Large-scale Biomass Production Using a Novel Cyanobacterium

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Background

With a fast generation time of 10 days, high biomass conversion and greenhouse gas fixation ability, and the capacity to produce lipids, cyanobacteria are viable platforms for biofuel production. These photosynthetic microorganisms offer a significant reduction in total emissions as they absorb carbon dioxide in cultivation ponds and are a good source of renewable energy.

Objectives

The focus of this study was to scale up the bioreactor and cultivate *Fremyella diplosiphon* in the greenhouse and outdoors. Naturally available brackish waters with salinity 12g/L was used. Biomass was harvested, biocrude extracted using hydrothermal liquefaction, and analyzed using Gas Chromatography Mass-Spectrometry (GC-MS).

Methods

Initial fish tank studies

Cultures at an OD_{750} of 0.2 were initially grown in fish tanks. Three replicated treatments were maintained and OD_{750} measured every three days for a period of 15 days.

Large-scale cultivation in 20 liters carboys

Modifications made:

(i). Instead of using tubes that start floating, plastic pipettes were cut and used to the desired length. This resulted in bigger bubbles, which improved aeration by preventing the settling of the cultures.

(ii). A drawback that was observed in carboys with open vents resulted in excessive evaporation. A modified pipette to reduce the vent size helped to lower evaporation rate.

(iii). A starting OD_{750} of 0.2 enabled the culture to establish quickly. There was no contamination observed in cultures.

Scale up cultivation

Growth and survival of *F. diplosiphon* strain SF33 was tested by scaling up to 15 L cultures in 20 gallon-sized carboys. Growth as a measure of OD_{750} was measured every three days for a period of 12 days. Temperature in the greenhouse and outdoor conditions was monitored using a data logger (Elitech GSP-6G).

