BIO-RAD Explorer™

STANDARDS BASED

Photosynthesis and Cellular Respiration Kit

A ThINQ!™ Investigation

Catalog #12005534EDU

General Biology | **Instructor's Guide**

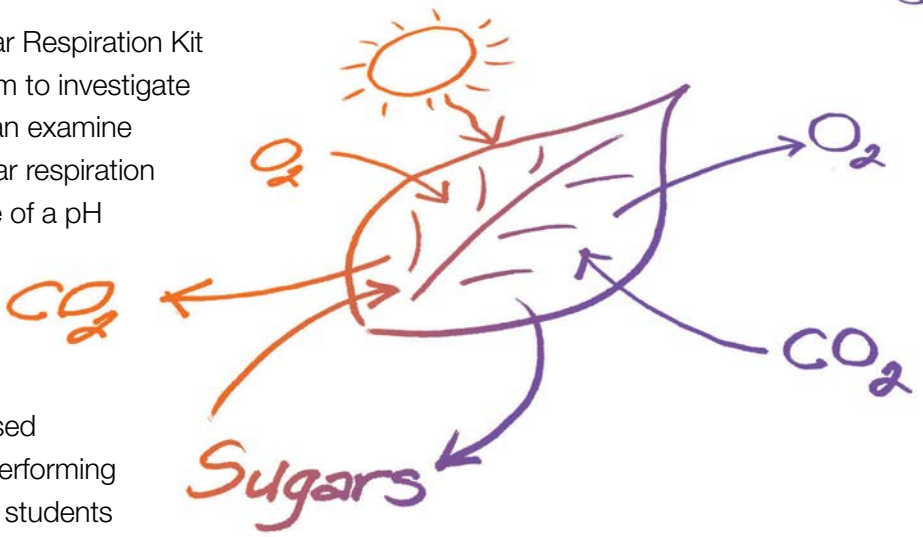
BIO-RAD

Dear Instructor

Thank you for inspiring the next generation of scientists, citizens, and decision makers to be curious about the world around them. Our goal is to provide you with tools to help your students think like real scientists.

This inquiry-based laboratory curriculum guides students through the scientific process of observing and asking questions about an engaging phenomenon, generating and revising models, planning and executing experiments, and analyzing data. The student manual poses a series of questions to focus and stimulate thinking about all aspects of the investigation. To facilitate the teacher's role, explanations and lesson plans are included in the instructor's guide.

The Photosynthesis and Cellular Respiration Kit uses algae as a model organism to investigate cellular processes. Students can examine both photosynthesis and cellular respiration simultaneously through the use of a pH indicator solution that changes color depending on (or in response to) the amount of CO_2 consumed during photosynthesis or released during cellular respiration. By performing the assay in a single organism, students are able to visually connect the two processes.



We strive to continually improve our curriculum and products, and your input is extremely important to us. We welcome your stories, comments, and suggestions.

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Contents

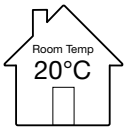
Kit Storage

When you receive the Photosynthesis and Cellular Respiration Kit:

1. Use a permanent marker to write the control numbers (found on the labels) and storage locations of the kit box and the Reagent Refill Pack on the front page of the Quick Start Guide. Also write the storage location of the Reagent Refill Pack on the kit box.

2. Put the reagent bag in the fridge (4°C). DO NOT FREEZE.

All other components can be stored at room temperature (RT).



3. Visit bio-rad.com/genbioalgae to download the Instructor's Guide, Student Guide, Standards Alignments, and additional resources.

Important Notes

- Begin the laboratory preparation at least 1 day before Lesson #1 and at least 3 days before Lesson #3. See p. 15 for further instructions
- **Always use distilled water for this kit**
  Tap water contains chlorine and will kill the algae

- **Technical support** is available at support@bio-rad.com or 1-800-4BIORAD, option 2

Safety Guidelines

Some countries outside the U.S. may require a special license to use this kit. Hawaii requires an application for a license with the state's Department of Agriculture to obtain permission to import algae. Please refer to your state's or country's legislative authorities for proper guidelines. Please obtain the Safety Data Sheets (SDS), which are available from Bio-Rad by calling 1-800-4BIORAD in the U.S. Visit bio-rad.com/genbioalgae for further information on reagents in this kit. Please consult your local environmental health and safety regulations for information on proper disposal of used items from this kit.

Algae

The strain of algae used in this kit, *Scenedesmus obliquus*, is a green freshwater unicellular alga that is commonly found in clean ponds, lakes, and rivers worldwide and is not pathogenic. Nonetheless, handling the *S. obliquus* strain requires use of standard microbiological practices, including but not limited to the following:

- Decontaminate work surfaces once a day and after any spill of living material
- Decontaminate all liquid or solid wastes before disposal
- All persons must wash their hands after they handle material containing algae and before exiting the laboratory
- Perform all procedures carefully to minimize creation of aerosols
- Use only mechanical pipetting devices; mouth pipetting is prohibited
- Eating, drinking, smoking, and applying cosmetics are not permitted in the work area
- Wearing protective eyewear and gloves is strongly recommended
- If an autoclave is not available, place algae beads and all solutions and components (transfer pipets, cuvettes, slides, and coverslips, etc.) that have come in contact with the algae beads in a fresh 10% bleach solution for at least 20 min to decontaminate.

Kit Components and Ordering Information

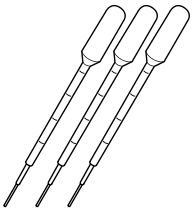
Each kit contains materials to outfit 24 student workstations.

Kit Components	Quantity
Algae beads	≥170 beads
10x CO_2 indicator solution	50 ml
Debeading solution	20 ml
0.2 ml PCR tubes with domed lids	150
Disposable plastic transfer pipets, sterile	60
Indicator Color Guide Sheets	3
Instructor's Answer Guide	1
Quick Start Guide	1

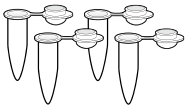
Required Materials (not included in this kit)	Quantity
For Lesson #1	
Clear plastic cups or conical tubes	2 per group
Plastic straws	1 per group
For Lesson #2	
(optional) Microscope slide and coverslip	1
(optional) Microscope	1
For Lesson #3	
Aluminum foil, 5" x 5"	1 per group, extra for prep
Plastic wrap or Parafilm	5 x 5"
Beaker, 150–250 ml	2
Beaker, 250–500 ml	1
Graduated cylinders, 250 ml and 25 ml	1 each
Distilled water	1 L
Lamp fitted with 60–100 W bulb	1 per group
Clock or timer	1 per group
Permanent marking pens	1 per group
Scissors	1 per group
Bleach (household variety), diluted to 10% solution	10 ml

Ordering Information

12005534EDU	**Photosynthesis and Cellular Respiration Kit for General Biology**, includes algae beads, 10x CO_2 indicator solution, debeading solution, PCR tubes, disposable plastic transfer pipets, indicator color guides, answer key. Instructor and student guides available for download.
17001238EDU	**Photosynthesis and Cellular Respiration Kit for AP Biology**, includes algae beads, 10x CO_2 indicator solution, debeading solution, disposable plastic cuvettes and caps, disposable plastic transfer pipets, indicator color guides, printed instructor's guide. Student guide available for download.
12002353EDU	**Photosynthesis and Cellular Respiration Reagent Refill Pack**, includes algae beads, 10x CO_2 indicator solution, debeading solution
1660480EDU	**Disposable Plastic Transfer Pipets**, nonsterile, 500
TWI0201EDU	**0.2 ml PCR tubes with domed caps**, pk of 1,000

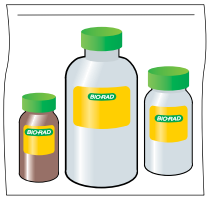


Disposable plastic transfer pipets

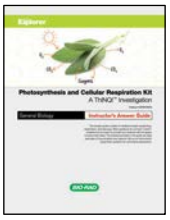


PCR tubes

Indicator color guides



Algae beads, 10x CO_2 indicator solution, and debeading solution



Instructor's Answer Guide

Tips & Tricks

For Lesson #1 students use straws to blow into cups containing indicator solution. To prevent spilling, students should blow slowly and carefully through the straws. Using larger clear plastic cups may be helpful to prevent spills.

By cutting the bulb portion off of a disposable plastic transfer pipet, students can use the stem portion as a straw during Lesson #1.

Tips & Tricks

For detailed information about lamp selection, see p. 11.

Using cell phone lights

Cell phone lights can be used during the in-class activity in place of a lamp and clock. However, they tend to have low light intensities resulting in slower color change. The closer algae bead samples are to the light source, the faster indicator color change will be observed.

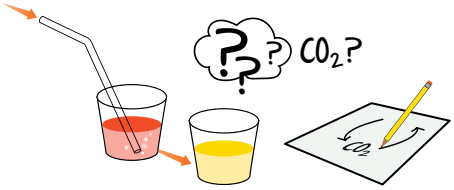
Kit Activity Overview

Lesson 1

Modeling the Inputs and Outputs of Photosynthesis and Cellular Respiration

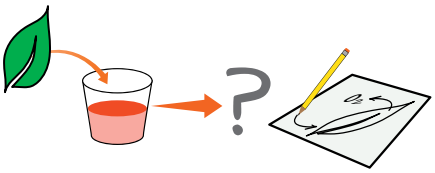
Observing indicator color change

Students observe a color change in the pH indicator after blowing into it. They create and revise models to represent what they observed and then discuss the causes.



Linking photosynthesis and cellular respiration

Students consider how a plant would affect the color of the pH indicator and create a new model to represent their predictions.

