The atmospheric CO$_2$ level has increased by 30% over the past 50 years and is predicted to increase up to 800 ppm by 2100 (Thuiller et al., 2005; Lindsey, 2020). Elevated CO$_2$ is expected to increase carbon storage and plant productivity due to the enhanced carbon fixation in ecosystems (Drake et al., 2011). In the longer term, rising CO$_2$ would increase the carbon : nitrogen ratio within plant tissue and lead to progressive nitrogen limitation in the ecosystem (Luo et al., 2004). Elevated CO$_2$ also has a number of direct effects on plant performance, including increasing water use efficiency by increasing biomass accumulation and reducing water loss (Li et al., 2003).

Growth chamber studies have played an important role in examining the effect of CO$_2$ levels on plant performance because they can simulate a range of growth conditions and provide a degree of replication and control that is difficult to achieve in the field. Manipulating CO$_2$ concentrations in a growth chamber requires a CO$_2$ sensor to monitor the concentration of the gas, as well as a system for delivering CO$_2$ to the chambers (Tissue and Oechel, 1981). In the longer term, rising CO$_2$ would increase the carbon : nitrogen ratio within plant tissue and lead to progressive nitrogen limitation in the ecosystem (Luo et al., 2004). Elevated CO$_2$ also has a number of direct effects on plant performance, including increasing water use efficiency by increasing biomass accumulation and reducing water loss (Li et al., 2003).

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### Key Words
- Arduino
- CO$_2$ concentration
- CO$_2$ sensor
- Growth chamber
- Microcontrollers
- Valves

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**PREMISE**: A CO$_2$ control system is important for investigating how elevated CO$_2$ affects plant growth. Our automatic CO$_2$ monitoring and control system offers an inexpensive and flexible way to make CO$_2$-enriched environments.

**METHOD AND RESULTS**: Using microcontrollers paired with non-dispersive infrared CO$_2$ sensors, relays, and valves, we developed a low-cost system for monitoring and controlling CO$_2$ levels in growth chambers.

**CONCLUSIONS**: Compared with existing commercially available CO$_2$ control systems, Arduino-based microcontrollers offer affordable access to the data logging of CO$_2$ levels in growth chambers, thereby reducing budget limitations for creating growth conditions with highly controlled CO$_2$ concentrations.

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The atmospheric CO$_2$ level has increased by 30% over the past 50 years and is predicted to increase up to 800 ppm by 2100 (Thuiller et al., 2005; Lindsey, 2020). Elevated CO$_2$ is expected to increase carbon storage and plant productivity due to the enhanced carbon fixation in ecosystems (Drake et al., 2011). In the longer term, rising CO$_2$ would increase the carbon : nitrogen ratio within plant tissue and lead to progressive nitrogen limitation in the ecosystem (Luo et al., 2004). Elevated CO$_2$ also has a number of direct effects on plant performance, including increasing water use efficiency by increasing biomass accumulation and reducing water loss (Li et al., 2003) and can monitor up to 24 chambers. Because earlier infrared sensors required that air samples be desiccated and that samples from each chamber be stabilized before the chamber CO$_2$ level could be assessed and adjusted, these systems can be cumbersome to use. A more recent study described a less expensive CO$_2$ injection system that was regulated by an infrared gas monitor, where CO$_2$ was mixed with ambient air to achieve a target CO$_2$ concentration before being added to the growth chambers (Godfree et al., 2011). The system relied on solenoid valves and flow meters to create the target CO$_2$ levels. However, its reliance on flow meters caused this system to take a longer time to reach the target CO$_2$ level because fresh air and pure CO$_2$ must be mixed before being added to the chambers; therefore, the CO$_2$ level ranges from 500 to 1000 ppm when the target CO$_2$ level is set at 600 ppm. A CO$_2$ control system that does not require CO$_2$ and fresh air to be mixed before being injected into the growth chamber would provide more stable CO$_2$ levels.

