

Algal Photobioreactor

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Authors:

Ares Torres, Sandra Pena, Britton Stark, Aditya Nirgun

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Department of Biological and Agricultural Engineering,
University of California, Davis

Abstract

The purpose of this project is to construct an algae photobioreactor that supports the growth of *Chlorella sorokiniana*. The first phase of the project involved generating potential project designs, analyzing their effectiveness using a Kepner-Treogoe decision analysis in order to decide which system would be the most effective. The second phase of the project was the construction period that resulted in a completed prototype. The third and final phase of the project was to evaluate the effectiveness of the design through standardized testing of its operation. The testing chosen was a combination of cost, size, and how fast the algae was able to grow in the chosen system. The design chosen in this project resulted in an inexpensive, portable, and simplistic system that was able to grow algae for a period of time. Due to inappropriate fertilizer, the ammonia levels in the system were increased dramatically which caused the algae's growth rate to be terminated and all samples showed the death of the algae. Based on the prior data, it is believed that the system would still work if the proper fertilizer was used in conjunction with the system.

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Introduction

As the demand for alternative fuel sources increases, research on the production of algae as a source of liquid biofuels is becoming imperative. Algae as a source of alternative fuels opens the gateway to other research that will improve the way current fuels are created and used. There is extensive research being done on the use of algae as a means to meet the world's requirement for fuels. Most of this research is underfunded due to the lack of public knowledge on the topic and the scepticism of the public on the use of renewable energies over that of fossil fuels. Our goal is to meet this demand by creating an algae photobioreactor that is inexpensive, portable, and efficient. We aim to produce at least $20 \text{ g m}^{-2} \text{ day}^{-1}$ of *Chlorella sorokiniana* using a bioreactor that does not exceed 3 m^2 . After experimentation, it was concluded that there was zero growth rate in the algae colony. This was caused by the fertilizer used to provide nutrients to the algae. Potential overdose of the fertilizer resulted in large amounts of ammonia which could have contributed to the death of the algae. Since this death was caused by an unintended error, the use of this kind of technology to grow algae is still very pertinent to the modern world as fossil fuels become increasingly finite and the world's consumption continues to grow exponentially.

Background

Algae are obligate aerobes, meaning they use some oxygen to grow. This means our task is to create a bioreactor that is an aerobic fermenter (McDuffie, 2013). The algae performs photosynthesis in its nutrient rich water and sun lit environment. It stores the energy produced as carbohydrates and these surplus starches are in turn stored as fatty acids and lipids (Patel, et al., 2012). These lipids are the biofuel we are seeking to isolate. The other carbohydrates are ethanol based (Oilgae, 2014). To achieve this goal with an efficient and sustainable procedure, we have to optimize the design of our bioreactor using a colony of *Chlorella sorokiniana*. *Chlorella sorokiniana* is “a unique single-celled freshwater micro algae. Its characteristic emerald-green color is due to its high content of chlorophyll. *Chlorella sorokiniana* reproduces at an extremely fast rate, renewing into four new cells every 17 – 24 hours” (Oilgae, 2016). The species thrives in temperatures between 20-41°C and its most optimal

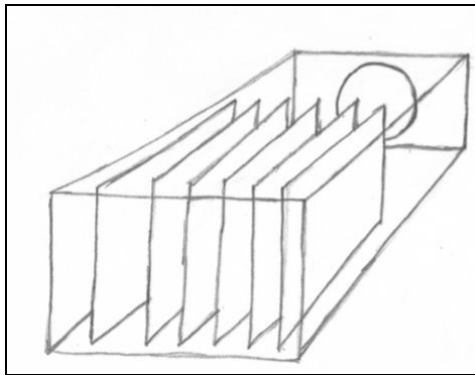
temperature is 38°C (Bechet, et al., 2013). It is known that heavy metals such as copper kill algae and other aquatic plants when introduced into the water column. In addition, high ammonia concentrations can kill an algae population if it is not balanced with nitrifying bacteria which are able to convert ammonia into nitrates.

Problem Statement

The original problem statement was to take existing bioreactor technology and modify it to meet our project limitations. After careful consideration of the parameters at hand and the time allotted, the revised problem statement was to create a novel algae bioreactor that met our project budget while still maintaining peak efficiency. Our project's major constraints consist of a budget of less than \$100 and its overall footprint of less than 3m². Our team assumed finding a liquid medium for the algae colony would be easier than the project presented.

