from changes in fossil fuel pools (Carbon Cycle Rationale): "stored carbon is not a factor really; it just displays how much carbon we can potentially produce, not how much we add."

• An eighth grader from Colorado simply compared the number of processes moving carbon into or out of the atmosphere without any consideration of the relative scale of those processes: "There are more sources of CO₂ entering the atmosphere than coming down. This is most likely a cause of the CO₂ increase."

This confusion about carbon movement at large scales was often fairy persistent in students' reasoning. Here, a ninth grader from Michigan responds to the same item:

• On the pre-test, substantial confusion about the nature of fossil fuels—"Burning trees makes a lot of fossil fuels, and those are very bad." After instruction, this confusion remained—"I don't really get all of the numbers, but deforestation and burning is very harmful."

By the end of the unit, performances on all of the key ideas significantly improved at a 5% significance level. When the learning gains were compared across different key ideas, learning gains on large scale systems were significantly higher than other key ideas, However, because student reasoning on those two items had started out so low, final proficiency values were still lower in large-scale reasoning than in other key ideas, despite the larger learning gains. Further, all the students demonstrated similar learning gains on items about large-scale systems, regardless of their initial proficiency (Fig. 5). In contrast, high-performing students made lower overall gains in their understanding of sub-systems than did the lowest-performing students.



Fig. 5 Average proficiency gains among initially low-, medium-, and high-performing students on items built around understanding mass changes (Mass), energy transformations (Energy), sub-perceptible systems (Micro), and large-scale phenomena (Large). *Solid bars* show average gain by all students for each key idea

Level of Teaching Coverage and Assessment

Both in terms of teaching unit design and individual teachers' pedagogical choices, we knew that not all of the carbon-transforming processes and the key ideas would be equally emphasized in this research. In order to account for these differences, we combined the number of lessons in the unit that each process or practice played a role, in with the number of teachers that taught those lessons. Although many teachers taught the majority of the lessons, those focusing on biosynthesis and large-scale reasoning were less frequently taught than the other key ideas, both because they were featured in fewer of the lessons and fewer of the teachers taught those lessons in their treatment of the entire unit (Table 7). Both biosynthesis and large-scale systems also received less attention in units taught instruction than their relative prominence on the assessments might indicate. Subsystems, on the other hand, tended to be taught substantially more than its weighting on the assessments.

Discussion

Tracing matter through biological processes is notoriously difficult for students and common misconceptions are persistent (Mohan et al. 2009; O'Connell 2010). Our curriculum included specific inquiry-based activities designed to help students overcome naïve conceptions. These activities included carefully measuring the mass of plants as seeds and as seedlings, with concurrent measurements of the growing medium to verify that mass gain was not substantially due to intake of material from that source. This experiment was accompanied by observations of gas (CO_2 and O_2) exchange between the air and plants growing under both light and dark conditions. These activities were bracketed by explanatory discussions of the nature of chemical bonds and energy, basic plant biochemistry, and patterns of carbon cycling at global scales. Throughout the unit, several key reasoning tools were implemented, including the Matter and Energy Process tool that helped students to think through

 Table 7
 For each process and key idea, the number of lessons that featured content related to that variable (out of 11 lessons), the average number of teachers who taught each of those lessons (of 25 teachers), the proportion of total instructional intensity spent on each variable, and the proportion of assessment items that feature that variable

	No. of lessons	Mean teachers/lesson	% of total instruction focus	% of assessment focus
Photosynthesis	6	14	51%	43%
Biosynthesis	1	11	7	24
Cell respiration	5	14	42	33
Mass	9	14	43%	45%
Energy	5	15	25	23
Subsystems	6	14	28	14
Large-scale systems	1	10	4	18

Percentages sum to 100% for processes and key ideas independently

transformations of matter and energy, and a Powers of 10 tool that placed relevant objects on a scale of sizes.

The targeted instruction in this study helped students to reason in a more scientifically principled way through important carbon-transforming processes. This was particularly the case for the processes of plant respiration and photosynthesis. Misconceptions about metabolism and gas exchange in plants are established early in students' education and persist in older students (Canal 1999; O'Connell 2010; Özay & Öztaş 2003). The curriculum used in this project was geared heavily towards helping students to trace matter through photosynthesis and respiration. In addition to the scaffolded inquiry experiences, students repeatedly worked with a simple visual (the Matter and Energy Process Tool; Fig. 1a) to check their reasoning against the laws of conservation of matter and energy. Students in turn demonstrated significant learning gains in their reasoning about photosynthesis and respiration. Specifically, students were more likely to describe biological systems like plants as a network of interacting subsystems that together transform materials and energy in tandem. Students were also much less likely to conflate matter and energy after this instruction and more likely to attempt to explain where materials went after transformations, rather than just explaining that they "went away."