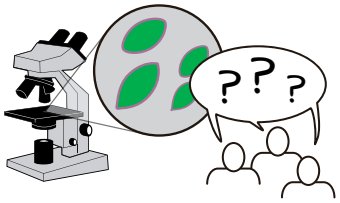


Lesson 2

Experimental design with algae beads

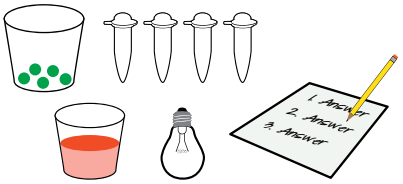
Using algae beads as a model organism

Algae is introduced to students as a model organism for measuring photosynthesis and cellular respiration in plants. Students view algae beads and example images of algae under the microscope. Students develop their own investigation questions that can be answered using algae beads and available materials.



Planning investigations with algae beads

Students design an experiment to collect evidence to help answer their investigation questions.

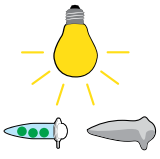


Lesson 3

Photosynthesis and Cellular Respiration Investigations

Conducting the experiments and analyzing results

Students carry out their experiments. They make claims using their experimental evidence to answer their investigation questions and write scientific explanations for their observations.



Post-laboratory questions

Optional post-laboratory questions guide students to expand their previous models to include heterotrophs.

Roadmap to the Instructor's and Student Guides

The **Instructor's Guide** is written to provide you, the instructor, with sufficient background information and planning tools for you to facilitate student inquiry and experimental design. It is designed to be used with or without the Student Guide and provides all the information and lesson details necessary to complete all activities. The Instructor's Guide includes:

- An **overview of the kit** with information about how the kit activities have been designed to support student learning and questions to help you guide your students
- A **focus on modeling** in the classroom that includes a brief introduction to the science practice of modeling and how it is implemented in this kit
- **Keys to success with algae beads** with care and background information about the algae beads as well as a section on how to support successful student experiments
- **Timelines** and **Advance Preparation Instructions**
- **Lesson plans** with step-by-step classroom instructions that can be used to prepare for instruction or in class during the lesson. To the left of certain lesson plan steps, there numbers that correspond to questions in the Student Guide. As you proceed through the lesson plan steps, you may have your students write answers to those corresponding questions in the Student Guide.

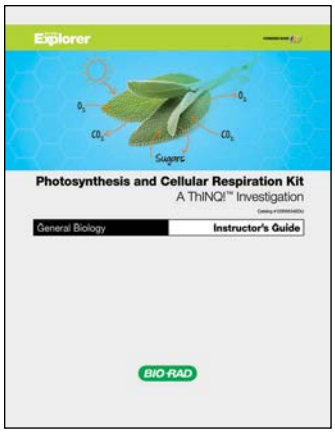
Question numbers in the Student Guide that correspond to the instructor lesson step.

1. ***Have students observe and describe the indicator solution.*** Record students' thinking on the board as a class and/or have them do so in their notebooks. Students do not yet know the function of the carbon dioxide

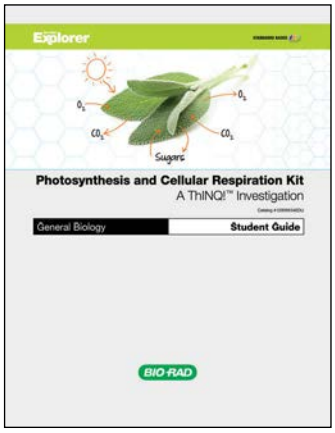
- **Appendices** with supplemental protocols, optional student background information, and FAQs

The **Student Guide** includes a minimal framework of questions and information to guide student thinking and discussion. In-depth instructions and background are intentionally omitted or found only in the instructor's Guide to foster the discovery of knowledge instead of rote memorization. Additional background on photosynthesis and cellular respiration can be found in Appendix D.

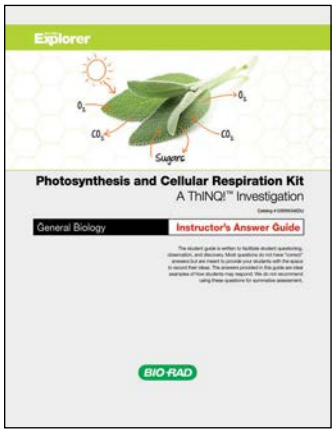
The Instructor's Answer Guide includes ideal examples of how students may respond to the questions presented in the Student Guide. However, most of the questions in the Student Guide do not have "correct" answers and are provided to your students as space to record their ideas.



Instructor's Guide



Student Guide



Instructor's Answer Guide

Curriculum Fit and Inquiry Support

Required prior knowledge

- Photosynthesis and cellular respiration are processes by which plants and animals generate and use energy
- The inputs of photosynthesis are light, carbon dioxide, and water. The outputs of photosynthesis include oxygen and glucose
- The inputs of cellular respiration are oxygen and glucose. The outputs of cellular respiration include carbon dioxide and water
- pH is a numerical indication of the acidity or basicity of a solution
- pH indicators change color in response to pH changes of a solution.

Curriculum fit and topic connections

- Photosynthesis and Cellular Respiration — the basic inputs and outputs of photosynthesis and cellular respiration are the heart of this kit
- Cellular Structures and Processes — specialized organelles house the complex metabolic and biochemical pathways of photosynthesis and cellular respiration. Make connections to other processes and organelles that are crucial to cell function
- Ecology and the Interdependence of Organisms — algae is both a model organism for plants and an important element of many ecosystems. Discuss how a bloom of algae affects the proliferation of other organisms within ecosystems
- Flow of Energy and Matter — what is produced by one process is used by another in a constant cycle. Create energy and matter maps at the molecular, cellular, and organismal levels to discuss how they're all connected
- Environmental Science — the chemistry of carbonic acid that allows photosynthesis and cellular respiration to be monitored in this kit is the same chemistry involved in ocean acidification as a result of carbon dioxide in the atmosphere

Standards Alignment

Visit bio-rad.com/genbioalgae for more information about standards alignment.

Incorporating the next generation of science education practices

The lessons in the Photosynthesis and Cellular Respiration Kit were designed to support three-dimensional learning as described by the National Research Council in *A Framework for K-12 Science Education* (2012, Washington, DC: National Academies Press). At the beginning of each lesson you will find brief explanations of how these dimensions, Science and Engineering Practices, Disciplinary Core Ideas, and Crosscuting Concepts, are integrated into the actvities.

Science & Engineering Practices

Science Practices are the fundamental methods that scientists use to attain knowledge and engineers use to solve complex problems. Engaging students in these practices enriches their understanding of the content itself and helps them understand how knowledge is gained. The lessons in this kit emphasize multiple scientific practices.

Disciplinary Core Ideas

Disciplinary Core Ideas represent fundamental content knowledge from specific scientific content areas.

Crosscutting Concepts

Crosscutting Concepts are ideas that span scientific disciplines and shape how science knowledge is understood and described. Crosscutting concepts are highlighted in the Instructor's Guide as a reminder of times to help your students recognize the connections across disciplines.

Engaging students in scientific questioning

The Photosynthesis and Cellular Respiration Kit for General Biology focuses on three major learning phases: exploration, knowledge construction, and application/reflection. You may find it useful to prompt students with the questions below in order to engage them in each phase. These questions can be used with your students at any point in the kit curriculum and may guide your thinking as you support your students' learning about photosynthesis and cellular respiration.

Lab Phase	Suggested Questions and Prompts to Support Student Learning and Discussion	Kit Specific Applications
Exploration	**Making observations that lead to an investigation question** What did you notice about the color of the solution when you blew into it using the straw? What might explain the change in color? What are you blowing into the solution that could cause a color change? What might happen if you add a plant to the solution? Would the color change? Why or why not?	Observing color changes in the CO_2 indicator solution
Knowledge Construction	**Clarifying the purpose of the core lab** What do you already know about photosynthesis (PS) and cellular respiration (CR)? How does your knowledge about PS and CR help you describe the phenomenon that you observed (color change in the indicator solution)? How do the processes of PS and CR relate to one another, if at all? What questions do you have about PS and CR and how would you use the materials available to you to test your ideas? What evidence would you need in order to answer your questions?	Modeling inputs and outputs of each process (for example, light, CO_2, sugar, ATP)
Application/ Reflection	**Analyzing and interpreting evidence** What is the investigation question? How does the evidence support your model of PS and CR? What changes need to be made to your model, if any? What justifications can you provide to support what counts as evidence in this investigation?	Drawing conclusions based on evidence and expanding models to include new information.

Example investigation questions

The activities in this kit help students develop investigation questions and design their own experiments through open inquiry to answer them. Below are a few example real-world connections and investigation questions that can be answered using the materials provided in this kit with minimal additional materials.

Cellular respiration in plants

Observation(s): Plants survive at night; plants produce glucose during photosynthesis

Investigation question: Do plants perform cellular respiration?

Student experiment: Place an algae bead sample in the dark for 30 minutes. Check for CO_2 indicator color change as evidence of cellular respiration.

Effects of light intensity

Observation(s): Plants do not grow indoors as well as outside; fewer plants grow on the forest floor among dense trees where light is filtered.

Investigation question: How does light intensity affect plant growth?

Student experiment: Place multiple samples of algae beads at different distances from a light source. Collect measurements every few minutes and compare the relative rates of photosynthesis for each sample.

Effects of light color

Observation(s): Plant leaves are usually green; red algae grow deeper in the ocean than green algae, which grow in shallow water. Organisms that are deeper in the ocean receive more blue light than red light because red light is filtered more by water than blue light.

Investigation question: Which colors of light are the most important for photosynthesis?

Student experiment: Use colored filters to expose algae bead samples to different light colors. Compare the relative rates of photosynthesis for each sample.

Effects of temperature

Observation: Desert plants do not grow well in cold climates even with the same rainfall.

Investigation question(s): What effect does temperature have on photosynthesis and/or cellular respiration? Are there ideal temperatures that support photosynthesis and cellular respiration?

Student experiment: Float algae bead sample tubes in water baths with different temperatures. Place both under the same light. Compare the relative rates of photosynthesis and/or cellular respiration for each sample.

Tips & Tricks

Using structured inquiry

If you would prefer to use structured inquiry with your students, a full experimental procedure for exploring photosynthesis and cellular respiration in algae is provided in Appendix A.

