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Investigating Plant and Animal Physiology with Vernier Sensors

Jennifer Olson and Cara Cario

Virginia Commonwealth University, Department of Biology, 1000 West Cary Street, Richmond, VA, 23284, USA

Extended Abstract

Measurements of plant and animal metabolism in lab classrooms can often be financially prohibitive due to the high cost of research-grade respirometry equipment. As such, lab exercises that focus on photosynthesis or cellular respiration often use indirect or overly simplistic methods (e.g., floating leaf disks or air bubbles in syringes), rather than incorporating whole, living plants and animals, especially in introductory lab courses.

We oversee an upper-level college lab course that investigates comparative physiological responses across plant and animal taxa, with frequent application to changing environmental conditions. Thus, measurements of cellular respiration and photosynthesis are often incorporated into our protocols. To facilitate these measurements, we use GoDirect® wireless gas sensors developed by Vernier® Science Education to detect changing levels of O_2 and CO_2 within a sealed container, closely mimicking a traditional (yet more costly) closed-circuit respirometry system (Lighton 2019). These sensors can connect to students' laptops or mobile devices via Bluetooth or USB and are controlled using the manufacturer's free software or mobile app, Graphical Analysis, which also collects and visually displays the data. The data can also be exported as .csv files for further analysis in spreadsheets or statistical software.

When introducing metabolic measurements to students in the classroom, we usually begin with a short demonstration measuring O_2 consumption and CO_2 production in sprouting green peas. The protocol closely follows the cellular respiration exercise (Experiment #11) published in the *Biology with Vernier* lab book (Masterman 2019). We use this demonstration to address common student misconceptions that plants only photosynthesize, and that photosynthesis occurs in all parts of the plant. This activity also introduces the gas sensors and how Graphical Analysis software allows us to graph the change in O_2 and CO_2 levels in real time. Conveniently, both gas sensors also have embedded temperature probes.

Two ways that students use the O_2 and CO_2 gas sensors in the integrative physiology lab course are to explore metabolic scaling in plants and animals and to evaluate metabolic thermal relations in ectotherms. Vernier has published similar experiments (for example: Masterman 2019; Melville 2019) and our protocols expand on these in multiple ways. In one of our plant-based experiments, students measure changes in O_2 and CO_2 concentration in the dark in multiple plant species and across a range of sizes. We have quantified sizes as both leaf surface area and mass. Also, unlike the experiments in the published Vernier lab book, which use multiple spinach leaves removed from multiple plants (Experiment #31, Masterman 2019), our protocols allow students to measure photosynthesis and respiration using intact plants, thereby preserving the entire plant (Figure 1). Subsequent experiments in the semester have also explored metabolic responses to nutrient deficiencies and limited water availability.

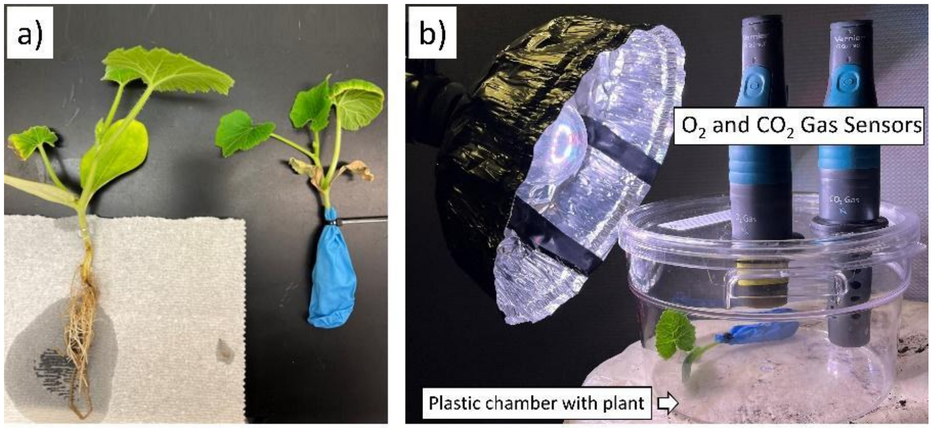

Figure 1. Measurements in pumpkin seedlings; a) setup, excluding damp roots and soil; b) chamber and sensor setup