The extent of improvement in photosynthesis and respiration varied dramatically with several factors, including the grade level and, more specifically, the initial proficiency of the students with the material. Lower-proficiency students generally gained more in their understanding than their higher proficiency peers, regardless of grade level, with ninth graders in particular evidencing large gains as a result of the unit. These differences among students may be due in part to the process-and-key idea framework that underlies both the unit and the assessments. Even for students with greater detailed knowledge of these processes, the emphasis on the same key ideas extending across all processes helped them to elucidate the importance of principles (e.g., conservation of matter and energy) regardless of the system. Students generally receive earlier and more frequent exposure in school to the mechanisms of growth (biosynthesis) and animal metabolism (i.e., cell respiration) than they do to specific mechanisms of photosynthesis (beyond the general "plant needs for growth"). This is reflected by pre-test abilities on the former two processes being much higher than those for the latter. By the latter part of high school, students have generally experienced more detailed instruction about photosynthesis, and thus stand to gain less from a unit focused largely on that process. In addition, older students, who have usually had more chemistry than younger students, are less likely not helped as much by instruction that includes molecular models to help students think about movements of carbon (i.e., mass) through these processes. This is seen in their low levels of improvement in the micro-practice compared to lower-proficiency students (Singer et al. 2003). In other words, our assessment system, which depended on a four-level rating scheme, likely creates a saturation effect for learners who begin near the top of that scale, since providing a more sophisticated answer on the post-test would not necessarily result in a much improved score if their pre-test answer already included acknowledgement of the key principles. Our results do strongly indicate that students who can pair prior instruction on details relevant to biological processes (especially a functional atomic-molecular understanding) with improved principled reasoning, stand to gain a great deal from this type of instruction. This can be seen, for example, in greatly improved reasoning for upper-class high school students who started out with

low pre-test scores, as compared to their classmates who were already reasoning at a high level.

With a majority of the teaching emphasis falling on photosynthesis and cell respiration, it is perhaps not surprising that students gained relatively little in their understanding of biosynthesis (which was a focus of instruction only in lesson 10). former two key ideas are required in multiple years' standards in most states, but biosynthesis does not get covered until later in high school and focuses mostly on digestive processes, not biosynthesis per se (the actual chemical rearrangement of biological macromolecules within organisms) (Assaraf et al. 2013; Banet & Núñez 1997). By helping students to think generally about how matter and energy remain coupled during biosynthesis, rather than joined or separated as in photosynthesis and cell respiration, this unit helped less proficient students reason better in this Area. The approach had little effect on more proficient students, who had apparently already gathered this basic idea. Although much more specific instruction about biosynthesis (potentially including more direct consideration of the structure and energetics of crucial molecules in living things) would be necessary to really boost student abilities in this area, it is promising that all students saw at least some gain in their reasoning even with a relatively modest emphasis in this unit. This is further evidence that the reasoning habits about key ideas the unit sought to cultivate in students, do help to build transferable reasoning patterns, that are useful across multiple processes for many students.

Students achieved higher learning gains on items about energy than on microscopic-scale items, although energy was taught directly less frequently than micro. A possible explanation of this pattern is that a series of common misconceptions stem from matter-energy confusion (e.g., food is "burned up" by metabolism, sun's energy is turned into sugar by plants, etc.). When students were learning about matter, they were learning how to trace matter separately from energy. Thus, although these activities did not explicitly address energy transformations, they did help students differentiate matter transformations from energy transformations, contributing to significant learning gains for energy in the process. Similarly, for many students, especially younger ones, more background in chemistry may be necessary before they can demonstrate larger gains in micro-scale reasoning. One set of tools that many instructors and researchers have found helpful in this regard is the use of manipulable molecular models which, help students to consistently conserve matter (atoms) across chemical reactions. Although we encouraged participating teachers to use these tools, if they had them, not all teachers ended up applying them to this curriculum.