A Focus on Modeling

What is modeling?

Creating models is a way to organize one's current understanding of events or systems through representations. It is also an important science practice. Modeling can take many forms, including three-dimensional constructions, drawings, and computer simulations, and can be used for multiple purposes in the classroom:

- Organizing student thoughts — creating a model helps to make students' own ideas more concrete and reveals areas of contradiction
- Making predictions — students can use their models to make predictions about future events or in new situations. A hallmark of a good model is one that provides enough detail to make such predictions but is simple enough to be easily understood
- Checking for understanding — student models provide an excellent opportunity for you to check their understanding and to ask guiding questions

How is modeling incorporated in this kit?

In this kit, students have multiple opportunities to create, revise, and extend drawn models based on their observations and reasoning. At each stage of evision, models should become more sophisticated and include more relevant information. Avoid correcting models that initially contain errors. Instead, help students revise them using new observations and information.

An example model development progression

1. **First model.** In Lesson #1 students draw models of the carbon dioxide indicator solution changing colors. Models may be very simple and include only basic observations. Encourage students to revise their models to include details that help to explain the phenomenon.

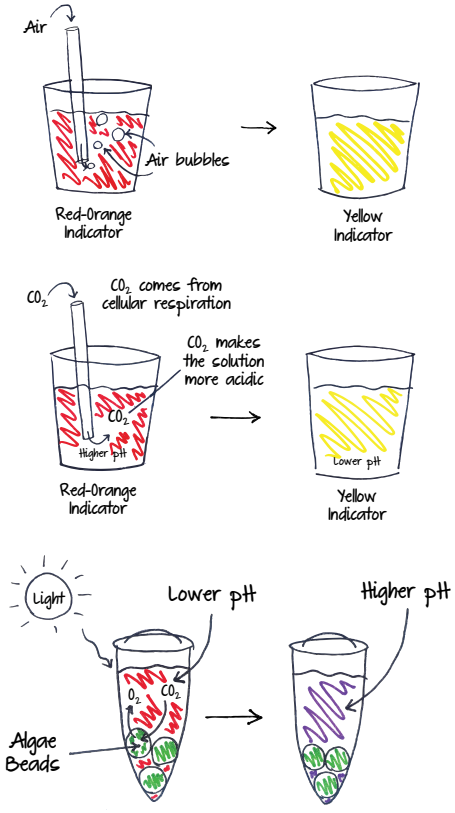
2. **Revised model.** Following a class discussion about pH, carbon dioxide, and cellular respiration, student models should include new information and demonstrate more interactions and relationships between the relevant components of the system. At this stage, students should be able to use their models to make predictions.

3. **New model.** Later in the lesson sequence, students will become familiar with algae beads and will discuss photosynthesis. They will draw new but related models that describe photosynthesis in algae beads. These student models should build on their previous models, and extend to include new concepts.

Science & Engineering Practices

Modeling

Students will engage in modeling in each lesson. Refer to the blue Science & Engineering Practices callout boxes at the beginning of each lesson for an overview of how modeling is used in that lesson.



Keys to Success with Algae Beads

What are algae beads?

Algae beads contain thousands of individual algae cells immobilized in sodium alginate to make them easy to handle. The beads are permeable to both solutions and gases including oxygen and carbon dioxide, which will diffuse in and out. Therefore, the algae can photosynthesize and respire within the alginate matrix.

How long will algae beads last?

Before activation, the algae beads will survive for at least 3 months at 4°C in the storage bottle. After activation, the beads will survive for at least 2 weeks in storage solution at room temperature and ambient lighting.

What kind of algae are in the beads?

The strain of algae contained in algae beads, *Scenedesmus obliquus*, is a green freshwater unicellular alga that is commonly found in clean ponds, lakes, and rivers worldwide and is not pathogenic.

What is the CO_2 indicator solution?

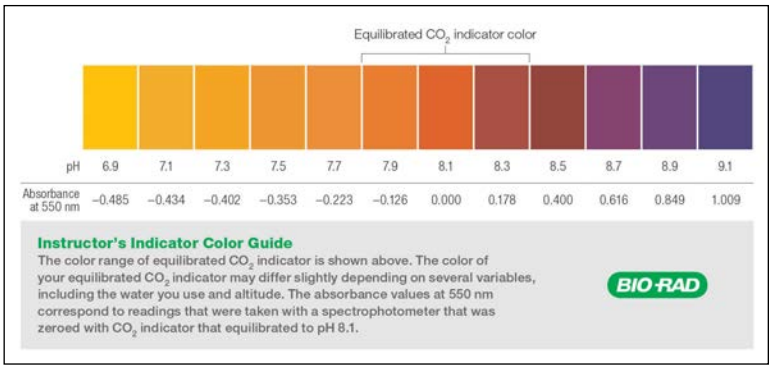
The CO_2 indicator in Photosynthesis and Cellular Respiration Kits is a mix of pH indicator dyes that allows students to track photosynthesis by CO_2 consumption and cellular respiration by CO_2 production. The color of the CO_2 indicator solution is sensitive to changes in pH caused by gaseous CO_2 dissolving in water to form carbonic acid according to this equilibrium:

$$CO_2 + H_2O \leftrightarrow H_2CO_3 \leftrightarrow HCO_3^- + H^+$$

As algae beads consume or produce CO_2 through photosynthesis and cellular respiration, respectively, the pH of the CO_2 indicator solution changes, causing an observable shift in its color. The full range of expected colors for the CO_2 indicator solution can be seen on the Indicator Color Guides provided in the kit and shown below.

After preparing the 1x CO_2 indicator solution, it must equilibrate overnight to atmospheric levels of CO_2 which will bring the pH to approximately 7.9–8.3, a reddish-orange color.

The pH range that can be measured by the indicator solution is approximately 6.9–9.1. During an experiment, the pH of the solution may change beyond this range, but no additional color change can be observed. That does not indicate that photosynthesis and/or cellular respiration have stopped, but the limits of the indicator solution have been reached.



Do's and don'ts of algae bead care

Don't use tap water

Chlorine in tap water will kill the algae. Distilled water is best.

Don't overheat algae beads

The algae are adapted to live at 30°C. Some lamps can give off too much heat and will overheat the algae beads during activation. As a general guideline, if the surface where the algae beads are being activated feels at all warm, then it is too warm for the algae to survive.

Don't freeze the algae beads

The algae WILL NOT tolerate freezing.

Do store refrigerated at 4°C

The algae can be stored at room temperature, but will last longer if stored at 4°C.

Do store algae beads in the storage solution

Algae beads should always be stored submerged to prevent them from drying out. When storing the algae beads longer than 24 hours, reuse the storage solution in which the algae beads are shipped. It is optimized to keep the beads functioning at their best. If you do not have the storage solution, use 1x CO_2 Indicator solution.

Do give algae beads both light AND dark

Algae, like plants, are accustomed to a daily light and dark cycle. For long-term care of the algae beads, be sure to provide a daily cycle of both light AND dark. Use a lamp for this purpose. See p. 11 for information on lamp selection.

Observing photosynthesis and cellular respiration using algae beads

Balancing observations of photosynthesis and cellular respiration

Algae are always performing a baseline level of cellular respiration, whether in the dark or in the light. However, when exposed to direct light the algae will ramp up their capacity to do photosynthesis, which will quickly become the dominant process. It can be difficult to detect cellular respiration when photosynthesis is very active.

The instructions and timing for activating and resting the algae beads will balance the processes so that students can observe both within the same class period. See the Advance Preparation Instructions for further details.

The importance of initial acclimation, activation, and resting

When you receive your algae beads, they are dormant and require a gentle acclimation to room temperature and ambient light followed by a long exposure to direct light to re-engage photosynthesis (activation). Activation is followed by a resting period in ambient light to slow and balance photosynthesis with cellular respiration as described in the above section. Following these steps, the algae beads will perform both processes quickly enough for students to observe them during a single 50-minute class period.

If you would like to speed up student observation of either photosynthesis or cellular respiration, then reduce or increase the resting period by several hours, respectively. For example, if you choose to have students observe only photosynthesis in one class period, do not rest the algae beads. This will keep photosynthesis highly active for student observation. Afterward, rest the beads in ambient light overnight (6–24 hr) and they will be ready for students to observe cellular respiration.

Reusing algae beads

Reusing algae beads in back-to-back classes

We do not recommend reusing the same algae beads for multiple class periods in the same day because the balance between photosynthesis and cellular respiration is disrupted during the experiments. Use freshly activated and rested algae beads for each class period on the same day.

Using algae beads for classes on subsequent day

For use in multiple class periods on subsequent days, we recommend activating and resting the algae beads in batches, one batch for each class period.

Activate the needed quantity of algae beads for your first day of classes. When the first batch of algae beads are resting, begin the activation for the next batch. Continue this process as needed.

Reusing algae beads

Algae beads can be reused multiple times when provided with good care and preparation. Algae beads that were exposed to direct light during student experiments should rest for 6–24 hours in ambient light while those that were put in the dark should be re-activated and rested before reuse. For simplicity, we recommend pooling all used algae beads each day and performing an activation and rest to restore the balance between photosynthesis and cellular respiration.

Successful Experiments with Algae Beads

Typical experiment setup

Adjust the lamp so that the bulb is 15–25 cm from and directly above the lab bench or table surface. This reduces the amount of heat on the beads while maintaining sufficient light intensity to quickly observe color change. However, dimmer or hotter lamps should be arranged closer to or farther from the surface, respectively, to ensure algae bead function. Be sure to test these variables prior to performing the overnight activation or in-class activity.

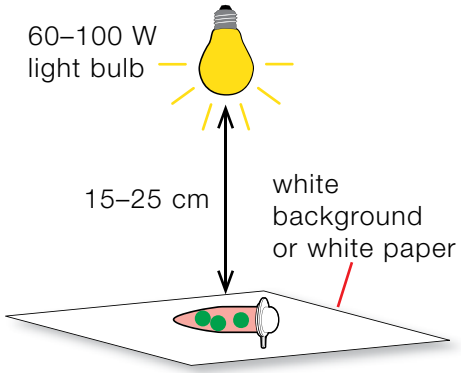
For best results, place a piece of paper or other white surface under the samples during experiments. A white surface will reflect light back to the algae beads and speed up photosynthesis.