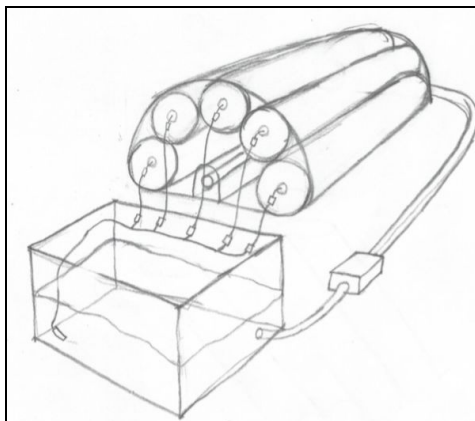
Initial Design Alternatives

Flat Plate



This system is an aquarium style tank that has the main innovative feature of flat mesh sheets that increase the surface area available for the algae to grow. A light source will be placed perpendicular to the mesh sheets with a reflector on the opposite side of the container to allow for proper lighting distribution. The main drawback for this design is its difficulty in providing even distribution of nutrients due to the mesh sheets.

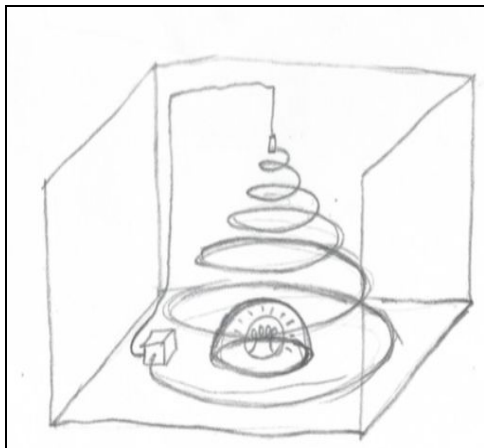
Horizontal Tubular



This system consists of a set of tubes that run parallel to the ground and a small tank that feeds into these tubes. These tubes are transparent and house the algae. Water circulation is provided by a water pump that is situated in the small tank and pushes water through the tubes that

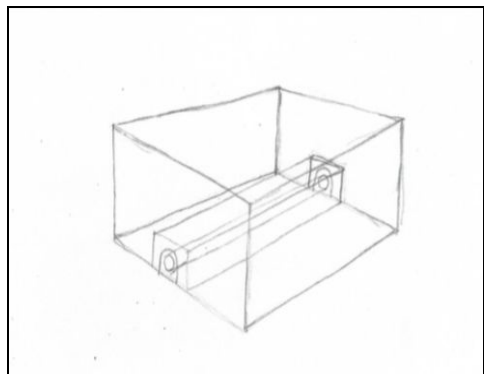
then exits back into the small tank. Carbon dioxide and nutrients are released into the small tank and a light source is placed under the tubes to provide light to them.

Spiral Design



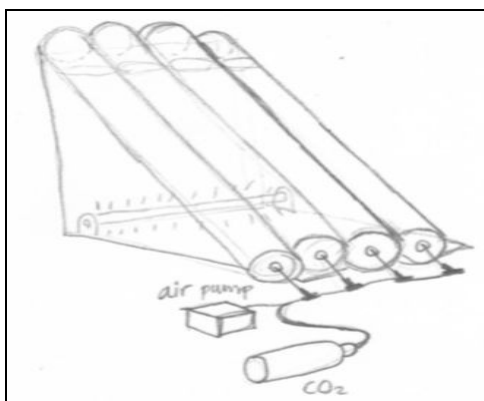
This system consists of a flexible frame in the shape of a cone surrounding a light source. Flexible tubing is then spiraled along the frame and algae is pumped into these tubes. A water pump forces water up and through the tubes until it is cycled through the bottom of the spiral. Carbon dioxide and nutrients will be supplied through an opening at the top of the spiral.

Submerged Light



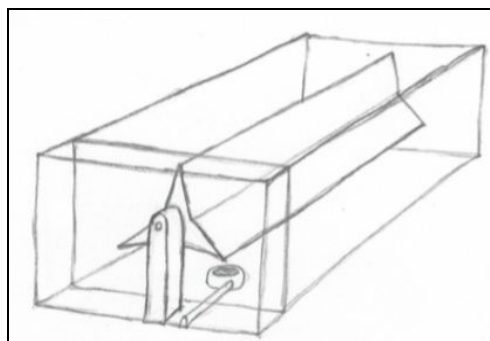
This system consists of a single container that will hold the algae and lighting source within it. The main feature of this system is the lighting source, which is contained in clear material that is submerged in the tank and allows for greater light exposure. An HOB filter will be placed on one corner of the tank to provide water circulation, evenly distributing nutrients throughout the tank.