Interestingly, students had the largest learning gains on the large-scale practice, even though that practice was by far the least taught practice across activities and teachers. This effect could be due simply to the fact that only two assessment items measured students' reasoning in this practice, so the gains could be largely statistical anomalies. But, another possibility is that most students had little initial understanding of large-scale movements of carbon (shown in their very low pre-test abilities), and thus any attention to these concepts was likely to bear larger fruit in terms of their reasoning. This possibility also reinforces the interconnected nature of the practices: improvement in one is unlikely to persist without concurrent improvement in. Others at the same time is likely to help students improve their reasoning in those other areas. This interconnection also allows students across a range of introductory proficiencies to productively engage with a curriculum its flexibility allows teachers to tailor much of the instruction to the level of understanding that their students already have while, also helping students with more knowledge of details to check that their understanding does not violate the fundamental principles involved.

A related explanation for some of the findings may be a difference between "constraint reasoning" and "connecting reasoning." Constraints such as the conservation of matter and conservation and degradation of energy can become important reasoning checks for students to test the feasibility of their own thinking about these processes. This type of reasoning was heavily emphasized in our instructional design and would help students to reason more effectively in all of the practices. Another skill emphasized in our instruction is the ability to connect similar processes across multiple scales. For example, the ability to articulate that global scale movement of carbon from atmospheric pools to the biosphere is essentially happening through the organismal-scale process of photosynthesis. This understanding reflects a high level of understanding across scales. It is possible that students who had initial difficulty with large-scale reasoning benefitted from the emphasis on conservation-based reasoning at the organismal scale combined with our activities that focused on connecting processes across scales. We might expect energy-focused items to be difficult for a similar reason, since it requires bridging molecular scales (chemical potential energy) with much larger scales, but our current assessments did not bear this out. This is likely due to the nature of our energy items, which did not actually require this complete cross-scale reasoning to be coded as a high-reasoning (levels 3 or 4) response.

In order to get a better handle on these differences, next steps could include construction of new items that more effectively tease apart these differences among processes and key ideas and within key ideas. In particular, the current items are somewhat limited in their ability to draw out assessable learning gains for students who were already fairly proficient at the start of the unit, even though their teachers affirmed the value of this approach for their students (improvements in the consistency of upper-level students' reasoning may have occurred but were not detectable with our assessment instruments due to a saturation effect). In addition, closer study of the actual teaching used in classrooms and their correlations with student reasoning performances would help to determine the veracity of any of these hypotheses. We also expect that the teachers' content knowledge and familiarity with the materials (i.e., pedagogical content knowledge) can strongly affect student reasoning performances.

Overall, we found that students could be assisted to build better understandings of important, complex, and traditionally bedeviling biological processes, through design of teaching materials that strongly incorporated several key principles: conservation and connection. Through a combination of targeted inquiry investigations and recurring use of visual reasoning tools to highlight key principles, students were better able to articulate their understanding of photosynthesis and respiration in a way that avoided many of the common misconceptions generally held about these processes.

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Compliance with Ethical Standards

Conflict of Interest The authors declare that they have no conflict of interest.

Appendix 1				
Table 8 The learning	progression			
	Level 1	Level 2	Level 3	Level 4
Explaining key ideas	Force-dynamic accounts	Hidden processes	Matter transmutation; matter-energy conversion	Matter transformation and energy transformation
Explaining mass	Changes based on perceptions and feelings Describe changes based on perceptions. For example, sunlight warms the tree. Plants need air, so that they will not suffocate.	Changes in "stuff" Explain events in terms of the following hidden mechanisms: 1) Changes involving only solids and liquids: Plants gaining weight from soil. 2) Changes involving only gases: CO ₂ and O ₂ conversion. 3) Material creation: Plants create mass when they have what they need. 4) Matter transmutation: Plants mix sulight, water, soil, end of a transmutation:	Changes involving organic molecules Explain events in terms of conversion between energy and organic molecules (e.g., Glucose molecule turns into kinetic energy) Explain events in terms of transmutation at the atomic-molecular scale: (e.g., Glucose molecular scale: (e.g., divoide and water. Do not identify oxygen.)	Atom re-arrangement Explain events in terms of matter transformation, and describe matter transformation as atom re-arranged to form new prod- ucts in chemical reactions. Therefore, the mass of different types of atoms is conserved.
Explaining energy	Cause-effect relation Describe cause and effect relation. Two causes (i.e., there is an actor such as plants, animals, and people; the actor has all its enablers) lead to one effect (i.e., the actor moves/grows)	and all togetter to triate toou. Tracing energy backward not forward Explain that energy powers hidden processes. It is often used up or becomes waste materials. Energy is a type of semi-matter. It can be created or used up.	Mass-energy conservation Use modified principles to explain events: Energy cannot be created or destroyed, but it can be converted into/from matter. Treat organic molecules as energy.	Energy transformation Explain events in terms of energy transformation: Energy transforms from one form to other forms; from one form to the form from one form to the form process. Recognize that organic molecules contain C–C and C–H bonds and that they provide energy in chemical reactions
Explaining large-scale systems	Sequence of events Describe sequences of events (e.g., plants grow,	Connecting hidden processes Describing solids and liquids moving in food chains/webs.	Connecting processes in terms of atoms and molecules	Carbon cycle and energy flow Explain carbon cycles in terms of transformation between