Algae bead samples and controls should be placed directly below the lamp on the white surface. Be sure that all samples are equidistant from the light bulb. Those that are farther from the bulb will receive less light and will photosynthesize more slowly.

Lamp selection

Several types of light bulbs may be used for the overnight activation and in-class activity. 60–100 W soft white incandescent bulbs work well, but tend to produce significant amounts of heat. 13–15 W compact fluorescent (CFL) bulbs and 6–8 W LED lamps work well and produce less heat than incandescent bulbs. However, it is always a good idea to test the heat output prior to overnight activation, since larger fluorescent bulbs can produce significant amounts of heat as well.

- **DO NOT USE A WARMING LAMP**
  (This type of lamp has a ceramic base)
- Adjustable or gooseneck lamps work well since their height can be easily adjusted



Recommended light bulb types and usage distances

Bulb type	Distance from samples
60 W Incandescent	15–25 cm
13 W CFL	10–20 cm
8.5 W LED bulb (60 W equivalent)	0–20 cm
5 W LED Desk Lamp	0–5 cm

Is your lamp too hot?

Algae cannot tolerate excessive heat but require ample light intensity to function. It is a good idea to first test the lamp and light bulb setup prior to overnight activation. Otherwise, you may kill your beads during overnight activation. Set up your activation surface and turn on the lamp for 5–10 min. Place your hand on the activation surface and decide whether it feels warm or cool. If it feels warm, then move the lamp farther away from the surface, and test again after 5–10 min. If the activation surface feels cool, then the conditions are appropriate for overnight activation.

Is the bulb bright enough?

Light intensity will directly affect the rate of photosynthesis and CO_2 indicator color change. You may use a PAR (photosynthetically active radiation) meter, such as the Vernier PAR Sensor, to measure light output from your lamp. We recommend 50–1,200 µmol (photons) m^{-2} s^{-1} at the sample surface for the photosynthesis activity.

Tips & Tricks

Using cell phone lights

Cell phone lights can be used for the in-class activity, but they tend to have low light intensities resulting in slower color change. The closer algae bead samples are to the light source, the faster indicator color change will be observed.

Troubleshooting student experiments

Are my algae beads functional?

If your algae beads can perform photosynthesis, then they're healthy and usable. Even beads that appear light green are capable of performing photosynthesis. To check, perform an overnight activation. If, after activation, the indicator solution color changes to purple and the algae beads remain green, then your beads are still functional. Otherwise, your beads are no longer functional. See the Advance Preparation Instructions for instructions on algae bead activation.

My students are not seeing indicator color change

Be sure to allot 30 min for student experiments to ensure visible color change of the 1x CO_2 indicator solution. If students still are not seeing color change quickly enough, bring the light source closer to the algae beads to speed up photosynthesis, but be sure not to overheat them. If still no color change is visible, perform an overnight activation to check the functionality of your algae beads. See Lamp selection.

Debeading isn't working

The algae bead storage solution will interfere with the debeading solution. Be sure to thoroughly wash the beads with distilled water during acclimation. Additionally, the algae bead may not completely dissolve in the time allotted for debeading, but enough algae cells will be released to observe under a microscope.

Algae cells aren't visible under the microscope after debeading

Be sure to vigorously swirl the debeading solution and dissolved algae bead by flicking the tip of the tube before pipeting a drop onto the microscope slide. Swirling will resuspend algae cells into the solution.

Example Preparation Schedule

Use this example schedule as a reference for planning the lessons and advance prep in a single week. When adjusting for a block schedule be sure to carefully follow the timing for acclimating, activating, and resting algae beads. Refer to the Advance Preparation Instructions for preparation details.

Monday	Tuesday	Wednesday	Thursday
Before the lesson • Prepare the 1x CO_2 indicator solution • Wash and acclimate the algae beads (minimum 6 hr) • Set up workstations	**Before the lesson** • Prepare example algae images or debeaded microscope slides (optional) • Set up common workstation	**Before the lesson** • Set up workstations	**Before the lesson** • No prep
Lesson #1: Modeling the Inputs and Outputs of Photosynthesis and Cellular Respiration	**Lesson #2**: Experimental Design with Algae Beads	**Lesson #3**: Photosynthesis and Cellular Respiration Investigation	**Lesson #3 (cont.)**: Analyzing Results and Post Lab Questions
After the lesson • Activate the algae beads overnight (18–24 hr) • Begin equilibrating the CO_2 indicator solution **Notes** • The timing of acclimation, activation, and resting of the algae beads is essential for proper function of the beads. Activating and resting for too little or too much time will affect performance • Nonequilibrated 1x CO_2 indicator solution is fine for Lesson #1. However, it must be equilibrated for Lesson #3	**After the lesson** • Dispense into PCR tubes and rest the algae beads overnight. (6–18 hr)	**After the lesson** • Pool algae beads for later reuse (optional)	

Preparing for multiple sections

If you are preparing materials for multiple class periods on different days, we recommend activating and resting the algae beads in batches, one batch for each class period.

Activate the needed quantity of algae beads (6–7 algae beads per student workstation) for your first day of classes. When the first batch of algae beads are resting, begin the activation for the next batch. Continue this process as needed.

Advance Preparation Instructions

The instructions below are listed in chronological order according to the example preparation schedule. If you have a significant gap between Lessons #1 and #3, adjust your schedule accordingly.

Required Materials (not included in this kit)	**Quantity**
Distilled water	1 L
25 ml graduated cylinder	1
250 ml graduated cylinder	1
250–500 ml beaker	1
150 or 250 ml beaker	2
Aluminum foil	
Scissors	
Plastic wrap or Parafilm	
Lamp fitted with 60–100 W bulb	1 per group

IMPORTANT!
Review the entire advance preparation instructions before you begin Lesson #1. Preparation timing is crucial for the function of the algae beads.

IMPORTANT!
Always use distilled water for this kit. The chlorine in tap water will kill the algae.

Tips & Tricks

For detailed information about lamp selection, see p. 11.

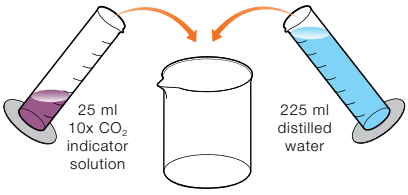
Using cell phone lights

Cell phone lights can be used for the in-class activity, but they tend to have low light intensities resulting in slower color change. The closer algae bead samples are to the light source, the faster indicator color change will be observed.

Anytime prior to Lesson #1

Dilute the concentrated 10x CO_2 indicator (for Lessons #1 and #3)

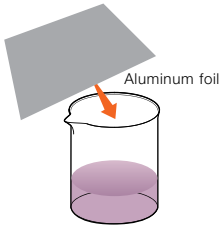
1. Add 25 ml of 10x CO_2 indicator and 225 ml distilled water to a 225 ml or larger beaker.



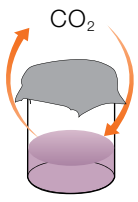
2. Gently swirl the beaker to mix the solution. The indicator is now ready for use in Lesson #1.

Continue equilibrating the 1x CO_2 indicator for later use (for Lesson #3)

3. Loosely cover the beaker of indicator solution with a tented sheet of aluminum foil.



4. Leave the beaker at room temperature overnight (minimum 18 hr) or up to 7 days to equilibrate with atmospheric CO_2. Do not fully seal the beaker as air exchange is required for equilibration. Equilibrated CO_2 indicator solution is required for Lesson #3.



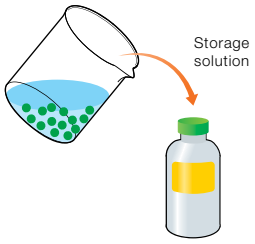
Optional stopping point

At least 3 days prior to Lesson #3

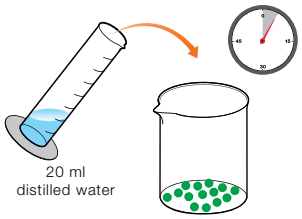
Acclimate the algae beads (for Lesson #3)

5. Transfer the algae beads from the bottle to a clean 150 ml beaker.

6. Pour any storage solution in the beaker back into the original storage bottle. Use a transfer pipet to remove any remaining storage solution from the algae beads. The storage solution can be reused to store unused or reused algae beads.



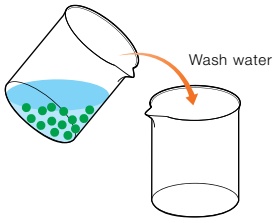
7. Wash the algae beads by adding 20 ml of distilled water to the beaker and incubating at room temperature for 5 min.



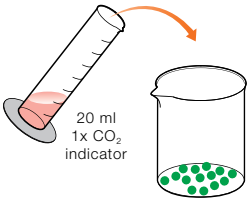
IMPORTANT!
Always use distilled water for this kit. The chlorine in tap water will kill the algae.

IMPORTANT!
Do not skip this step. Residual storage solution inside the beads will inhibit the debeading and CO_2 indicator solutions.

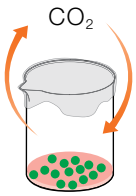
8. Pour the wash water into a second beaker in case any algae beads pour out. Use a transfer pipet to remove any remaining wash water from the algae beads. Discard the wash water.



9. Add 20 ml of 1x CO_2 indicator solution to the beads. The CO_2 indicator solution does not need to be air equilibrated for this step.



10. Loosely cover the beaker with plastic wrap or Parafilm and allow the algae beads to acclimate at room temperature in ambient light or darkness for 6–24 hr.