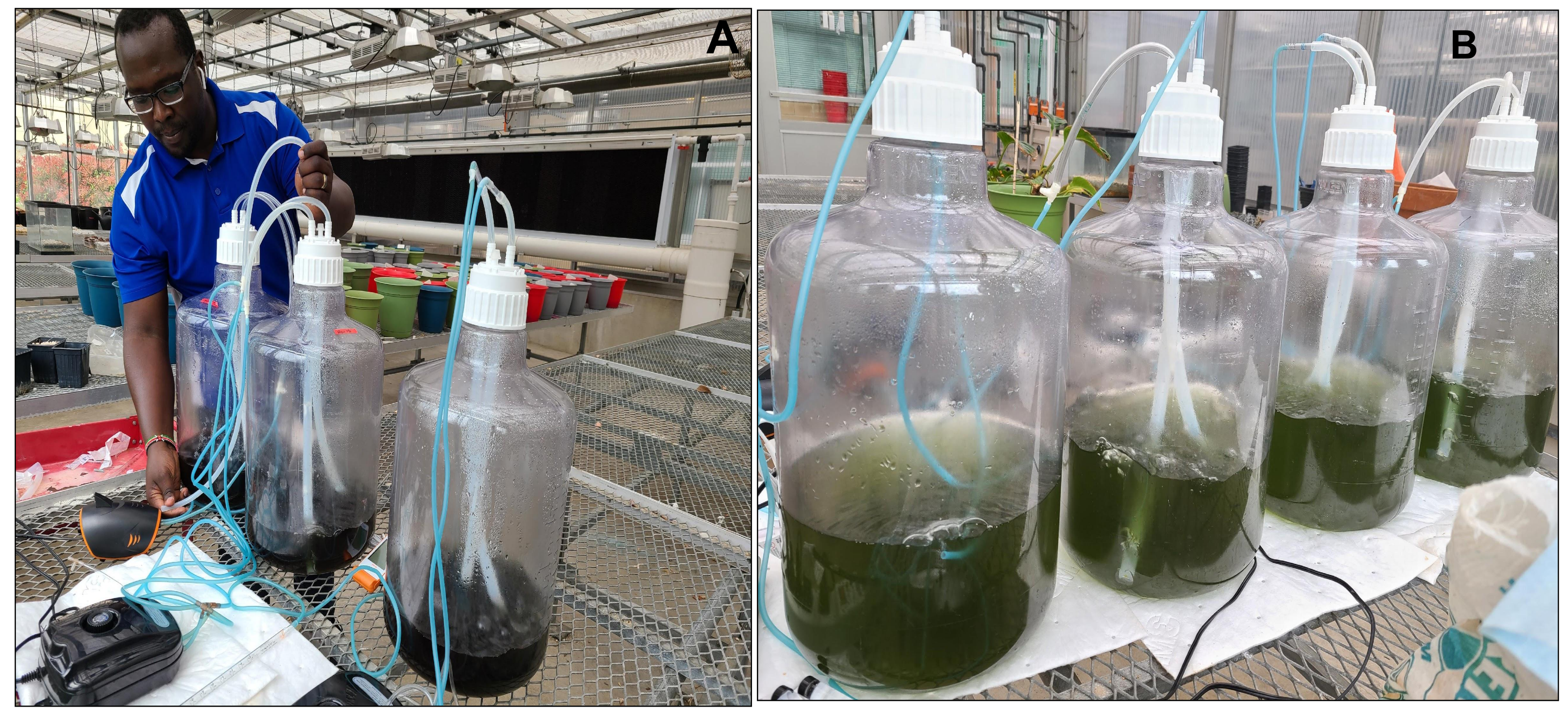
Pigment and lipid and quantification

- Pigments were quantified at the beginning and end of cultivation period using a microplate reader.
- Biocrude was extracted using hydrothermal liquefaction and analyzed using GC-MS.

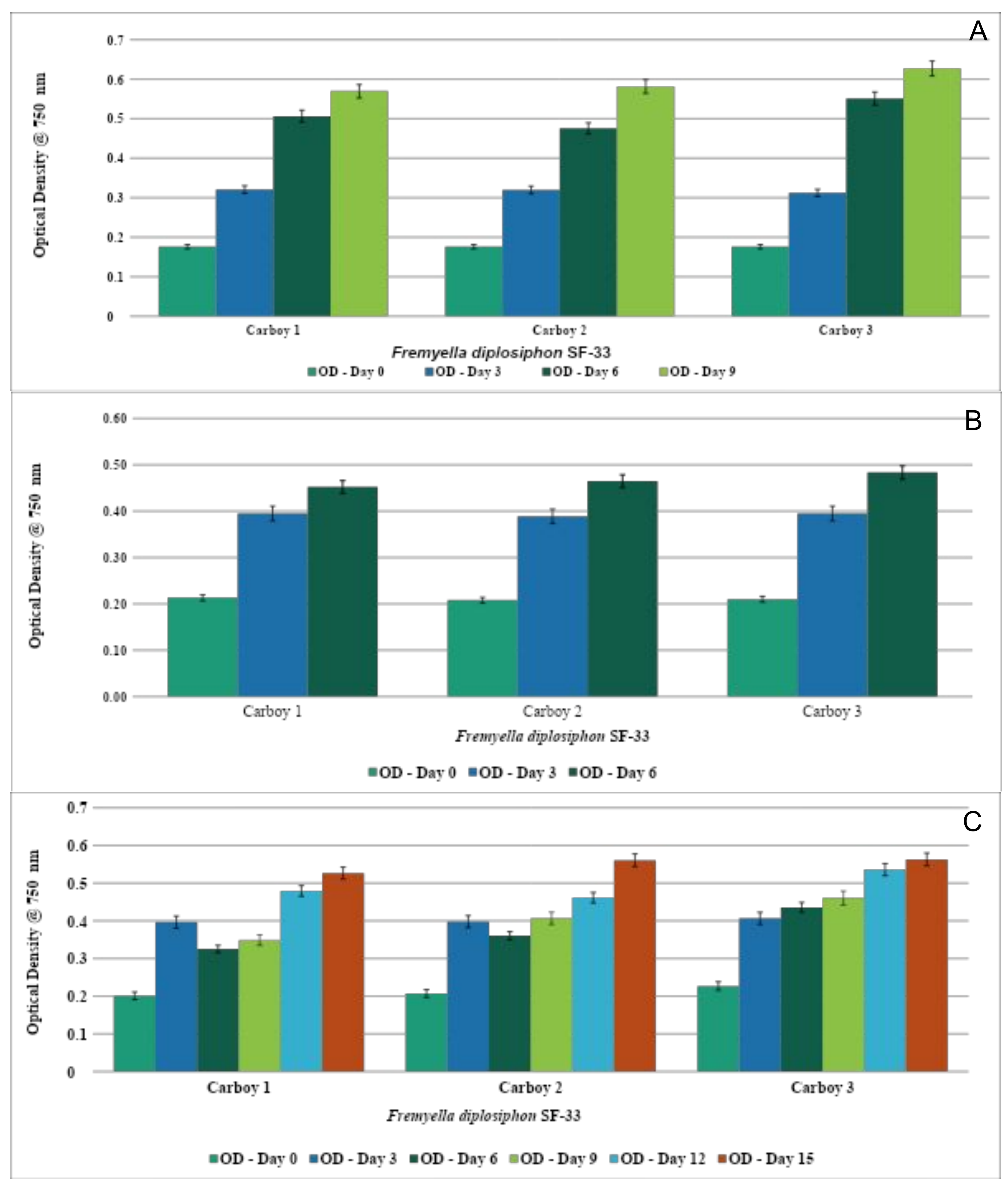
Results and Discussion

Growth: Cultures grown both in greenhouse (5, 10 and 15L) and outdoor (15L) conditions demonstrated the ability of the strain to grow under fluctuating environmental conditions, ranging from 13 to 32° C. This indicates that the strain is capable of further scale up process in outdoor conditions, greatly enhancing its potential cultivation for biofuel production.

