Conferences

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The Green Solution: Optimizing productivity of bi-substrate microalgae cultures

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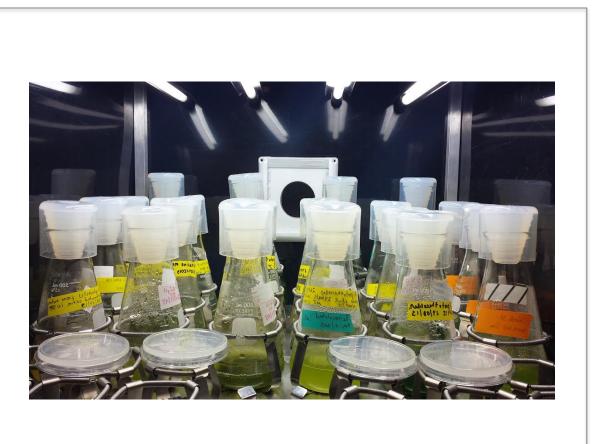
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The Green Solution **Optimizing productivity of bi-substrate microalgae cultures**

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Background

Glycerol is a waste generated from biodiesel production. We can capitalize on its low-cost and availability by utilizing it as a carbon source for cultivating Chlorella Protothecoides, an algae strain known for thriving under heterotrophic conditions. Microalgae is often employed for CO_2 capture, biofuel production and most importantly, it can be processed for antioxidants and lipids. These products are often found in health supplements and food additives, with a current market value of US\$2.1 billion.



Goal

Determine:

- Feasibility of recycled biodiesel glycerol as an alternative carbon substrate to glucose in microalgal biomass production
- The effect of changing glycerol to glucose ratios in achieving an optimal condition for maximizing biomass production (100:0, 25:75, 50:50, 25:75, 0:100)
- Interplay of nitrogen deficiency on optimal glycerol to glucose ratio (10:1, 40:1, 80:1,120:1)

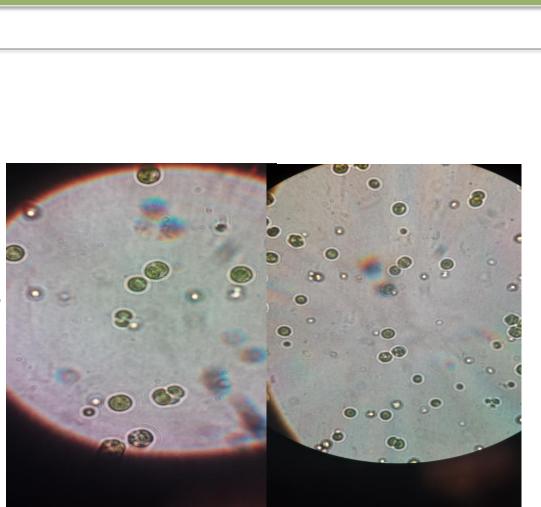
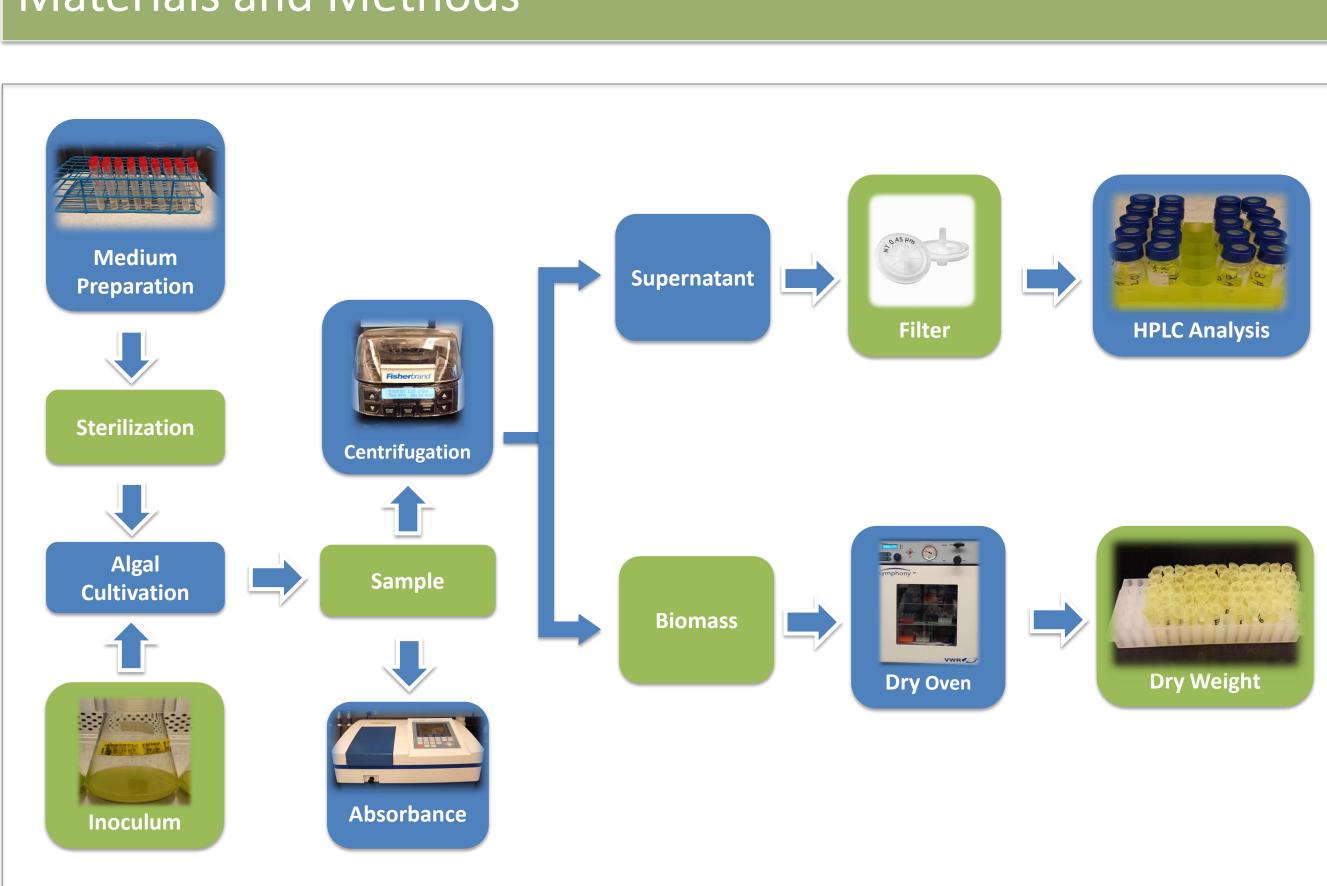