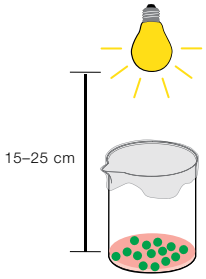


Optional stopping point

2 days prior to Lesson #3

Activate beads (for Lesson #3)

16. Place the beaker of beads 15–25 cm away from a bright light source overnight (18–24 hr). Ensure that the beads are distributed in a single layer, each with equal access to light. See lamp selection on p. 11 for more details.



17. The next day, turn off the lamp when you are ready to dispense the beads.

Tips & Tricks

If you are preparing to activate algae beads for several sections over multiple days, we recommend activating the beads in batches.

Activate the needed quantity of beads for your first day of classes. When the first batch of beads are resting after activation, begin the activation for the next batch of beads. Continue this process until you have activated enough beads for all of your sections. See Keys to Success with Algae beads for more details.

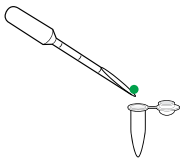
1–7 days before Lesson #2

Prepare a microscope slide for instructor demonstration (optional, for Lesson #2)

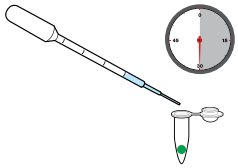
Debeading an algae bead and viewing the algae under a microscope is an optional activity that is not factored into the lesson timing. Follow the instructions below if you choose to do this activity as an instructor-led demonstration. If you would prefer to have your students debead and create microscope slides themselves, follow the directions in Appendix B instead. Visit **bio-rad.com/debeading** for an instructional video for debeading algae beads and preparing a microscope slide.

1,000x magnification image of debeaded algae cells under the microscope. Additional images can be found in Appendix B.

11. Scoop one **<u>washed</u>** algae bead into a PCR tube with a transfer pipet scoop. Fill the PCR tube with debeading solution using a transfer pipet.

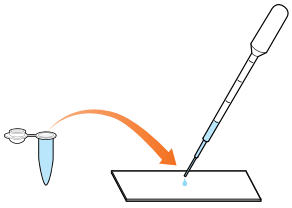


12. Soak at room temperature for 30 min. Mix vigorously every 5 min by flicking the tube.

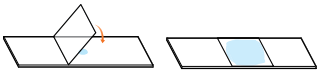


13. Flick the tube to resuspend the algae cells. The bead may not be fully dissolved, but enough algae will be released into solution to be visible under the microscope.

14. Transfer 1 drop of the solution onto a microscope slide.



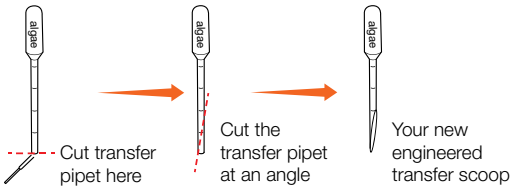
15. Place a coverslip over the drop on the slide. The slide is now ready for use.



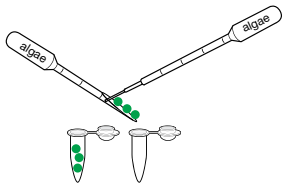
1 day prior to Lesson #3

Dispense and rest beads (for Lesson #3)

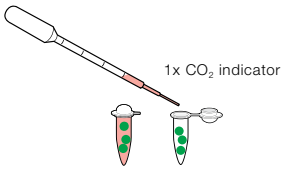
18. Cut the tip of a transfer pipet diagonally to make a scoop.



19. Use the scoop to transfer 3 algae beads into each of 2 PCR tubes **per student workstation**.



20. Use a clean transfer pipet to fill each PCR tube to the top with equilibrated 1x CO_2 indicator solution and cap the tubes.



21. Rest the algae beads by placing the tubes in ambient lighting for 6–18 hr (overnight darkness is fine).

Tips & Tricks

This rest period allows photosynthesis activity to slow down enough for cellular respiration to be observed when students perform their experiments (Lesson #3). Longer rest times allow cellular respiration to be the dominant process and shorter rest times allow photosynthesis to be the dominant process. See Keys to Success with Algae Beads for more details.

Lesson 1

Modeling the Inputs and Outputs of Photosynthesis and Cellular Respiration

Lesson overview

PART 1: Observing indicator color change (25 min)

- Students blow into the carbon dioxide indicator solution and observe the color change.
- Students create and revise a model of their observations.

PART 2: Linking photosynthesis and cellular respiration (25 min)

- Students take an inventory of the inputs and outputs of photosynthesis and cellular respiration and identify connections between them
- Students predict what might happen if a photosynthesizing organism were put into the indicator solution

Learning outcomes

- Students will practice describing phenomena and developing models to illustrate their understanding
- Students will find connections between the inputs and outputs of photosynthesis and cellular respiration

Prior knowledge needed

- Photosynthesis and cellular respiration are processes by which plants and animals generate and use energy
- Carbon dioxide is produced during cellular respiration, and used up during photosynthesis

Classroom preparation

- Split the class into small groups of up to 4 students
- Prepare student workstations

Student Workstations	Quantity
Drinking straw or transfer pipet with bulb removed	1
1x CO_2 indicator solution in a clear plastic cup	5 ml

Science & Engineering Practices

Modeling

Students create and revise models to illustrate and explain color changes of the CO_2 indicator solution. See p. 8 for an explanation of modeling as a science and engineering practice.

Disciplinary Core Ideas

Matter and Energy Flow in Organisms

Students will learn how the products of cellular respiration are consumed by photosynthesis and vice versa; energy flows and transforms from light to chemical energy.

Crosscutting Concepts

Scale, Proportion, and Quantity

Photosynthesis and cellular respiration can be discussed at the molecular, cellular, and macro (CO_2 indicator color change) levels. Encourage students to make connections between levels of scale as they create and discuss their models.

Energy and Matter: Flows, Cycles, and Conservation

Discuss with students how the products of cellular respiration are consumed by photosynthesis and vice versa in a cycle of matter and flow of energy.

Cause and Effect: Mechanism and Prediction

Students will discuss the potential causes of the CO_2 indicator solution color change. They apply their understanding to predict the effect of introducing plants to the indicator solution.

PART 1 (25 min): Observing indicator color change

Goal: Students understand how the carbon dioxide indicator solution works and develop a model to describe it.

Question numbers in the Student Guide that correspond to the instructor lesson step.

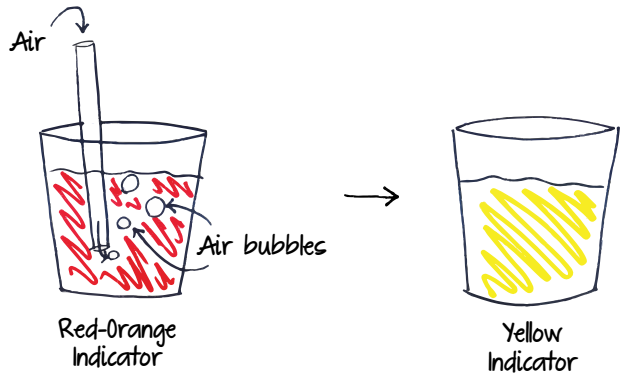
1. ***Have students observe and describe the indicator solution.*** Record students' thinking on the board as a class and/or have them do so in their notebooks. Students do not yet know the function of the carbon dioxide indicator. Until students have discovered how the indicator works, it is referred to simply as "indicator."

2. ***Have one student from each group blow gently through a straw (for about 5 sec) into the indicator solution.*** The others in the group should observe. The indicator solution will change from an orange-red to yellow.

3. **Invite students to think independently about potential causes of the color change.**

4. **Have students create an initial model of what they observed.** Using a large sheet of paper or their notebooks students should draw a model that illustrates what they observed and provide a brief explanation.

   Student models may look something like this:



5. ***Discuss with your students as a class what caused the indicator solution to change colors.*** Help students connect the color change and the addition of carbon dioxide from cellular respiration in their bodies.

Tips & Tricks

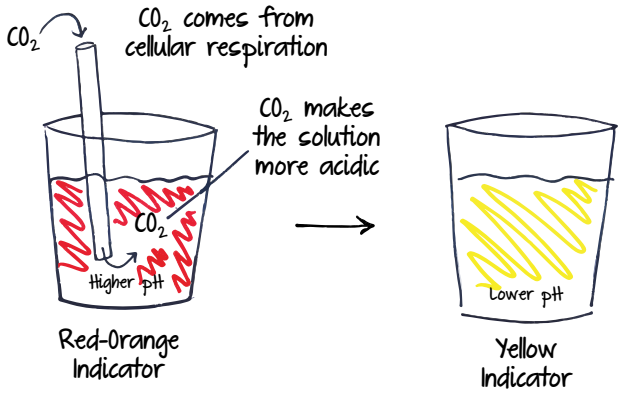
Warmup question for students

What do you know about photosynthesis and cellular respiration? Write down anything you know about these processes.

Example guiding questions:

- What did you notice about the color of the solution when you blew into it?
  It became more yellow.
- What are you breathing out when you blow into the indicator?
  Carbon dioxide, water vapor, air, odor
- How does carbon dioxide change the pH of the solution?
  It makes it more acidic. It lowers the pH.
- What color does the indicator change to when it becomes more acidic?
  Yellow
- What does the yellow color indicate?
  More carbon dioxide/more acid/lower pH
- How could we test whether carbon dioxide is causing the color change?
  By blowing plain atmospheric air into the indicator as a comparison.

Have students revise their models to include the role of carbon dioxide. Student drawings may look something like this:



Tips & Tricks

Design a quick experiment

Have students use a transfer pipet to repeatedly bubble atmospheric air into the CO_2 indicator solution. Compare the results of doing so with the results of blowing through a straw into the CO_2 indicator solution.

PART 2 (25 min): Linking photosynthesis and cellular respiration

Goal: Students create an inventory of the inputs and outputs of photosynthesis and cellular respiration using the indicator as a concrete reference point.

Question numbers in the Student Guide that correspond to the instructor lesson step.

6. ***Have students create an inventory of the inputs and outputs of both photosynthesis and cellular respiration.*** Use the following table as a guide. Be sure to point out that light is required for photosynthesis but not for cellular respiration.