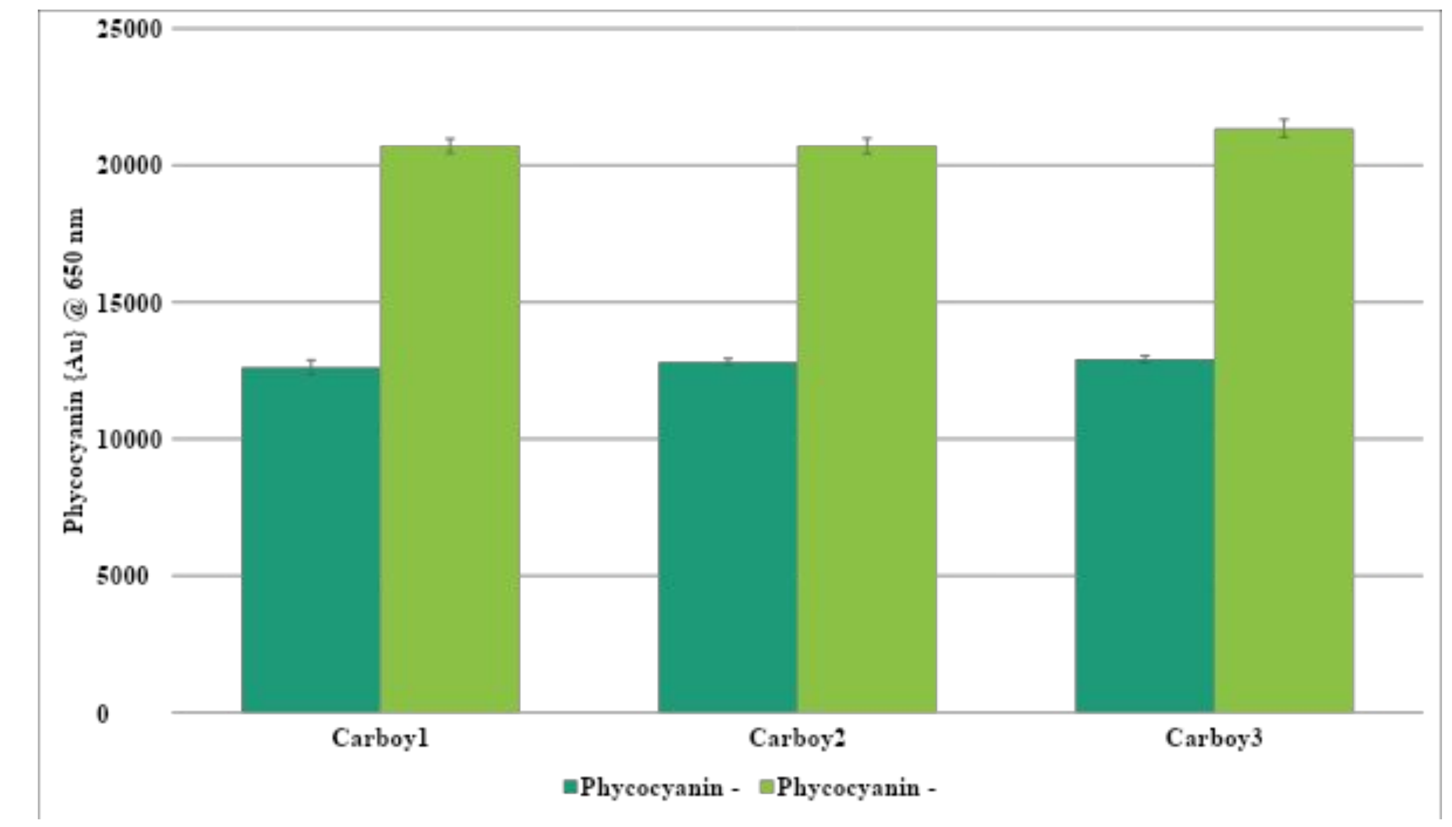
Pigments: There was a significant increase in the phycocyanin levels on day 12 in 5, 10 and 15 L carboys grown in the greenhouse and 15 L carboys outdoors. In addition to biofuels, other high value co-products were identified, increasing potential revenue channels. Phycocyanin is one example, which is of great interest to the nutraceutical market due to its antioxidant properties. **Lipids:** Biocrude analyzed using GC-MS revealed the precursors for a variety of bioenergy applications.