Figure 1: *Chlorella protothecoides*

Materials and Methods



Results

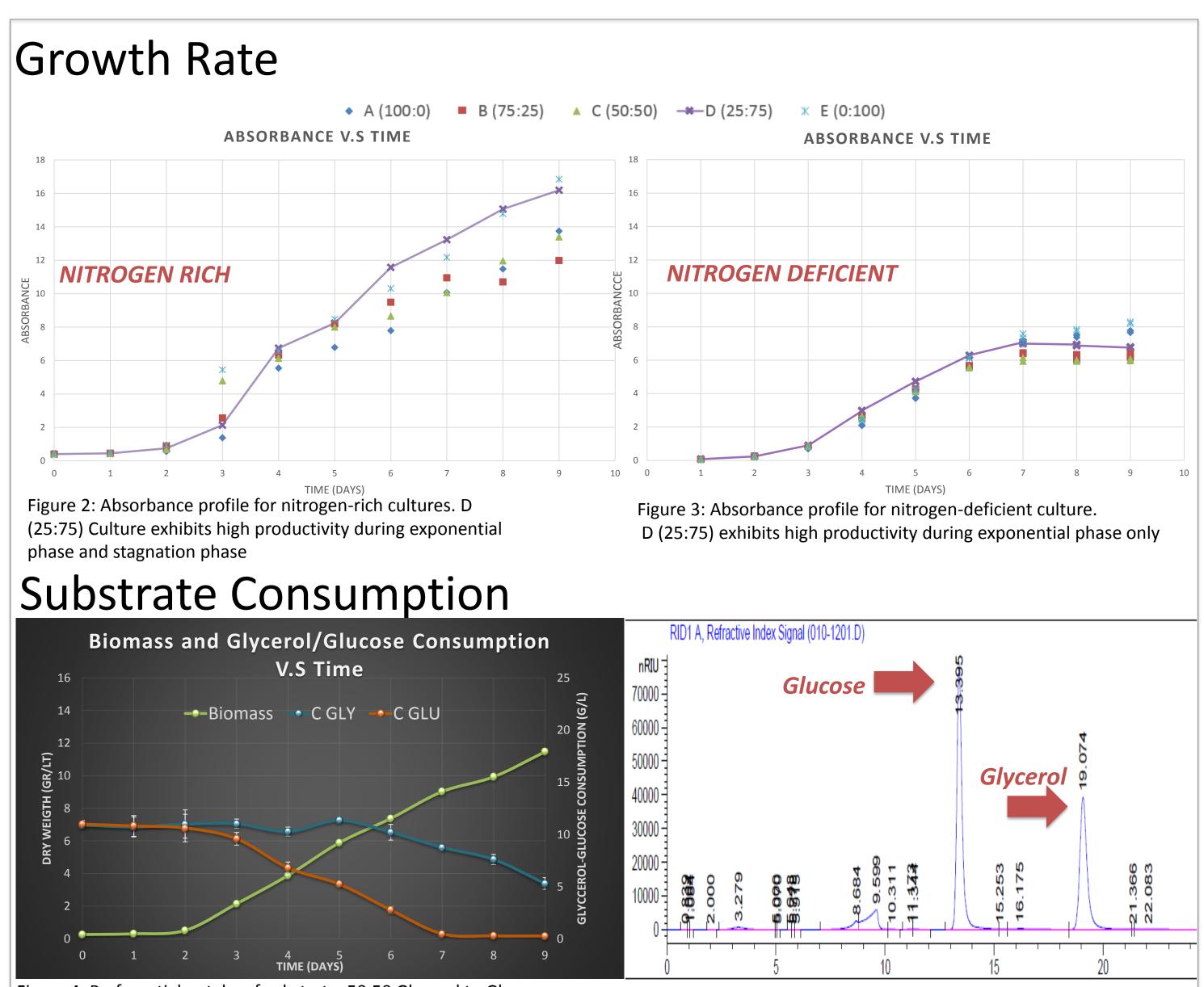


