The University of the West Indies,

Cave Hill Campus,

Bridgetown, Barbados

Biofuels Research at Cave Hill

Principal Investigator: Professor Winston F Tinto

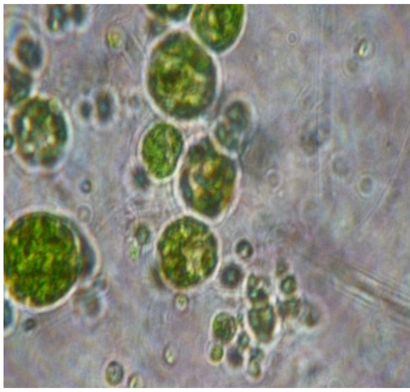
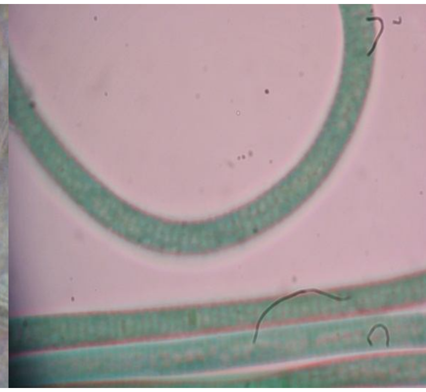
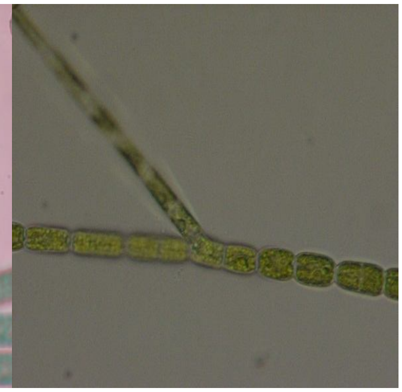
Introduction

Petroleum-based fuels contribute to global warming and are predicted to expire within this century. There is a growing need for alternative renewable fuels to meet future energy demands. Biodiesel is increasingly regarded as a feasible alternative to petroleum-based fuels. Usually, biodiesel is derived from soybean oil in the US and rapeseed oil in Europe, with varying quantities of other plant oils, most notably *Jatropha* oilseed, being used in some other countries. Bioethanol is also being proposed as a biofuel to replace fossil fuel by utilizing sugarcane (mainly Brazil) or corn (mainly US) as biomass feedstock. Biobutanol on the other hand, is preferable to bioethanol since it can be used without retrofitting existing vehicles. Also, butanol is not as corrosive as ethanol and does not absorb water to the same degree as ethanol and can mix with gasoline in any proportion. However, these sources cannot realistically meet forecasted fuel demands due to competition with food crops for prime agricultural land and size of supply. Biodiesel from microalgae may be a viable alternative for producing fuel in the quantities required, due in part to microalgae's greater biomass productivity than conventional plant oilseed. A major advantage of using biodiesel is that it does not contribute as much to greenhouse gas (GHG) emissions as fossil-based fuels. Cellulosic ethanol and butanol will make a major contribution to future energy demands once economical ways of converting fibre to fermentable sugars can be found. The biofuels research at Cave Hill involves the bio-prospecting for high lipid producing microalgae from freshwater, brackish water and seawater. Another area involves the bio-prospecting for fungi and bacteria that can convert fibre, especially from high fibre energy cane, into fermentable sugars. This will involve screening existing sugarcane plantations for the microorganisms that are involved in the decay of sugarcane fibres under natural conditions. Marine fungi from diverse sources will also be investigated.

Microalgae Research Project

Research on biodiesel production from microalgae indicates that some algae are capable of growing rapidly producing large quantities (roughly 20 to 50% of dried weight) of neutral lipids, also known as triacylglycerols (TAGs), when grown under nitrogen-deficient conditions. Further, some of these microalgal species that produce large amounts of oil can grow in wastewater, brackish water and seawater, which minimises their freshwater usage. However, there are a number of challenges in implementing a biofuels system that is based on microalgal biomass. This includes very limited and incomplete studies on life-cycle assessment (LCA), limited knowledge of growing rates under unconventional conditions, the problems associated with cultivation using open-pond systems versus closed systems using different photobioreactor designs, and the necessity for improvement of harvesting and processing methods. The majority of research reported in the literature to date is concerned with downstream activity such as large scale processing of microalgal biomass and the production of biofuel. However, little emphasis is placed on bio-prospecting to obtain promising algal strains from diverse environments. Moreover, very little is known about indigenous microalgal lipid productivity in the Caribbean. The use of indigenous algae is preferable because of the lowered risk to biodiversity and the necessity to use algal strains that are native to the environment in which they will be cultivated.

Biofuels research at Cave Hill commenced in 2011 and the first major objective was to carry out research on high lipid producing microalgae from terrestrial, brackish water and the marine environment. The work was initiated with collection of samples from the soil and fresh water since this was most convenient. Fourteen single-cell and filamentous microalgal samples have been collected from freshwater locations across Barbados. They include species from the genera *Chlorella, Oscillatoria, Spirogyra* and *Rhizoclonium.*

Chlorella sp. *Oscillatoria sp.* *Rhizoconium sp.*

Work has now commenced on collecting microalgae from brackish water and the marine environment. This should yield a greater diversity of these organisms. Among the microorganisms to be targeted would be the blue-green algae or cyanobacteria, since these are very versatile at producing lipids and they can also produce compounds that can be useful in the nutraceutical, cosmetic and pharmaceutical industries. These high value co-products can be of economic importance while striving to develop biofuels from microalgae. One of the major objectives of this project will be to establish a microalgae culture collection that can be exploited for intellectual property development.