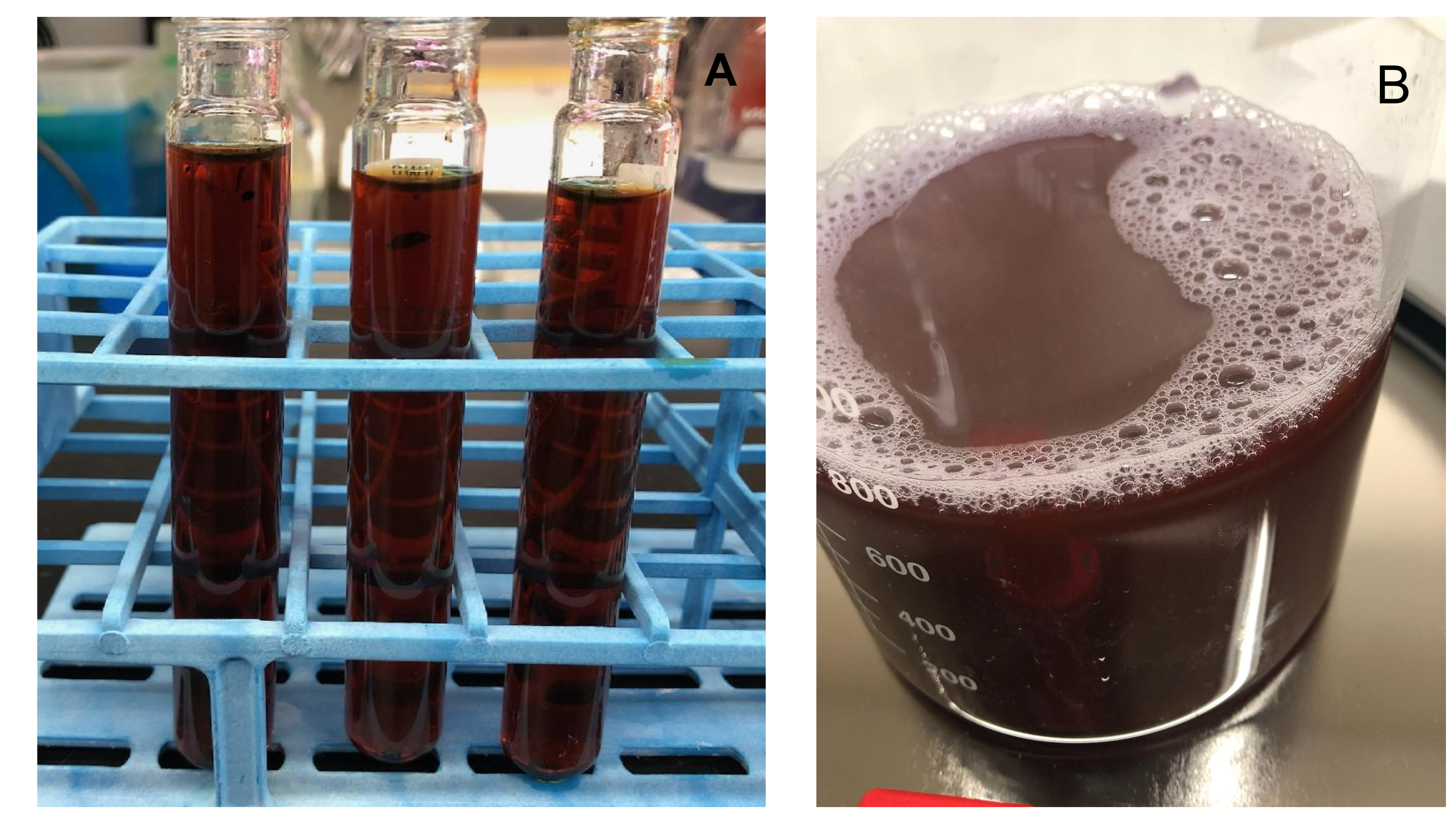
Carboy cultivation of *F. diplosiphon cultivation* in the greenhouse. (A) 5 liter carboy culture, (B) 10 liter carboy cultures.



Growth of *F. diplosiphon* SF-33 grown in 20L carboys. (A) 5 liter, (B) 10 liter, and (C) 15 liter culture



Phycocyanin quantification in *F. diplosiphon* strain SF-33 at an excitation of 590 nm and emission of 650 nm.



***F. diplosiphon* SF-33 (A) extracted biocrude via hydrothermal liquefaction and (B) pigments from biomass**

Conclusions

With the current initiatives taken, an era of using biofuels as an alternative to fossil fuels is on the horizon. Future studies will be aimed to use hydrothermal liquefaction as a method for thermochemical conversion of cyanobacterial biomass that will lead to the production of biocrude. Thus, processing time as well as cost reduction will lead to a sustainable biofuel process.

Future Work

Future Studies: After scale-up at Patuxent Environmental and Aquatic Research Laboratory (PEARL), we will perform American Society for Testing and Materials analyses to determine the resultant biofuel's properties and compare it to industry standards.

Acknowledgement

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