Figure 4: Preferential uptake of substrate, 50:50 Glycerol to Glucose

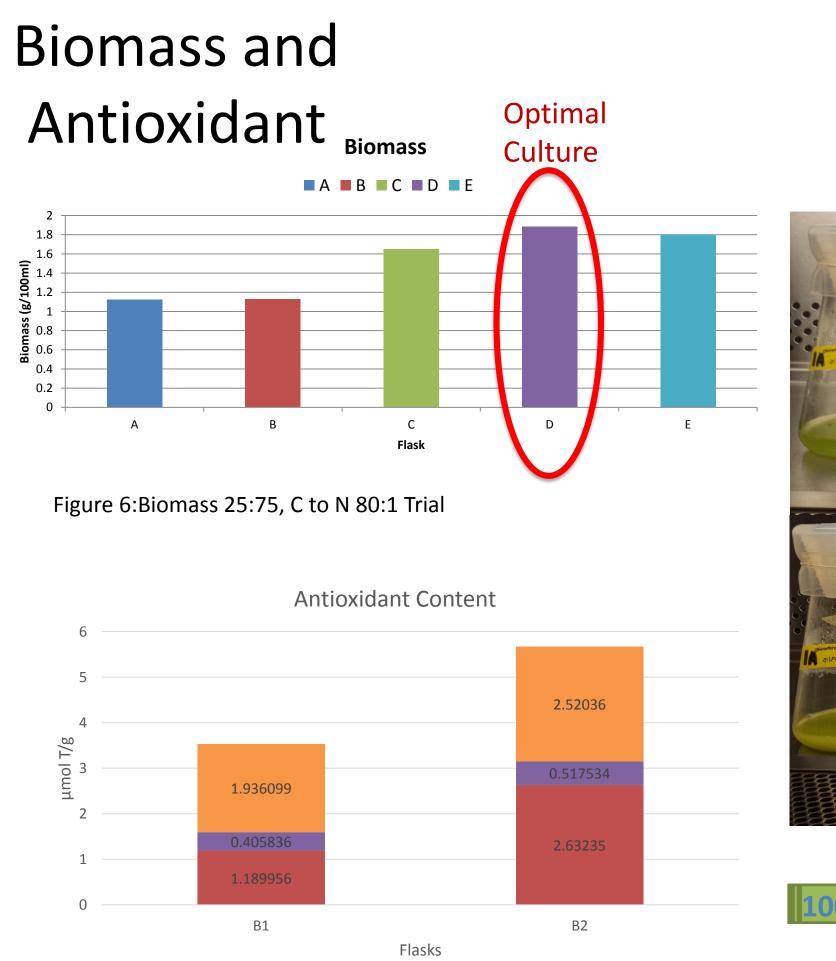
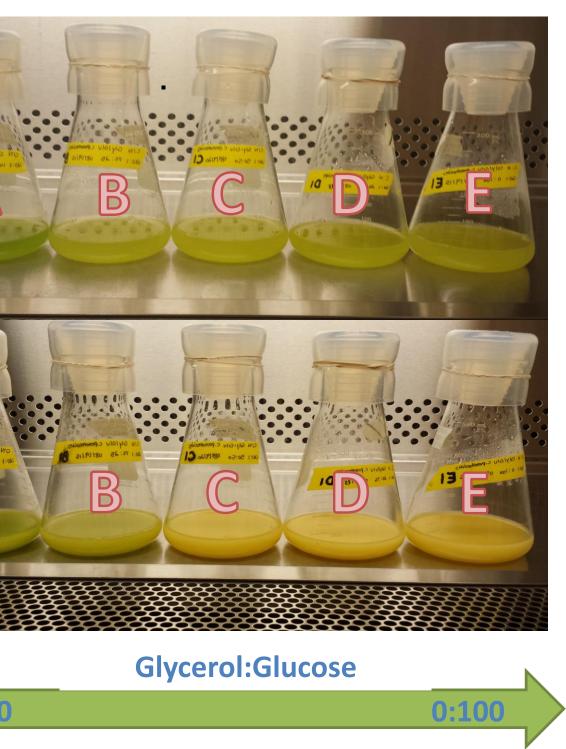