Here, we describe a simple, low-cost CO$_2$ control system that uses microcontrollers paired with non-dispersive infrared (NDIR) CO$_2$ sensors to monitor and control the CO$_2$ level in growth chambers. Like other infrared sensors, NDIR sensors rely on the principle that a target gas (e.g., CO$_2$) absorbs a specific band of infrared light while transmitting other wavelengths of light, thus reducing the energy reaching an infrared light detector (Dinh et al., 2016). A challenge with this technology is the need to reduce interference from non-target gases, such as water vapor, although recent
advances in filtering in NDIR sensors have greatly reduced this interference (Yasuda et al., 2012). NDIR sensing technology has therefore been proven to be durable and stable for detecting CO2 in a range of environmental settings; however, because temperature and duration of use can affect sensors (Yasuda et al., 2012), periodic calibration is still essential. Furthermore, the electrical noise associated with growth chambers (especially from lighting systems) can potentially interfere with the communication between a CO2 sensor and its data acquisition system. To avoid this, we paired each sensor with a microcontroller in close physical proximity to the sensor and used serial communication, as opposed to a more electrically sensitive digital communication between the sensor and microcontroller. This setup offers an inexpensive and flexible way of making CO2-enriched environments, including the data logging of CO2 levels in growth chambers, and can easily be modified to accommodate additional sensors and environmental controls.

METHODS AND RESULTS

Our system is based on K30 NDIR CO2 sensors (Senseair AB, Delsbo, Sweden). They are inexpensive and have integrated calibration and communication circuits (see Table 1 for materials list). They are not sensitive to water vapor, meaning there is no need to use a desiccant to absorb water before sampling. NDIR CO2 sensors come in a range of sensitivities and can send digital or analog signals to computer interfaces. The model we used can be calibrated at 0 or 400 ppm CO2 by adding a contact switch to its circuit board (CO2meter.com, 2020). The sensors also easily connect to air supply tubing for calibration. Calibration takes approximately 3–5 min, pumping an air stream of 0 or 400 ppm CO2 air over the sensor, and then closing the appropriate contact on the sensor circuit board. The sensors’ circuit board and all of the other electrical components of our system have standard 2.5-mm electrical connector spacings, allowing the wiring to be connected with header pins and connectors (Dupont, JST type, or screw terminals), which are widely available from electronic parts suppliers.

Because growth chambers generate considerable electrical noise, CO2 sensors need to be in close proximity to their signal processors to ensure that sensor readings do not degrade. In our system, each CO2 sensor is therefore connected to a separate microcontroller mounted on each growth chamber (Fig. 1), which then controls a gas solenoid valve, via a relay switch, on a central manifold connected to a CO2 supply tank (Figs. 2, 3). We used Arduino Uno-type microcontrollers to communicate with the CO2 sensors. We used a data logging shield (Adafruit, New York, New York, USA), a real-time clock, and an SD card socket connected to each Arduino, following the manufacturer’s instructions (Earl, 2013). The large SD cardholder can fit any SD card and store up to 32 GB of information (FAT16 or FAT32 format). The microcontroller can also communicate directly to a computer via a USB or wireless connection, allowing data to be monitored in real time and the microcontroller to be reprogrammed as needed.

### TABLE 1. Materials list to build the automatic CO2 monitoring and control system.

| Materials                        | Cost (US$) | Model no. | Supplier |
|----------------------------------|------------|-----------|----------|
| CO2 sensor                       | $85        | K30       | https://www.co2meter.com |
| Microcontrollerb                 | $20        | Arduino Uno | https://store.arduino.cc/usa/arduino-uno-rev3 |
| Data logging shield              | $14        | 1141      | https://www.adafruit.com/ |
| Eight-channel relay              | $12        | TS0012    | https://www.sunfounder.com |
| ¼-inch NC gas solenoid valveb     | $10        | 2W-025-08 |          |
| Gas manifold                     | $100       |           |          |
| 2.54-mm connectors               | $10        |           |          |
| 22-gauge wire                    | $20        |           |          |
| Tank regulator                   | $200       |           |          |
| Power supplies (9 V, 12 V, 5 V)  | $30        |           |          |
| CO2 supply tank                  | $30        |           |          |

*One per sensor.

*Items without a supplier are widely available from a number of suppliers.