Level 1 and then animals eat pl				
and then animals eat pl	Level 2		Level 3	Level 4
	Describe "gas cycle" and animals/hume	" between plants ans	Explain carbon cycle as carbon atoms moving in food chains/webs without chemical reaction involved; Ex- plain energy cycle instead of energy flow	organic and inorganic carbon; Explain energy flows with recognition of heat dissipation
Explaining Macroscopic Observat microscopic Describe objects, orga sub-systems or parts of them tha observed at the macroscopic scale	vations Cellular structure ganisms, Describe sub-system that can be of living and nonl (e.g., cells, particl	s or particles living things es, etc.)	Atomic-molecular structure Recognize that organisms and materials are all made of atoms and molecules; Identify carbon and organic molecules in living things	Associating C-C and C-H bonds with energy Recognize that organic molecules such as glucose provide energy because they contain C-C and C-H bonds

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Appendix 2. A Coding Rubric for an Item about Energy in Photosynthesis

Here is an example of our rubric system for an item asking students to distinguish which things (water, light, air, soil nutrients) provide energy for plant growth. It is an item about energy (i.e., a key idea) in photosynthesis (i.e., a carbon-transforming process):

(Question text: "energy for plants") Which of the following is(are) energy source(s) for plants? Circle yes or no for each of the following.

a. Water	YES	NO
b. Light	YES	NO
c. Air	YES	NO
d. Nutrients in soil	YES	NO
e. Plants make their own energy.	YES	NO

Please explain ALL your answers.

Level	Indicators	Example response
4	1. identify light as the external	"Water, air, nutrients in soil, and plants
(Upper	energy source for plants AND	making their own energy are not energy
Anchor)	2. may choose others as long as	sources. They are products that have
	they clearly avoid converting	energy in them light is a source of energy
	matter into energy	and it is a part of photosynthesis which
		plants use"
Difference	Level 4 responses clearly desci	ribe light as the only energy source, while
b/w 3 & 4	level 3 sometimes inc	cludes others as energy sources.
3	1. identify plants as making	"Water is an energy source for plants
	their own energy through	because it allows photosynthesis to take
	photosynthesis OR	place. The same is true for light and air
	2. works with conservation of	they allow photosynthesis . Nutrients in
	energy, but does so incorrectly	the soil I feel don't aid in giving the plant
		energy. Through photosynthesis plants
		produce glucose which serves as there
		source of energy."
Difference	Level 3 responses begin to disti	inguish based on matter and energy, often
b/w 2 & 3	through role in photos	synthesis, while Level 2 does not.
2	1. explain how different things	"the reason that water, light, and the
	support different plant	nutrients in the are energy sources fore
	functions OR	plants is because they all have plants is
	2. distinguish among needs	because they all have plant vitamins. The
	based on some hidden	reason that air is not a energy source for
	mechanism (e.g. vitamins) OR	plants is because it doesn't carry plant
	3. explain plants make own	vitamins"
	food without tracing through	
	to glucose or photosynthesis	
Difference	Level 2 responses describe some	e sort of unseen mechanism as key to
b/w1&2	growth; level 1 focuses on how t	hings help plants to survive.
1	1. explanations for growth	"A plant is just like a human they need
	based on human-like analogy	water, light, air , and nutrients"
	OR	
	2. focuses on needs of plant to	

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