

**Development of a Prototype Algal Reactor for Removing
CO₂ from Cabin Air**

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Development of a Prototype Algal Reactor for Removing CO₂ from Cabin Air

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Controlling carbon dioxide in spacecraft cabin air may be accomplished using algal photobioreactors (PBRs). The purpose of this project was to evaluate the use of a commercial microcontroller, the Arduino Mega 2560, for measuring key photobioreactor variables: dissolved oxygen, pH, temperature, light, and carbon dioxide. The Arduino platform is an open-source physical computing platform composed of a compact microcontroller board and a C++/C computer language (Arduino 1.0.5). The functionality of the Arduino platform can be expanded by the use of numerous add-ons or 'shields'. The Arduino Mega 2560 was equipped with the following shields: datalogger, BNC shield for reading pH sensor, a Mega Moto shield for controlling CO₂ addition, as well as multiple sensors. The dissolved oxygen (DO) probe was calibrated using a nitrogen bubbling technique and the pH probe was calibrated via an Omega pH simulator. The PBR was constructed using a 2 L beaker, a 66 L box for addition of CO₂, a micro porous membrane, a diaphragm pump, four 25 watt light bulbs, a MasterFlex speed controller, and a fan. The algae (wild type *Synechocystis* PCC6803) was grown in an aerated flask until the algae was dense enough to be used in the main reactor. After the algae was grown, it was transferred to the 2 L beaker where CO₂ consumption and O₂ production was measured using the microcontroller sensor suite. The data was recorded via the datalogger and transferred to a computer for analysis.

Nomenclature

<i>PBR</i>	=	Photobioreactor
<i>DO</i>	=	Dissolved O ₂
<i>RTC</i>	=	Real Time Clock
<i>BNC</i>	=	Bayonet Neill-Concelman Radio Frequency connector
USB	=	Universal Serial Bus
CO ₂	=	carbon dioxide
ppm	=	parts per million
SD	=	Secure Digital
O ₂	=	oxygen
BLAST	=	Basic Local Alignment Search Tool
I/O	=	Input/Output

I. Introduction

Algal photobioreactors are used naturally and synthetically in many climate conditions to fix carbon dioxide or to produce biomass. PBRs utilize the photosynthetic capability of algae to fix CO₂ to produce O₂, which may be used in space in life support functions. In order to ensure survival of the crew, the algae must be grown in conditions

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where pH, light levels, and a carbon source are optimal. These factors must be monitored via a cost-effective system. In this study, a system was developed to analyze the bioreactor and produce data relevant to the algae's capacity to remove CO₂ via photosynthesis. This system involved the use of Arduino microcontrollers and multiple sensors to detect dissolved O₂ levels, carbon uptake, and pH. The algae, *Chlorella Vulgaris*, was used due to its ability to grow quickly and to use a high gas exchange rate. *Chlorella Vulgaris* is also high in nutrients and can be used as a food source for astronauts.

II. Materials and Methods

A. Arduino Components

In order to test the ability of the algae to fix CO₂ and produce O₂, a data logging system needed to be developed to detect multiple variables. The Arduino Mega 2560 was used because it contains 54 digital inputs/outputs, and 16 analog inputs. The analog inputs provide the ability for sensors to be interfaced with the Arduino Mega board. The Arduino Mega 2560 was equipped with the following shields: datalogger, BNC shield for reading pH sensor, a Mega Moto shield for controlling CO₂ addition, as well as multiple sensors. The datalogging shield contained a SD card input that supports FAT16 or FAT32 format, and an RTC clock that supports a button cell. The SD card recorded and retained all the data. The BNC shield supported coaxial cable connections from a pH probe, and contains two variable resistors for calibration of the pH probe. The MegaMoto motor shield regulates the valve that allows for additional CO₂ to enter the system.



Figure 1. Setup of the Arduino Shields

The Arduino Mega 2560 controlling platform also supports a USB connection and a power jack. The USB connection serves as a method of mounting programs or sketches onto the board itself via a compiler known as Arduino 1.0.5. The power jack allows for the board to be maintained on an external power supply. This is useful when the system has to be running for several days or weeks without a computer. This particular Arduino model has advantages over other arduino models. For example, this arduino has an additional reference voltage commands such as INTERNAL1V1, which sets the board to 1.1 V reference. This helps when a specific sensor is sensitive to a small voltage range.