Vertical Tubular



This system is similar to the horizontal tubular design, however the tubes are laid straight and angled at 45° from the ground. A light source is placed underneath the tubes. Carbon dioxide will be delivered via airline tubes that connect directly to an air pump system. Nutrients will be fed into the water through a separate tube system.

Water Wheel



This system consists of a glass tank that contains a multi-pane water wheel that spins about an axis parallel to the longest edge of the tank. The water wheel's rotation will be powered by a motor located outside of the tank. Carbon dioxide and essential nutrients will be released into the water by tubes and circulated by the water wheel.

Milk Bag



This system consists of a transparent plastic sheeting that allows light to penetrate from all sides. A small airline will be inserted through a small port on the bag to create water circulation and introduce both carbon dioxide and oxygen. A port on the roof of the bag will allow for sample extraction and fertilizer insertion. The bag will be braced by several supports to increase the weight tolerances.

Design Selection

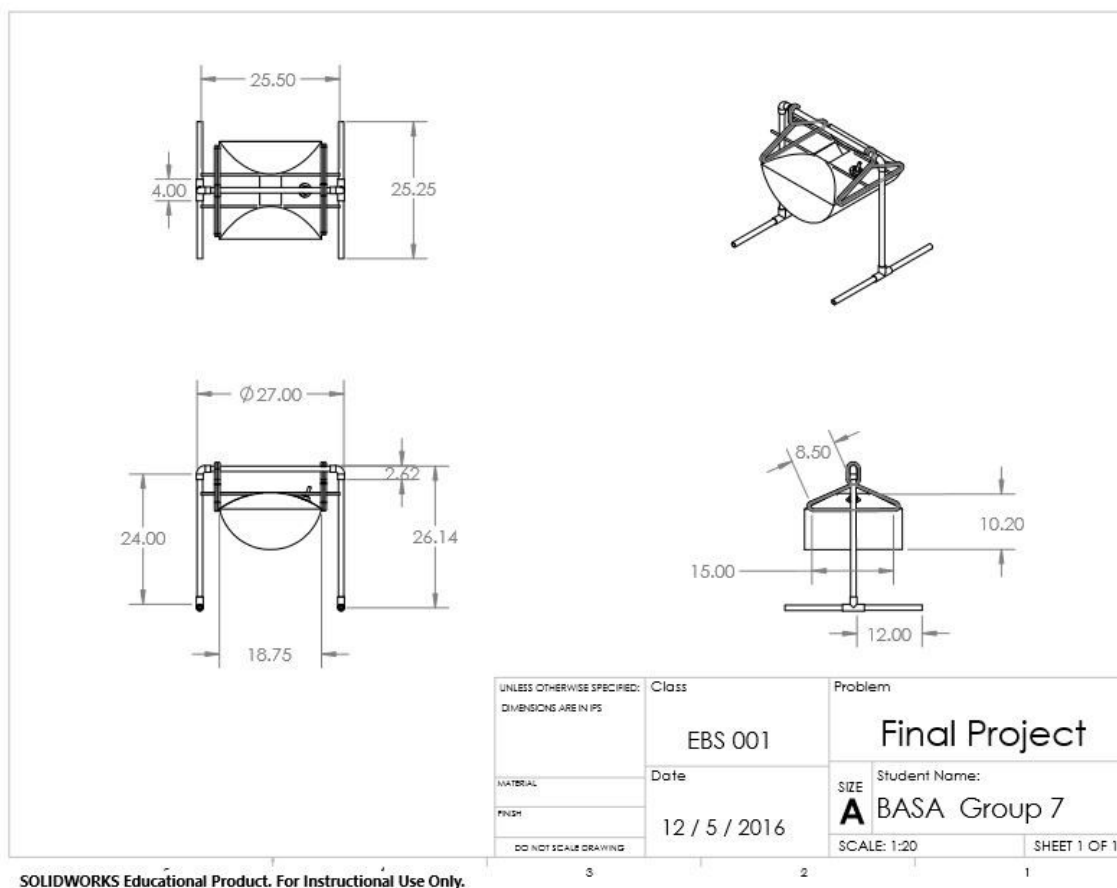
Table 1. Analyzes alternative designs

K-T Decision Analysis															
Alternative Solution		A: Submerged Light		B: Flat Plate		C: Water Wheel		D: Horizontal Tubular Design		E: Spiral Design		F: Vertical Tubular Design		G: Milk Bag Design	
Musts															
1. Size < 3 sq. Meters		Yes		Yes		Yes		Yes		Yes		Yes		Yes	
2. Manufacturing Cost < \$100		Yes		Yes		Yes		Yes		Yes		Yes		Yes	
3. Efficient Water Circulation		Yes		No		No		Yes		No		Yes		Yes	
Keep Design?															
Wants	Weight	Rating	Score	Rating	Score	Rating	Score	Rating	Score	Rating	Score	Rating	Score	Rating	Score
Low Energy Consumption	5	7	35					5	25			5	25	7	35
Aesthetic	3	4	12					8	24			7	21	4	12
Noise Level	4	8	32					6	24			6	24	9	36
Isolated Environment	7	3	21					9	63			9	63	7	49
Simplicity	6	9	54					2	12			5	30	9	54
Efficient Carbon Dioxide	9	6	54					8	72			7	63	7	63
Total Score		Total	208	Total		Total		Total	220	Total		Total	226	Total	249

The “musts” were based on the assignment criteria and the efficiency of water circulation through each system. The “want” factors were based on the key components of a successful bioreactor as well as consumer oriented design initiatives. The most important factors of a bioreactor, such as an isolated environment and efficient carbon dioxide distribution, were

weighed the highest. Low energy consumption and simplicity were rated second highest. Finally, the “wants” that do not contribute to the system directly were rated the lowest.

Design Description



SOLIDWORKS Educational Product. For Instructional Use Only.