	Photosynthesis	Cellular respiration
Inputs	Light (energy) Carbon dioxide Water	Glucose (chemical energy) Oxygen
Outputs	Oxygen Glucose (chemical energy) ATP	ATP Carbon dioxide Water
Organelle	Chloroplasts	Mitochondria
Example organism	Plants	Animals

Tips & Tricks

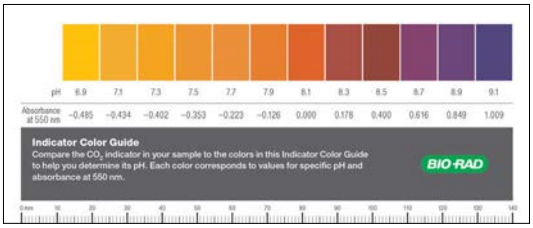
Cellular respiration in plants

At this stage, students may not yet know that plants also do cellular respiration. Allow them to ask the question and discover that fact through their experimentation later in the lesson sequence.

7. ***Discuss with your students the connection between photosynthesis and cellular respiration.***
Example discussion questions:

- How are photosynthesis and cellular respiration connected?
- Why do plants perform photosynthesis?
- Why do plants make glucose?
- Do plants perform cellular respiration?

8. ***Show students the indicator color guide and explain the relationship between the CO_2 indicator solution color, carbon dioxide level, and pH of the solution.***
Connect students' observations of the indicator color change with the range on the color guide. What does a purple color indicate?



9. ***Ask students "How would the color change if you put a plant in the indicator solution?"*** Prompt students to think about the inputs and outputs of photosynthesis (including light). Remind them of the dependence of the indicator color on carbon dioxide levels.

Lesson 2

Experimental Design with Algae Beads

Lesson Overview

PART 1: Using algae beads as a model organism (15 min)

- Students examine algae beads
- Students view algae under the microscope (15 min)
  - (Optional) Instructor demo debeading (+15 min)
  - (Optional) Student-led debeading activity (+ 30 min)
- Students create a model to describe their predictions of how algae might interact with the indicator solution

PART 2: Planning investigations with algae beads (35 min)

- Students design their own experiment using provided materials

Learning outcomes

- Students will practice developing models to explain energy transfer processes in plants and algae
- Students will plan an experiment that will provide evidence to answer a question
- Students will practice formulating the components of a scientific investigation, including a scientific question, hypothesis, variables, and controls

Prior knowledge needed

- The carbon dioxide indicator changes colors in response to the amount of carbon dioxide in the solution
- The inputs of photosynthesis are light, carbon dioxide, and water; the outputs of photosynthesis are oxygen, glucose, and ATP
- The inputs of cellular respiration are oxygen and glucose; the outputs of cellular respiration are carbon dioxide, water, and ATP

Classroom preparation

- Split the class into small groups of up to 4 students
- Prepare an image of debeaded algae under the microscope
  See Advance Preparation Instructions for details and alternatives
- Prepare a common workstation including the following materials for students to reference during their experimental design. If you are having students prepare their own microscope slides, refer to Appendix B for additional materials used in part 1

Common Workstation Materials

Carbon dioxide indicator in a cup or beaker
Algae beads in a beaker
Transfer pipets
Extra PCR tubes
Sheets of aluminum foil, 5 x 5"
Student Indicator Color Guide
Lamp fitted with 60–100 W bulb

Science & Engineering Practices

Modeling

Students will create and revise models to represent color changes of the CO_2 indicator solution. See p. 8 for an explanation of modeling as a science and engineering practice.

Asking Questions

Students will practice writing defined investigation questions about energy processes in plants and algae.

Planning Investigations

Students will design experiments to collect evidence to answer their investigation questions.

Disciplinary Core Ideas

Matter and Energy Flow in Organisms

Students will design experiments to learn more about the processes of photosynthesis and cellular respiration.

Crosscutting Concepts

Energy and Matter: Flows, Cycles, and Conservation

Students will consider how the products of cellular respiration are consumed by photosynthesis and vice versa; energy flows and transforms from light to chemical energy.

Cause and Effect: Mechanism and Prediction

Students will apply their understanding of the cause and effect of CO_2 indicator solution color change as they design an effective investigation into photosynthesis and cellular respiration.

PART 1 (20 min): Using algae beads as a model organism

Goal: Introduce algae beads as a model organism.

Question numbers in the Student Guide that correspond to the instructor lesson step.

10. ***Present algae beads to the students as a model organism for plants.***

Have students view the algae beads and make observations. Although algae are not plants, they are a simple photosynthesizing model organism.

11. ***Show students images of algae cells under the microscope and have them make observations.***

Algae beads themselves are not individual algae cells but instead contain thousands of algae cells. Images can be found in Appendix B.

12. ***Ask students: "What would happen if you put algae beads in the carbon dioxide indicator?"***

Remind students of their answers to question 9. Predictions should include how the color of the indicator solution will change. Then ask students to share their thinking about why they made that prediction.

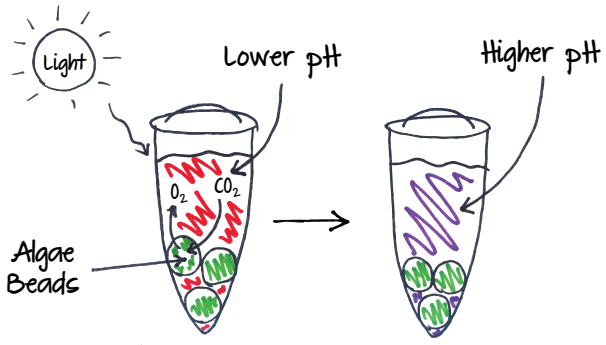
(Optional) Show students the time lapse video of algae beads in carbon dioxide indicator and exposed to light.

Visit bio-rad.com/algaetimelapse to view the video.

13. ***Have students create a new model of algae beads in the indicator.***

Models should include the relationships between the inputs and outputs of photosynthesis (and possibly cellular respiration) within light and dark conditions.

Student models may look something like this:



Tips & Tricks

Warmup question for students

Describe two reasons scientists use model organisms in the laboratory.

Tips & Tricks

Debead an algae bead (optional)

Instead of showing images of algae cell to your students, you may debead an algae bead and have students view the algae under a microscope. This optional activity may be done as an instructor-led demonstration (see Advance Preparation Instructions) or as a student-led activity (See Appendix B). Allow for additional classroom time if you include the student-led activity.

Tips & Tricks

Provide indicator color guides

Students can use the indicator color guide to help them understand the cause of the color change as well as the full color range of the indicator solution. The indicator color guides also provides a visual link between color and pH.

PART 2 (30–40 min): Experimental design

Goal: Students design an experiment with available materials to answer their investigation question.

Structured inquiry

If your students are not yet ready to design their own experiments, you can provide them with a structured investigation question and protocol found in Appendix A. If you use the structured protocol, skip part 2 and proceed directly to Lesson #3.

Question numbers in the Student Guide that correspond to the instructor lesson step.

14. Prompt students to ask questions about algae beads, photosynthesis, cellular respiration, and how algae beads can be used to understand these processes in nature.

Example prompt questions:

- What do you already know about photosynthesis and cellular respiration?
- What questions do you have about photosynthesis and cellular respiration and how would you use algae beads and indicator to test your ideas?
- How did your knowledge about photosynthesis and cellular respiration help you describe the color change you observed in the indicator solution?
- What measurement or observation could you make to demonstrate algae beads doing photosynthesis or cellular respiration?

Assemble a whole-class list of student experimental questions for everyone to see.

15. Explain the tools that are available to your students when they design their experiments.

Common Workstation Materials
Carbon dioxide indicator solution in a cup or beaker
Algae beads in a beaker
Transfer pipets
Extra PCR tubes
Sheets of aluminum foil, 5 x 5"
Student Indicator Color Guide
Lamp fitted with 60–100 W bulb

16. ***Ask students: "Which question from our list could you answer using the materials we have?"***

The materials listed above limit which experimental questions can be answered in order to help focus your students. Add or remove available items as appropriate. See Curriculum Fit and Inquiry Support for example experiments students could do.

Have each student group select one investigation question they want to pursue.

Review students questions and provide guidance.

Have student groups design their experiments.

Students may use the Experimental Design and Planning section of the Student Guide during this process or they may write in their notebooks.

Be sure students include the following elements:

- Investigation question
- Materials
- Independent variable
- Dependent variables
- Constants
- Controls
- Protocol
- Predicted results

Review student experimental design and provide guidance as needed.

As a class, through peer review, or by consulting with one group at a time provide students with feedback on their experimental designs. Discuss relevant variables and appropriate controls. Commonly missed controls include:

- Controlling for lamp heat: place all tubes under the same light, including foil-wrapped "dark" tubes, to ensure that all samples are exposed to the same heat from the lamp
- Controlling for the effects of light: place additional tubes with indicator solution but without algae beads under the light to demonstrate that the indicator solution does not change color because of light exposure alone

Lesson 3

Photosynthesis and Cellular Respiration Investigation

Lesson Overview

First day

PART 1: Conducting the experiments (50 min)

- Instructor reviews available materials, pre-lab and post-lab experimental steps, and student protocols
- Students complete their investigations

Second day

PART 2: Analyzing experimental results and revising models (30 min)

- Students engage in a whole class discussion to review their methods and findings from the previous class period
- Students make claims based on their collected evidence for which model is supported
- Students revise their models of energy transfer in algae beads

PART 3: Post lab questions and discussions (20 min)

- Students consider the scenario of adding a snail and algae beads to the carbon dioxide indicator

Learning outcomes

- Students carry out experimental protocols and gather evidence
- Students provide a scientific explanation for their experimental observations
- Students can predict the impact of heterotrophs on carbon dioxide levels within a system

Prior knowledge needed

- The carbon dioxide indicator changes color based on the amount of carbon dioxide in the solution
- The inputs of photosynthesis include light, carbon dioxide, and water; the outputs of photosynthesis include oxygen and glucose
- The inputs of cellular respiration include oxygen and glucose; the outputs of cellular respiration include carbon dioxide and water

Classroom preparation

- Split the class into small groups of up to 4 students
- Prepare student workstations

Science & Engineering Practices

Modeling

Students create and revise models to represent color changes of the CO_2 indicator solution. See p. 8 for an explanation of modeling as a science and engineering practice.