### FIGURE 1. Wiring connections between the K30 CO2 sensor (right) and the Arduino Uno–type microcontroller (left) using serial communication. A JST connector was soldered to the K30 circuit board and a calibration switch was soldered to the board to enable the recalibration of the CO2 sensor. Calibration gas can be passed through the air sampling tube during calibration. The 5 V and ground pins of the CO2 sensor are connected to the corresponding 5 V and ground pins of the microcontrollers (red and black wires, respectively). Pin 8 (RX on the microcontroller) connects to the TX pin on the sensor (yellow wire), and pin 9 (TX on the microcontroller) connects to the RX pins of the CO2 sensor (green wire). The data logging shield (not shown) stacks onto the microcontroller. For use, the sensor is placed in an acrylic box with vent holes and mounted on the side of the growth chamber.
The CO₂ sensors receive their power supply from the microcontroller. Arduino-type microcontrollers can have a range of voltage inputs, but we found that providing 9 V of power to the microcontroller worked best. If supplied with less than 7 V, the microcontroller and sensor may be unstable and fail to record sensor readings. If using 12 V or higher, the voltage regulator of the microcontroller may overheat. Serial communications between the CO₂ sensor and the microcontroller require a four-wire connection (Fig. 1), made via the connectors on the data logging shield stacked on the microcontroller. The sensors can also communicate with the microcontrollers using a digital protocol, which could theoretically allow multiple sensors from many chambers to be connected to a single microcontroller; however, due to the electrical interference in growth chambers, each sensor needs to be placed in close proximity to a microcontroller so serial communication is more stable. The 5 V and ground pins of the sensor are connected to the corresponding 5 V and ground pins of the microcontroller. The transmitting (TX) pin of the sensor is connected to...
brass tubing and T connectors, which are commonly available from with 0.25-inch connections can be attached to a manifold made of voltages, but 12 V models are the most commonly available. Valves trical relays (Figs. 2, 3). Gas solenoid valves come in a range of tank connected to a manifold of solenoid valves activated by elec-

manifold to each growth chamber, and the CO2 is released into the els) in the growth chambers recorded over a 24-h period.

FIGURE 4. An example of CO2 levels (600 ppm and 800 ppm target lev-

eels, monitoring time, valve open time, etc. can be edited within the code based on the experimental conditions. Even a brief injection of CO2 will cause a rapid increase in the CO2 concentration in the chamber; thus, the CO2 level that opens the valve must be set below the desired chamber CO2 concentration and the valve should open for a short enough period to result in a CO2 increase to near the desired concentration. In this example, the CO2 sen-
sors monitor the CO2 level every 30 s. If the CO2 level in a cham-

ber drops below the set value, the corresponding gas valve is set to open for 0.2 s and will not reopen for 30 s, allowing the air to circulate in the chamber before adding more CO2. We tested the system using Conviron A1000 chambers (Conviron, Winnipeg, Canada), which have a 1000-L volume and were set at 24°C and ca. 36% humidity. The growth chambers had their external air supply valves closed and these were also covered in cellophane tape to reduce air leakage. An internal fan provides rapid air movement within the chamber.

Each microcontroller is connected to a relay switch controlling a valve on the CO2 injection system (Fig. 2). Relay boards come in various configurations, and it is convenient but not essential to have a relay board with at least as many channels as there are sensors. Our system uses an eight-channel relay board with its own 5 V power supply; relay boards can be powered by a microcontroller, but this is not recommended due to the relay board’s power consumption. The relay board can be wired such that the relays are in a closed position when not powered, which avoids in-
jecting CO2 into a chamber if there is a power interruption. Each microcontroller 5 V pin is connected to the 5 V pin of the relay board. A digital pin on each microcontroller is connected with a pin on the relay board, with each controlling a different relay switch on the board.

The microcontroller is programmed using Arduino software (https://www.arduino.cc/en/main/software). The code provided here (Appendix S1) is an example of a chamber set to achieve an 800 ppm CO2 concentration (Fig. 4). The chamber numbers, CO2
CONCLUSIONS

Our CO$_2$ control system gives researchers access to a low-cost setup for monitoring and controlling CO$_2$ levels in controlled environmental conditions. The CO$_2$ control system can be run in multiple chambers simultaneously, and we provide a versatile code for the CO$_2$ control system, which can also be used in other areas (e.g., monitoring and controlling temperature, humidity, and soil moisture). The system can be easily adapted for different experimental conditions, and its low cost allows the replication of conditions to be achieved for more statistically robust growth experiments.

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AUTHOR CONTRIBUTIONS

H.C. designed and executed this work under the guidance of J.M. H.C. wrote the article, and J.M. was involved in the editing and correcting of the manuscript. Both authors approved the final manuscript.

SUPPORTING INFORMATION

Additional Supporting Information may be found online in the supporting information tab for this article.

APPENDIX S1. Arduino code for a growth chamber CO$_2$ control system.

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