Description of design: This design involves a bag suspended from a frame with lighting and air pump circulation. The frame is built from PVC and clothing hangers attached to the bag with duct tape, dowels, and string for additional support. The bag is repurposed from its original use as a container for the milk dispenser in the campus dining commons. We inserted our air pump airline tubing into the spout on the milk bag. The light is positioned above the bag using the dowells.

Fabrication: We assembled the PVC pieces for the frame and cut some of the PVC to make two sets of legs for a stable base. We duct taped the ends of the bag and dowels to the hangers and tied string around the underside for support. Finally, we cut back flaps at the top of the bag and taped them to the dowels.

Operating Conditions:

Table 2. Optimal conditions for system efficiency

Operating Conditions	Description
Temperature	This design is operational between the temperatures of 20°C and 41°C.
Amount of water in the bag	This design is equipped to hold up to 4 liters of water however at this volume there is noticeable straining on the hangers and duct tape supporting the bag. The ideal water amount we found was 3 liters.
Sunlight	Keeping sunlight exposure to a minimum is preferred to better control both the temperature and the light intensity of the system. In addition, UV light exposure via sunlight can potentially degrade the plastic bag, leaking harmful chemicals into the water.

Design Testing: Methods

We started with a small scale test where we tested the relative growth rate of algae in both reverse osmosis water with fertilizer and water from a greenhouse. Both types of water were

placed in containers which were fed with airline tubes providing circulation. The containers were then placed in front of the same light source we planned to use for the final design. We planned to visually evaluate the growth in the two water conditions. The greenhouse water supported the algae culture but we did not observe any further growth. The reverse osmosis water with fertilizer resulted in a thin bubbly film at the surface indicating an apparent death of all the algae. We concluded that the MiracleGro fertilizer caused the death of the algae because it contained 0.07% water-soluble copper. We could not use the greenhouse water for our final trials because it already contained an inseparable culture of algae that would have interfered with our results. We decided to use deionized water and a different fertilizer that did not contain copper ingredients.

In our final test we used 15 mL of Household Plant Food in 3 liters of water. For each sample, we pre-weighted the paper filters and recorded its mass. We then extracted 1 mL of algal water, placed it onto the filter, dried it until there was no liquid, and then weighed and recorded the solid matter left behind. These samples and measurements were taken everyday for 6 days.

Design Testing: Results & Analysis

Table 3- Weight of filter paper before placing algae (initial weight). Final weight consists of dry mass of algae and filter paper.