The Arduino Mega 2560 allows for more digital inputs/outputs and analog inputs than other Arduino models. This microcontroller platform is also cost effective and smaller than other controller platforms such as OPTO-22.

The sensors attached to the Arduino system needed calibration in order to output accurate data. The sensors were calibrated using a multiple point calibration method. The DO probe was calibrated by measuring the voltage difference between an O₂ zero environment and an O₂ rich environment. A saturated solution of sodium sulfite was used as the O₂ zero environment and the voltage difference was recorded as the probe was taken out of the solution. An equation was then generated and used in the program for the DO probe. The pH probe was calibrated using a pH simulator set to pH 4 and pH 10. The simulator was set to pH 4 and the smooth trim was changed until the serial monitor displayed pH 4. Next, the simulator was set to pH 10 and the rough trim was changed until the serial monitor displayed pH 10. A linear map function was used to program an equation into the BNC shield. The CO₂ sensor was calibrated using a similar method. The CO₂ sensor was zeroed using pure N₂ and then air was blown on to the sensor. The voltage was recorded as the CO₂ sensor acclimated to the new environment. An equation was developed and the result of the equation was recorded on the SD card.

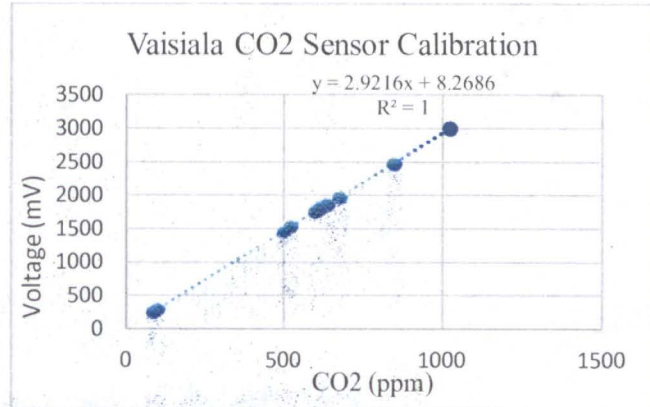


Figure 2. Calibration of the CO₂ sensor under CO₂ poor conditions and CO₂ rich conditions

B. Development of Code

The code for each individual part was written in Arduino 1.0.5, a language similar to C/C++. The code consists of structure, variables, and functions. Structures contain compound operators, bitwise operators, pointer access operators, Boolean operators, and control structures. Variables include constants, data types, scopes/qualifiers, and utilities. Functions include digital I/O, analog I/O, time, and math. The most utilized segments were analog I/O, control structures, and data types.

The environment used to write code for the Arduino is the Arduino 1.0.5. This program was downloaded from the main Arduino website (Arduino.cc). The program divides code written into files called sketch. Each sketch has its own code that gives the Arduino a specific function. For example, the SD cardinfo sketch allows the user to check the status of the SD card inserted into the Arduino.

Each shield had its own code and an example code was obtained from the manufacturer.

C. Photo-Bioreactor Setup

The PBR consisted of multiple components that helped facilitate constant movement of nutrient poor algae and nutrient rich algae. The 66 L, airtight intake box functioned as a reservoir of CO₂. The algae was supplied CO₂ via a