Carrying Out Investigations

Students collect and translate data into meaning and ultimately a scientific explanation.

Disciplinary Core Ideas

Matter and Energy Flow in Organisms and Ecosystems

Students will conduct experiments to learn more about the processes of photosynthesis and cellular respiration and how both energy and matter are exchanged within and between organisms.

Crosscutting Concepts

Energy and Matter: Flows, Cycles, and Conservation

Students will consider how the products of cellular respiration are consumed by photosynthesis and vice versa; energy flows and transforms from light to chemical energy.

Cause and Effect: Mechanism and Prediction

Students predict the outcomes of their experiments based on their understanding of the causes of color change of the CO_2 indicator solution.

Student Workstations items	Quantity per workstation
Small cup of 1x CO_2 indicator solution, 1 ml	1
PCR tube with 3 activated algae beads and filled with 1x CO_2 indicator solution	2
Transfer pipets	5
Empty PCR tubes	2
Waste container	1
Sheet of aluminum foil, 5" x 5'"	1
Student indicator color guide	1
Marking pen	1
Lamp, 60–100 W equivalent	1
Clock or timer	1

First Day

PART 1 (50 min): Conducting the experiments

Goal: Students complete the experiments they designed.

Review the available materials with students.

Have students conduct their experiments.

As students complete their experiments and record data, they should record any changes to the methods or materials they used in the Experimental Design and Planning section of the student guide or in their own notebooks.

Tips & Tricks

For detailed information about lamp selection, see p. 11.

Using cell phone lights

Cell phone lights can be used during the in-class activity in place of a lamp and clock. However, they tend to have low light intensities resulting in slower color change. The closer algae bead samples are to the light source, the faster indicator color change will be observed.

Tips & Tricks

Warmup question for students

What is your investigation question? What evidence do you expect to collect in your experiment?

Tips & Tricks

Troubleshoot your students' experiments

For more information about troublshooting student experiments, see p. 13.

Second Day

PART 2 (20–30 min): Analyzing experimental results and revising models

Goal: Students develop a scientific explanation for their experimental observations and revise their models of energy transfer in algae based on those explanations.

As a whole class, have students explain their investigation questions, methods, and observations.

Invite students to highlight any surprises that they noticed and any conflicts with what they had predicted would happen.

Have student groups write scientific explanations for their investigation questions based on their evidence.

Use claim, evidence, and reasoning to help students answer their investigation questions and then write scientific explanations for their results.

Have students revise their previous models of energy transfer in algae based on their new claims.

PART 3 (20–30 min): Post-lab questions and discussion

Goal: Assess student learning and have students expand their models of energy transfer to include heterotrophs.

In class or as homework, have students complete the post-lab questions.

The post-lab questions are an opportunity for students to apply the knowledge they gained in the experiment to a broader context that includes heterotrophs in a mini ecosystem.

Once students have completed the post-lab questions, ask students to explain their answers.

Students may have questions about whether enough oxygen will be produced by the algae beads to keep the snail alive. This brings up the concepts of rates of reaction and is an opportunity to discuss how one could measure those rates.

Tips & Tricks

Use peer review

Have student groups review the claims of other groups and provide feedback or critiques.

Appendix A

Lesson #3 Quick Guide:
Photosynthesis and Cellular Respiration Investigation — Structured Inquiry

Investigation question

Plants and algae produce glucose through photosynthesis. Glucose is a form of stored chemical energy. Animals consume glucose from plants using cellular respiration.

Do algae also consume the glucose they produce by performing cellular respiration?

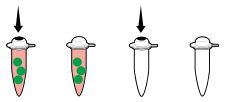
Focus questions

1. ***How will the pH of the CO_2 indicator solution change as algae beads perform photosynthesis? How will the CO_2 indicator solution color change?***

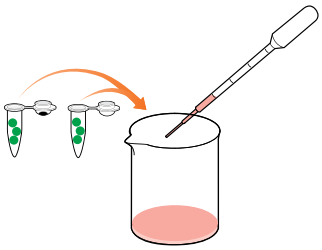
2. ***How will the pH of the CO_2 indicator solution change if the algae beads perform more cellular respiration than photosynthesis? How will the CO_2 indicator solution color change?***

Protocol

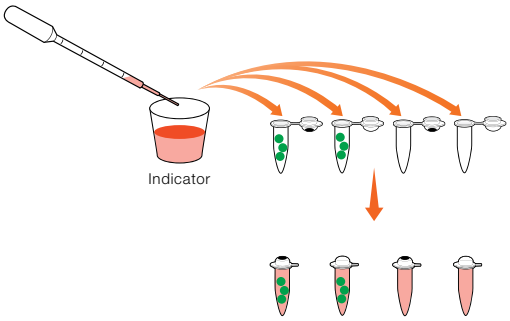
1. ***Mark the cap of one PCR tube containing algae beads with a dot. Also mark the cap of an empty PCR tube with a dot. The tubes with dots are your "dark" tubes and will be covered in foil during the experiment.***



2. ***Use a transfer pipet to remove any solution from each of the PCR tubes with beads. Be careful not to remove or crush the beads.***

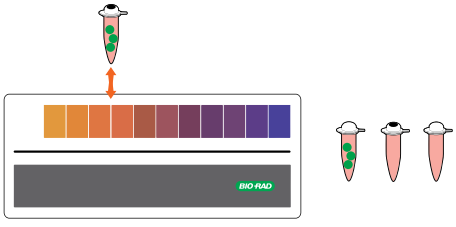


3. ***Using a clean transfer pipet, add CO_2 indicator solution to each of the four PCR tubes. Add enough solution to reach the top of each tube, and then close the lids.***

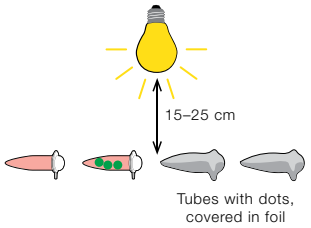


4. ***With the caps closed, turn the tubes with beads upside down and flick the tips to dislodge any algae beads that are stuck at the bottom. The algae beads should move freely in the CO_2 indicator solution.***

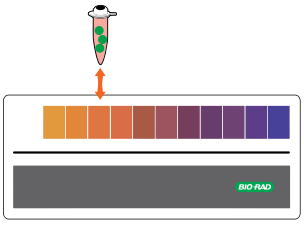
5. ***Compare the starting color of the CO_2 indicator in your tubes to the sections on the indicator color guide. Record the pH and starting time in the table on the next page or in your notebook.***



6. ***Wrap the tubes with dots on their caps in foil. Place all four tubes under the light source. Be sure that all tubes are approximately the same distance from the light source.***



7. ***Every 5 min, for 30 minutes, mix the CO_2 indicator solution in the tubes by flicking the tubes. Compare the color of the CO_2 indicator solution in the tubes to the indicator color guide. Record the pH of the color on the guide that matches the indicator color. Repeat for each time point and record your results in the table below.***



	pH readings			
Time	**+beads, light**	**+beads, dark**	**–beads, light**	**–beads, dark**
Start time:				

Analysis and conclusions

Rewrite your investigation question

Write a <u>claim</u> that answers the experimental question

Provide <u>evidence</u> to support your claim

Explain your <u>reasoning</u> for how the evidence supports your claim

Provide a <u>scientific explanation</u> of the results of your experiment

Create a revised model of algae beads and indicator solution that includes the new information you learned from the results of your experiment.

Explanation — Write 2–3 complete sentences to describe your model.

Appendix B

Student-Led Debeading of Algae Beads

Goal: Students view algae cells from debeaded algae beads.

Notes

- Allot an additional 40 min of class time for this activity.
- If an extra class period is required to accommodate this activity, be sure to adjust your algae bead preparation schedule to ensure that activation begins two days prior to Lesson #3.
- It is essential that the algae beads have been washed with distilled water before attempting to debead. The storage solution will inhibit the debeading solution. See Advance Preparation Instructions for details.

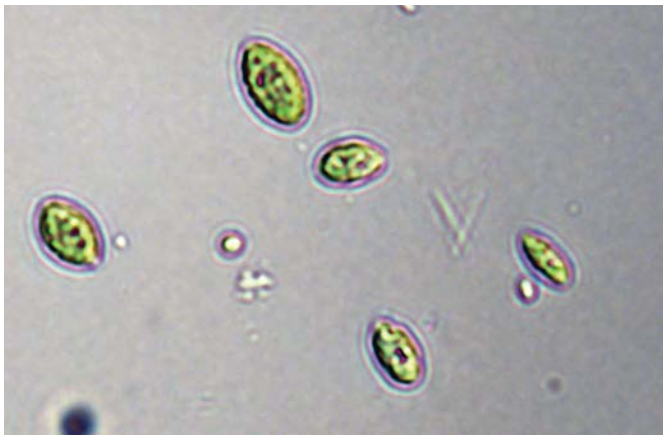
Classroom preparation

- Before the activity, scoop one washed algae bead into an empty PCR tube for each student group.
- Provide copies of the student protocol on the next page to students.
- Prepare student workstations