	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6
Initial Weight (g m^{-2} day $^{-1}$)	.8115	.8302	.8303	.8167	.8255	.8379
Final Weight (g m^{-2} day $^{-1}$)	.8222	.8399	.8402	.8252	.8350	.8473
Weight of algae	.0107	.0097	.0099	.0085	.0095	.0094

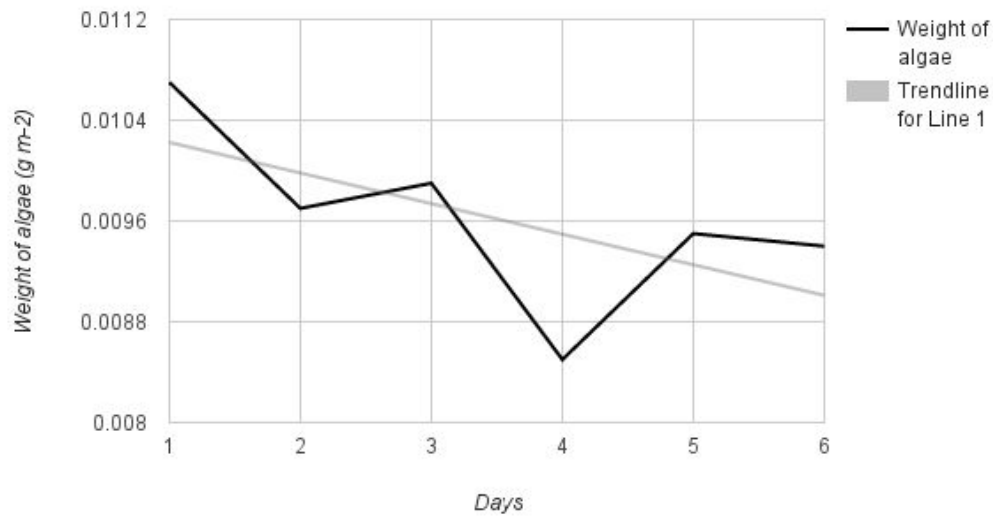


Figure 1- Growth rate of algae ($\text{g m}^{-2} \text{ day}^{-1}$) over the course of 6 days.

As shown in the data and indicated by the best fit line there was no growth in the algae throughout the 6 days. We believe the cause of this were the high levels of ammonia concentration, leading to the death of the algae. The ammonia was introduced into the system via the fertilizer. By adding less fertilizer or introducing nitrifying bacteria we could have fixed the issue.

We used a linear regression model to perform a goodness of fit test for our data. Our null hypothesis was that our value $\rho \leq 0$ or that there was evidence that there was negative or no change in the daily sample weights and our alternate hypothesis was $\rho > 0$ or that there was evidence that there was an increase in the weights of the daily samples. Our significance level was $\alpha = 0.05$. The formula for the linear regression test statistic is $t_s = r \sqrt{\frac{n-2}{1-r^2}}$ with degrees of freedom = $n - 2$. The formula for the Coefficient of Determination is $r^2 = \frac{\sum(y_i - \bar{y})^2 - \sum e_i^2}{\sum(y_i - \bar{y})^2}$. Our Coefficient of Determination is 0.4019, our test statistic is 1.63946 and a DoF of 4, our 95% confidence interval is $\{-0.0006541, 0.0001648\}$, our p-value is between .10 and .05 thus we failed to reject our null hypothesis. This means that we are 95% confident that the true slope of

the line of best fit for our data is within our interval and as a result, we cannot determine that there was significant evidence that our data showed a growth in the weights of the daily samples.

Economic Analysis

Table 4. List of items and their cost in dollars

Item	Cost
PVC pipes	\$9
Air pump	\$15
Air stones	\$3.99
MiracleGro Fertilizer	\$4.33
Total	\$32.32

Table 5. External Costs

Airline tubing	\$5
Hangers	\$4
Light fixture	\$40
Milk bag	\$2
Dowells	\$6.66 (Shop time: 5 minutes)
String	\$0.25
Total	\$57.91

Design Manufacturing Information

After certain computations, it will take approximately \$200 to build a full scale unit of this design. Although the design is uncomplicated to construct, the materials used may not have enough strength to hold a full scale unit. Materials like duct tape and hangers are fairly weak and not recommended to be used long term. In addition, there is no way to scale up the milk bag