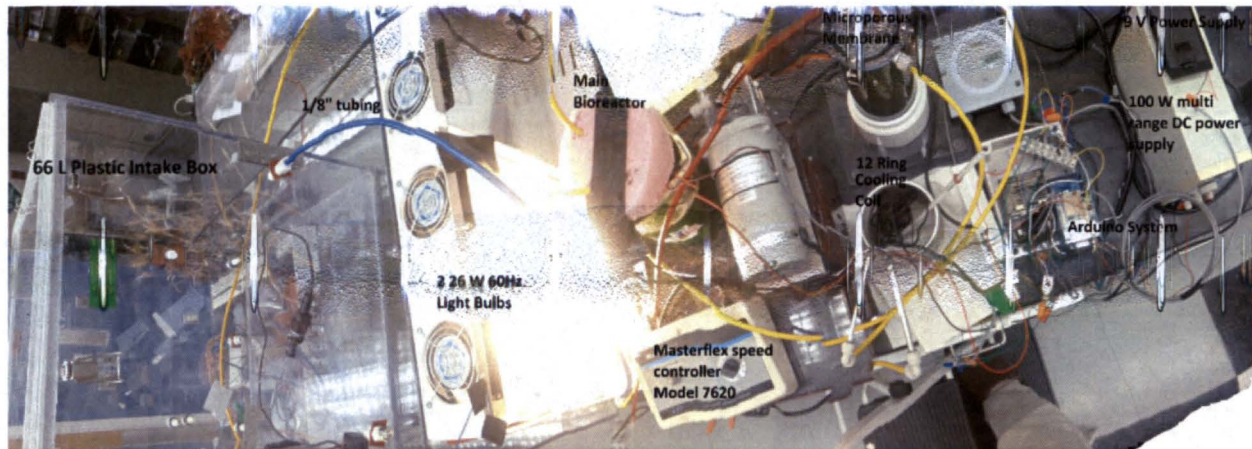


Figure 3. Bioreactor Setup

micro porous membrane from the intake box via a diaphragm pump. O₂ produced was removed via the micro

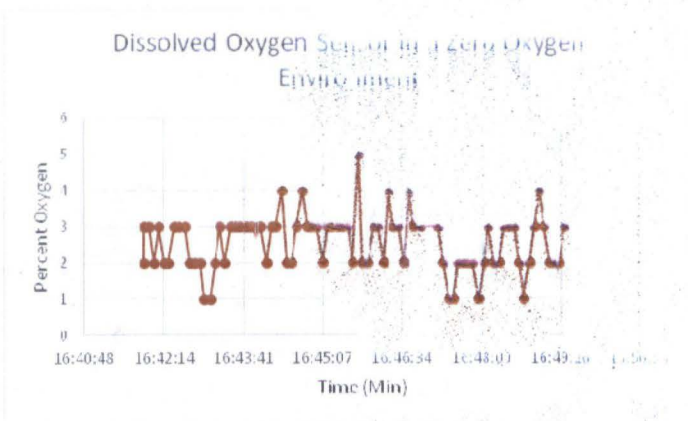
porous membrane and transferred into the intake box. A dissolved O₂ sensor was placed in the intake box to record the amount of O₂ obtained. The intake box contained a CO₂ sensor, which monitored and regulated CO₂ flow into the micro porous membrane. The micro porous membrane consisted of 30 3/10 cm micro porous tubes. If the CO₂ sensor read above 2000 ppm, then the addition of CO₂ was stopped. If the CO₂ sensor read below 2000 ppm, then CO₂ was added to the system. The main reactor included a pH sensor, a DO sensor, a temperature sensor, and a photosensor. The main reactor contained nutrient rich algae and exposed them to warm light. A Master Flex speed controller model 7620 transferred the nutrient rich algae from the main reactor to the microporous membrane. This membrane contained 3/10 cm 30 micro porous tubes that allowed the O₂ to leave and the carbon dioxide to enter. The nutrient rich algae was moved through a 12 ring metal heat coil and pushed into the main reactor chamber. Tubing that connected various part of the system were 1/8 inch with 1/8 plastic fittings. A Styrofoam cap was constructed to allow wires and tubing into the main reactor.

Multiple sensor are needed in order to measure the efficiency of the algae and the efficiency of the reactor. temperature probe, one light probe, Two DO probes, one CO₂ probe, and one pH probe was distributed into the system. A DO probe was placed in the main reactor and a DO probe was placed in the intake box. A temperature probe, a light probe, and a pH probe was placed in the PBR. The CO₂ probe was placed into the intake box with a DO probe.

D. The Algae *Synechocystis* sp. PCC 6803

Synechocystis has been studied extensively because of its ability to adapt to a wide variety of conditions. This strain can grow without the need for photosynthesis when given a carbon source to grow. *Synechocystis* also contains a complete genome sequence, which is useful to compare to other genome sequences via BLAST.

E. Modifications to the Photo-Bioreactor



The sensors and shields attached to the main Arduino Mega 2560 were functioning properly; however, the DO probes was noisy because the 10 bit resolution A/D converter of the Arduino 2560 could not resolve the low signals emitted by the DO probes (40 mV).

Fig 2. Sensor noise due to Poor A/D converter resolution.

In order to obtain meaningful biological responses, the Arduino Mega 2560 microcontroller was substituted by a Campbell Scientific CR23X datalogger. A new program was written , however, the same calibration functions were used. This system allowed for a higher resolution, which allows for more accuracy. This micrologger was connected with all the probes and the micrologger was set to record every 10 seconds for 3 days while the lights were left on. The data was recorded in a batch file. The batch file was imported and graphed on Microsoft Excel 2013.