We have also measured changes in O_2 and CO_2 levels in a variety of invertebrates, including house crickets (*Acheta domesticus*), tobacco hornworms (*Manduca sexta;* Figure 2), and fiddler crabs (*Uca sp.*). Within individual species, metabolic rate has also been measured across multiple sizes, life stages, and temperatures. Our basic protocol is based on Vernier's experiment investigating the effect of temperature on gas exchange in crickets (Experiment #23, Masterman 2019). The Vernier protocol instructs students to measure ten animals in a single chamber and calculate the average mass and respiration rate. However, we have found that the CO_2 sensor, specifically, is sensitive enough to measure gas exchange in a single cricket as small as 0.22 g. When investigating metabolic scaling in invertebrates, our students measure *Manduca* across a 10-fold range of mass, in addition to crickets that have a much smaller range of mass (approximately two-fold). Previous results have demonstrated a significant relationship between metabolic rate and mass across both species combined and when examining *Manduca* alone, but the trend across the small size range of crickets alone is not significant. Thus, choosing appropriately sized animals should be considered by instructors developing similar activities.

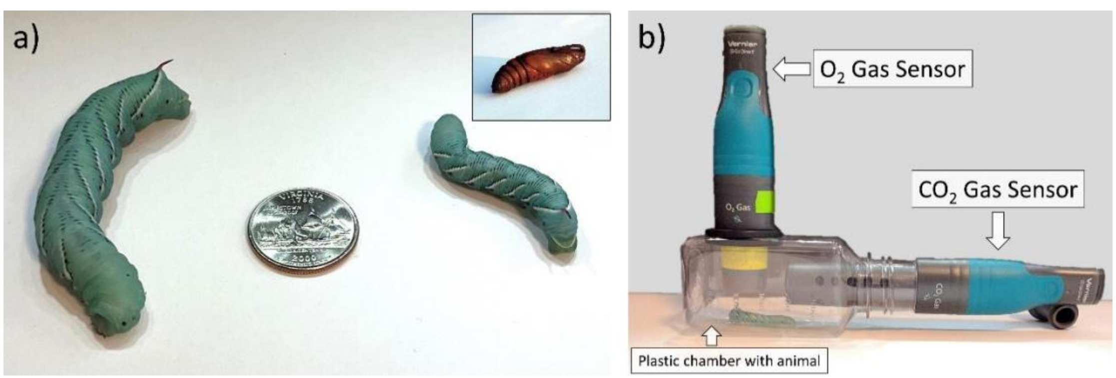

Figure 2. Metabolic measurements in tobacco hornworms; a) multiple size and life stages; b) chamber and sensor setup

Participants in the mini workshop collected respirometry data on plants and animals with Vernier GoDirect® O_2 and CO_2 gas sensors using plastic chambers that accommodate both probes simultaneously. Multiple sizes of tobacco hornworm (*Manduca sexta*) larvae and pupae were available for those who wanted to measure animals. Participants investigating plants could choose between pumpkin seedlings (*Cucurbita sp.*) grown in control (nutrient-rich) soil vs. those grown in nutrient-poor soil. Following their measurements, we presented sample results from the expanded experiments in our integrative physiology course to demonstrate methods to visually present and statistically analyze the data.

Learning curves, measurement techniques, and troubleshooting are often hurdles when adopting new classroom equipment. Thus, we also shared tips and solutions to problems we have encountered. Some examples include trouble pairing to the correct probe when there are many in the classroom, inserting the probes with enough force to create an airtight seal with the chamber, ensuring that animals move as little as possible and avoid defecating, maintaining consistent light exposure for plants, and minimizing the impact of diurnal cycles and ambient weather conditions on plant respiration or photosynthesis measurements.

Keywords: metabolism, respiration, photosynthesis, Vernier, *Manduca*, gas exchange, scaling, temperature

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Correspondence to: Jennifer Olson, jrolson@vcu.edu

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