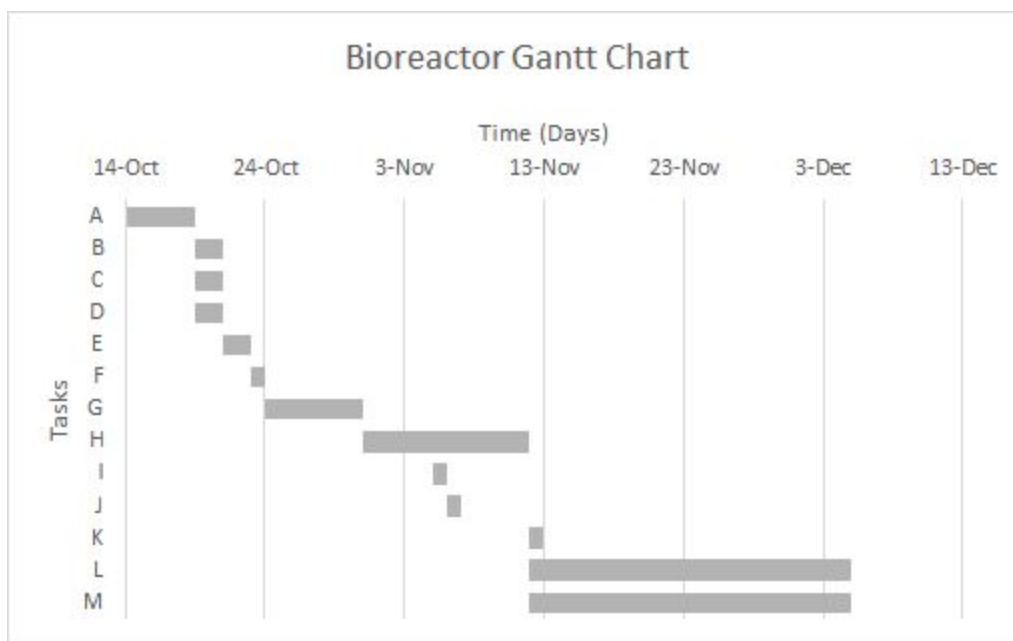
itself requiring the use of multiple milk bags which could increase the size and complexity of the design.

Future Recommendations & Conclusions

After completing experimental tests and analyzing the data, our team concluded that our design had several room for improvement. If the project were to be redone, it would be beneficial to find means of controlling the system's temperature. Adding nitrifying bacteria would help reduce the amount of ammonia in the water. Testing in both natural and artificial light to determine which light source would be more beneficial for this design. Our team determined that the addition of copper kills algae. Mechanically, our project meets the demands in our problem statement, but our medium for the algae requires further adjustments.

Project Management

Our team was able to effectively complete many of the tasks on time according to the Gantt Chart. However, our team encountered a few major obstacles. After creating a list of supplies to be purchased, we realized that obtaining tubes in the diameter and height that we required would be too expensive for our budget. We then created an entirely new design with a focus in using inexpensive materials. After obtaining our new materials, we began testing different types of water to determine the best medium for our algae. Both trials failed due to existing copper concentration in our fertilizer water and our greenhouse water being contaminated by a different type of algae. Our team then bought a different fertilizer without copper and used the rest of our algae to make our final trial. However, that trial failed as well because our fertilizer introduced too much ammonia into our water. If given the chance to do this again we would begin the testing of different mediums sooner.



Team Norms and Statement of Individual Team Member Responsibilities

When the project was first assigned, our team designated certain tasks for each of us to complete. As the project progressed, these tasks became more specific and crucial to the completion of the assignment. As a team, we each contributed ideas to the project's final design, the steps needed to complete its construction, and made sure we stayed focused and on schedule. Britton Stark created a sketch of the project's logo, designed the project on Solidworks, and provided materials to the project's design. Ares Torres was in charge of formatting the team's assignments. He collected the different types of water that were needed to test the algae's growth, took samples of the algae water, and assisted in recording data. He also created the project's logo on Solidworks that was later printed using a 3D printer. Aditya Nirgun sketched potential project designs, purchased essential materials from Ace Hardware Store, completed and recorded tests on the different water samples. Sandra Pena was in charge of arranging meetings, completing the meeting minutes, submitting preliminary assignments, and assisted purchasing materials.

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