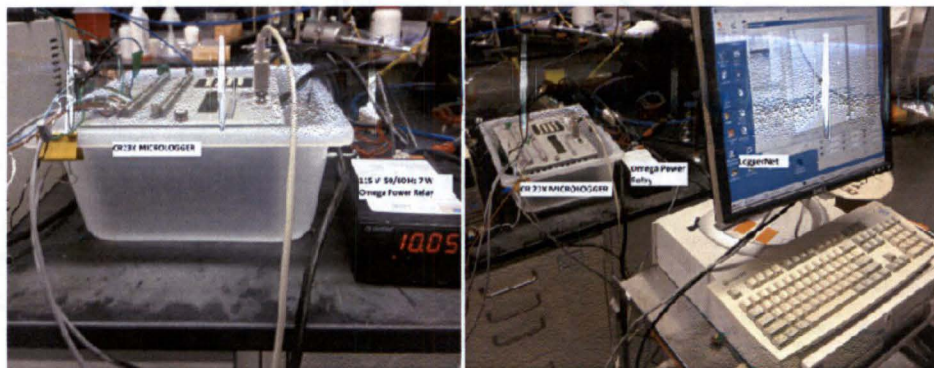
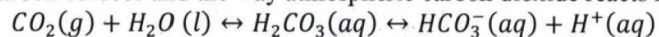


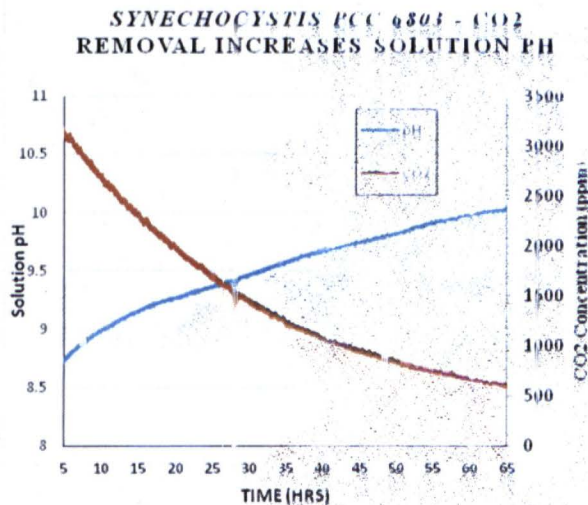
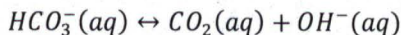
Fig 3. The Campbell Scientific CR-23X Datalogger was used to monitor the PBR sensors at a higher resolution

F. Results

Various relationships were observed during the time period. Carbon dioxide levels and pH were inversely proportional to each other. As carbon dioxide levels fell, pH increased. This trend is due to the ability of *Synechocystis* to convert carbon sources and the way atmospheric carbon dioxide reacts in water.



As carbon dioxide from the chamber is added through the micro porous membrane, the carbon dioxide ionizes to become bicarbonate and H^+ in water. The algae utilizes the CO_2 for photosynthesis and produces additional hydroxyl groups, which raises the pH.



A proportional relationship was observed between pH and dissolved oxygen. When pH increased, dissolved oxygen concentrations increased.

Another trend that developed was a sinusoidal movement of dissolved oxygen in solution. This is an indication of a

Variables that remained constant were light and temperature.

G. Discussion

The Arduino Mega 2560 was used successfully to read and record the data collected by the light, CO_2 , solution temperature, and pH sensors. However, we found that the dissolved oxygen sensors could not be accurately read by the 10-bit A/D (analog to digital) converter of the Arduino Mega 2560. In order to obtain meaningful biological data, a higher resolution A/d board or an amplifier board were needed. A DO probe shield can be used but we did not have enough time to implement it, thus a CR23X datalogger was used instead.

The CR23X data was used to demonstrate that the algal PBR can indeed remove CO_2 and that this CO_2 removal results in an increase in solution pH (Fig 4). This graph also shows that pH control may also be needed for ensuring optimal growth as algae growth is reduced at high pH (ref).

Conclusions

The Arduino platform is a very flexible platform due to the many shields available for many functions required for operating and monitoring key environmental parameters of an algal PBR, including solution pH, temperature, CO_2 concentration and for recording data. There was a large learning curve in programming the each of the functions needed, but we found that these were achievable and that an algal PBR system can be implemented with the Arduino Mega 2560 given enough time and with the proper shields.

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Periodicals

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All of the preceding information is required

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