Figure 8: Top (Day 3), Bottom (Day 9) Variation in Chlorophyll content as a function of Glycerol to Glucose ratio.

Figure 7: Antioxidant content for Glucose/Glycerol 25:75, C to N 80:1 Trial

■ Hexane ■ Ethyl Acetate ■ Water

Figure 5: Liquid Chromatograph of medium supernatant



Conclusions

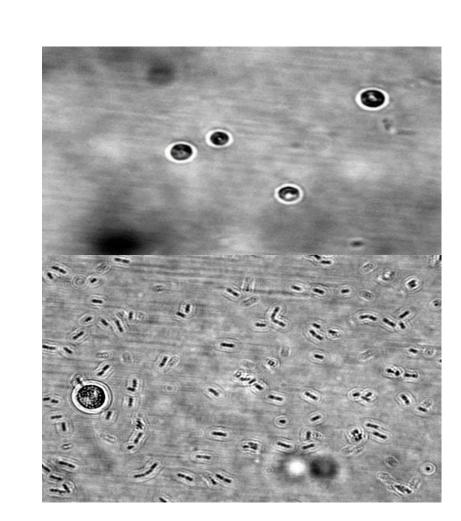


Figure 9: (Top) Healthy culture vs. (Bottom) Contaminated culture

Future Work

- Evaluate Antioxidant, Chlorophyll-A, Chlorophyll-B and lipid compositions of extractable biomass
- Characterize and create Kinetic model of optimal ratio between substrates and nitrogen
- Additional trials to confirm finding and recalibrate results

References

[1]. Heredia-Arroyo, T., Wei, W., & Hu, B. (2010). Oil accumulation via heterotrophic/mixotrophic Chlorella protothecoides. *Applied biochemistry and biotechnology*, 162(7), 1978-1995. [2]. Shi, X. M., Zhang, X. W., & Chen, F. (2000). Heterotrophic production of biomass and lutein by Chlorella protothecoides on various nitrogen sources. Enzyme and Microbial Technology, 27(3), 312-318. [3]. Shi, X. M., Liu, H. J., Zhang, X. W., & Chen, F. (1999). Production of biomass and lutein by Chlorella protothecoides at various glucose concentrations in heterotrophic cultures. *Process biochemistry*, 34(4), 341-347. [4]. O'Grady, J., & Morgan, J. A. (2011). Heterotrophic growth and lipid production of Chlorella protothecoides on glycerol. *Bioprocess and Biosystems engineering*, 34(1), 121-125. [5]. Chen, Y. H., & Walker, T. H. (2011). Biomass and lipid production of heterotrophic microalgae Chlorella protothecoides by using biodiesel-derived crude glycerol. *Biotechnology letters*, 33(10), 1973-1983. [6]. Cerón-García, M. C., Macías-Sánchez, M. D., Sánchez-Mirón, A., García-Camacho, F., & Molina-Grima, E. (2013). A process for biodiesel production involving the heterotrophic fermentation of Chlorella protothecoides with glycerol as the carbon source. Applied Energy, 103, 341-349

Acknowledgements





Optimal ratios are 25:75 Glycerol to Glucose, 10:1 Carbon to Nitrogen 25:75 Glycerol to Glucose exhibits

- highest productivity rate
- Nitrogen deficiency results in Biomass reduction
- Glycerol maintains Chlorophyll pigment within *Chlorella protothecoides*
- Glycerol rich cultures are more resistant to contamination





I would like to thank everyone in the Bioprocess Development lab for their insight and guidance.