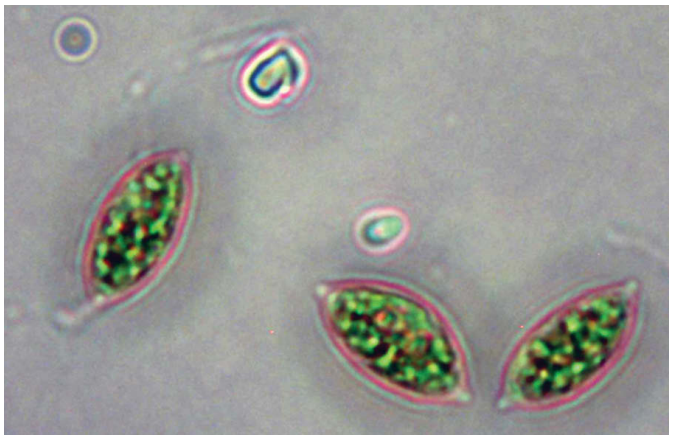
Materials

Student Workstations	Quantity
PCR tube with a single washed algae bead	1
Debeading solution in a small cup	0.5 ml
Disposable Plastic Transfer Pipets	2
Microscope slide	1
Coverslip	1
Microscope	1

Example images of debeaded algae under the microscope



400x magnification

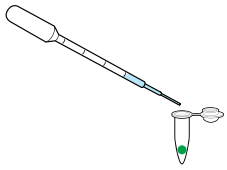


1,000x magnification

Debeading Algae Beads

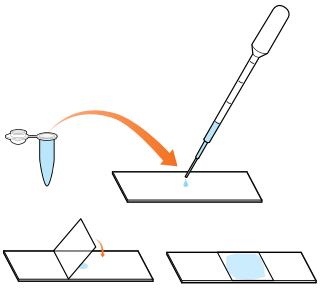
Protocol

1. *Use a transfer pipet to add debeading solution to the PCR tube that contains an algae bead.*



2. *Incubate the solution at room temperature for 30 min. Flick the tube vigorously every 5 min. After 30 min, enough algae cells will have been released to proceed with the microscopy activity. The bead may still look intact.*

3. *Flick the tube to mix. Then use a new transfer pipet to transfer 1 drop of dissolved algae bead solution to the center of a microscope slide. Place a coverslip over the microscope slide.*



4. *Observe the algae under a microscope.*

5. *Draw a sketch of the algae under the microscope and write a brief description. Include details about color and appearance.*

Appendix C

Frequently Asked Questions

How long will algae beads last?

Before activation, the algae beads will survive for at least 3 months at 4°C in the storage bottle. After activation, the beads will survive for at least 2 weeks in a CO_2 indicator solution at room temperature under ambient lighting. See Keys to Success with Algae Beads for further details.

What kind of algae are in the beads?

The strain of algae used in this kit, *Scenedesmus obliquus*, is a green freshwater unicellular alga that is commonly found in clean ponds, lakes, and rivers worldwide and is not pathogenic.

Are there any safety procedures to follow when using or disposing of the algae beads?

Handling the algae beads in this kit requires use of standard microbiological practices, including but not limited to the following:

- Decontaminate work surfaces once a day and after any spill of living material
- Decontaminate all liquid or solid wastes before disposal
- All persons must wash their hands after they handle material containing algae and before exiting the laboratory
- Perform all procedures carefully to minimize creation of aerosols
- Use only mechanical pipetting devices; mouth pipetting is prohibited
- Eating, drinking, smoking, and applying cosmetics are not permitted in the work area
- Wearing protective eyewear and gloves is strongly recommended

If an autoclave is not available, place algae beads and all solutions and components (transfer pipets, cuvettes, slides, coverslips, etc.) that have come in contact with the algae beads in a fresh 10% bleach solution for at least 20 minutes for decontamination. Please consult your local Environmental Health and Safety organization for disposal requirements in your area.

Can I use tap water to wash the algae beads?

No. Always use distilled water for this kit. Tap water contains chlorine and will kill the algae.

How should the algae beads be stored?

Algae beads in the storage bottle should always be submersed in storage solution to prevent the beads from drying out.

Why do I need to activate the algae beads?

The algae in the kit are provided in a dormant state. For the experiment to yield visible and measurable results within 30 minutes, their photosynthetic machinery needs be active. To ensure consistent activation, the beads should be distributed in a single layer, with no obstruction of the light path to each bead. If the algae beads are not activated prior to the experiment, it will take longer to obtain results. See Keys to Success with Algae Beads for further details.

What kind of light should I use to activate the algae beads?

A 60–100 W soft white incandescent light bulb works well. Alternatively, a 13–15 W compact fluorescent (CFL) or a 6–8 W LED will also work well. Make sure that the light does not heat up the beads and that the beads remain submersed in CO_2 indicator solution (do not allow the CO_2 indicator solution to evaporate). This can cause them to overheat (temperatures exceeding 30°C) or photobleach. See Keys to Success with Algae Beads for further details.

Why do the algae beads need to rest after activation?

This rest period allows active photosynthesis to slow down enough for cellular respiration to be observed when students perform the their experiments. Longer rest times allow cellular respiration to be the dominant process and shorter rest times allow photosynthesis to be the dominant process. See Keys to Success with Algae Beads for further details.

What are the beads made of?

The beads are made of sodium alginate, a polymer that is not harmful to the algae. The algae can photosynthesize and respire within the alginate matrix.

How does the CO_2 indicator solution work?

The CO_2 indicator solution is a colorimetric pH-sensitive indicator that responds to fluctuations of the carbonic acid formation that spontaneously occurs when CO_2 dissolves in water. After preparing the CO_2 indicator, it must have enough time to equilibrate with atmospheric levels of CO_2. Atmospheric CO_2 levels will cause the CO_2 indicator to equilibrate to a pH of approximately 7.9–8.3, which will turn it dark orange (refer to indicator color guide).

Can I use the algae beads in back-to-back class periods?

Using algae beads in multiple class periods on the day same is not recommended because the balance between the rates of photosynthesis and cellular respiration changes through use and will be inconsistent between classes. We recommend dispensing enough algae beads into PCR tubes ahead of time to supply all your class periods in one day. See Keys to Success with Algae Beads for further details.

Where can I find answers to the questions in the student manual?

The Instructor's Answer Guide is provided separately as a printed insert in the kit box. It is provided in this format to minimize the opportunity for students to see the answers. Please do not scan or distribute the Answer Guide to further prevent students from finding the answers.

Can I get more Indicator Color Guides?

If you run our of Indicator Color Guides, visit bio-rad.com/colorguide to download a PDF to print more. However, depending on your printer, the colors may vary significantly from the Indicator Color Guides provided in the kit.

Can I reuse transfer pipets?

Yes. Be sure to rinse transfer pipets thoroughly with distilled water before reuse.

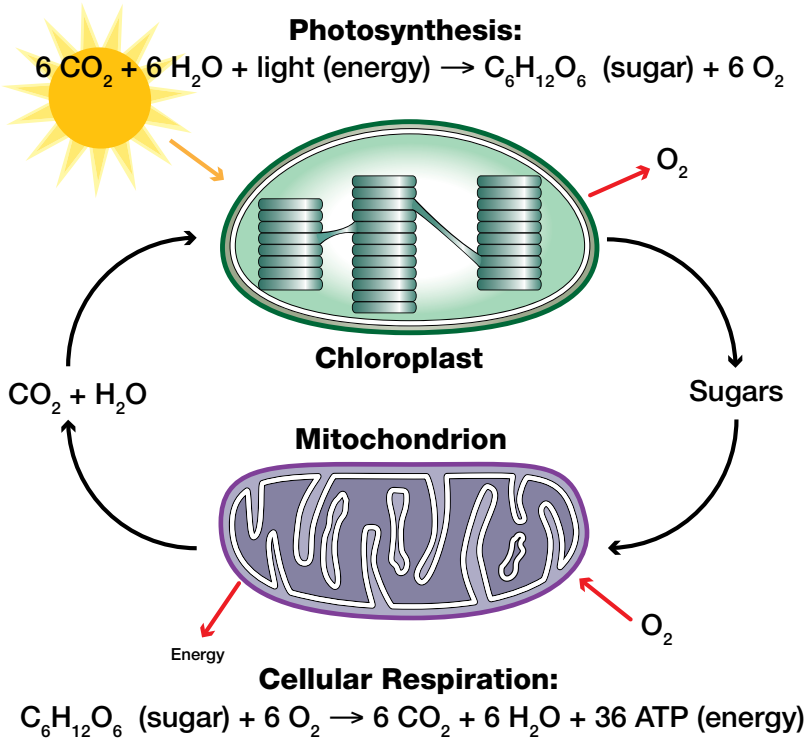
For further questions contact Technical Support at support@bio-rad.com or 1-800-4BIORAD, option 2.

Appendix D

Introduction to Photosynthesis and Cellular Respiration

Photosynthesis and cellular respiration occur within the same cell

It is important to understand that, although only autotrophs like plants and algae perform photosynthesis, ALL organisms (you, the neighbor's cat, and the tree at the end of the street) perform cellular respiration. In fact, the reactions that break down glucose in the presence of oxygen are universal. Even autotrophs, who produce their own food, use cellular respiration to break down the sugars to extract energy. Photosynthesis is the capture and transformation of light energy into chemical energy and cellular respiration is the burning of glucose (chemical energy) to grow and to do the work of living. Both plants and animals (including microorganisms) need oxygen to perform aerobic cellular respiration. Even plant roots, which cannot perform photosynthesis under ground, need oxygen for survival. This is why overly wet or saturated soils are bad for root growth and function, as well as to the decomposition processes carried out by microorganisms in the soil.



In autotrophs such as algae, these processes occur within the same cells! In fact, if you could look inside one of the algae cells used in the lab investigations, you'd see a large central chloroplast as well as smaller mitochondria — all within the same cell. Though photosynthesis and cellular respiration are connected by common inputs and outputs, algae cells balance the rates of photosynthesis and cellular respiration as needed to survive environmental changes.

Summary

The following table summarizes some of the hallmarks of photosynthesis and cellular respiration.

Table 1. Hallmarks of photosynthesis and cellular respiration.

	Photosynthesis	Cellular Respiration
Input	CO_2, H_2O, light energy	Glucose, O_2
Output	Glucose, O_2	CO_2, H_2O, chemical energy (glucose)
Organism type	Autotrophs (producers) only	Autotrophs and heterotrophs
Organelle	Chloroplast	Mitochondrion
Requires light	Yes	No

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