Algal cultures growing in the Laboratory.

Energy Cane Research Project

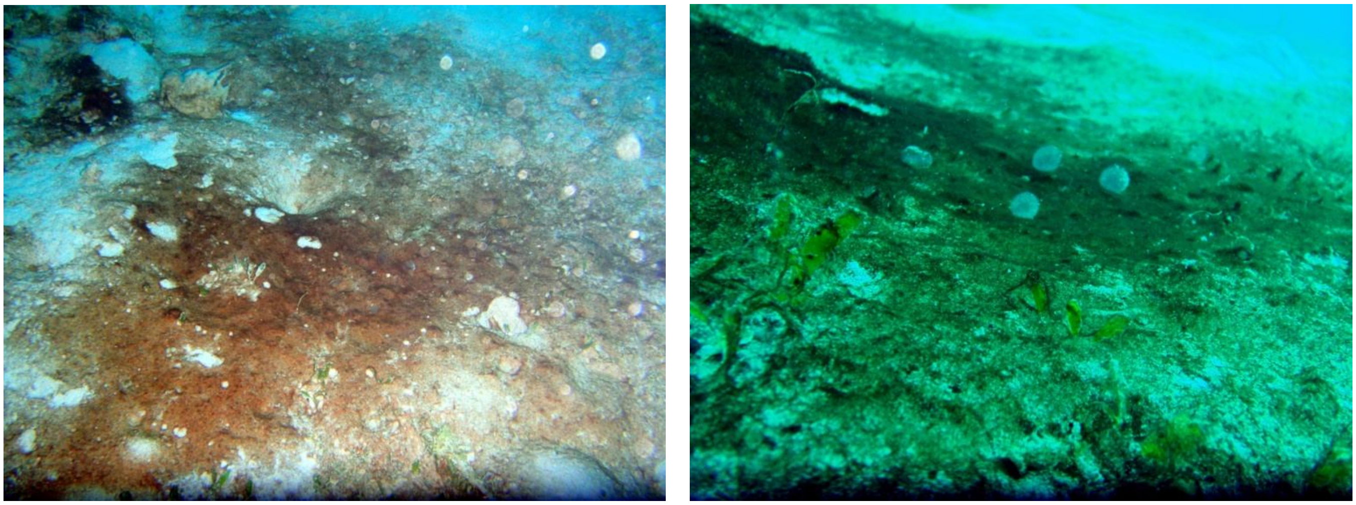
The use of the cane fibre as a feedstock for bioethanol and biobutanol production through fermentation is a promising option that is being actively investigated in some laboratories worldwide. This process, using genetically modified *Clostridia*, can also co-produce acetone and hydrogen gas. However, the use of naturally occurring microorganisms is a preferred option due to the risks associated with using genetically modified organisms.

This research project seeks to establish high fibre multipurpose cane varieties and at the same time identify useful fungal and bacterial strains that can efficiently convert these fibres to fermentable sugars. Another approach being actively investigated world-wide is enzyme bio-prospecting. This involves searching for fibre degrading enzymes from microorganisms found in the gut of plant eating organisms.

The major initial focus of this research project involves the bio-prospecting for fungi and bacteria from sugarcane plantations. Marine fungi will also be collected from symbiotic marine invertebrates, seaweeds and marine sediments. A culture collection will then be established and systematically investigated for microorganisms that can convert the sugarcane fibres to fermentable sugars to produce bioethanol, biobutanol. Lignin and other high value products will also be isolated in the process.

Value-added Co- Products

The only way that microalgae can presently lead to a viable economic production system for biofuels is by co-production of high-value products. Many microalgae produce high-value polyunsaturated fatty acids (PUFA) and other chemicals that can be used as nutritional supplements, pharmaceuticals and cosmetics. These microalgae, especially the blue-green algae (cyanobacteria) are known to produce natural products with anti-bacterial, anti-viral and anti-tumor properties, among others. Also, *Streptomyces* bacteria, some of which can also be high lipid producing, can be found in the soil and the marine environment. These microorganisms are responsible for the production of the majority of antibiotics in use today. Due to the increasing observation of multi-drug resistant strains of pathogenic microorganisms, we are currently



Massive colonies of Cyanobacteria growing off the coast of Barbados at Brown's Beach

investigating the use of marine *Streptomyces* and cyanobacteria as a source of new drugs to treat diseases associated with these pathogens. Although we can culture cyanobacteria in the laboratory, these microorganisms can be found growing as massive colonies off the coast of Barbados.

<u>Selected Literature on Biofuels Research</u>

1) Sheehan, J., Dunahay, T., Benemann, J. & Roessler, P. (1998) A look back at the U.S. Department of Energy's Aquatic Species Program-Biodiesel from Algae. National Renewable Energy Laboratory NREL/TP-580-24190, www.nrel.gov/docs/fy04osti/34796.pdf.

2) Sharma, Y. C. & Singh, B. (2008) Development of biodiesel: Current scenario. *Renewable and Sustainable Energy Reviews* **13**, 1646-1651.

3) Demirbas, A. (2009) Progress and recent trends in biodiesel fuels. *Energy Conversion and Management* **50**, 14-34.

4) Ross, A. B., Jones, J. M., Kubacki, T. & Bridgeman, T. (2008) Classification of macroalgae as fuel and its thermochemical behaviour. *Bioresource Technology* **99**, 6494-6504.

5) Singh, J. & Gu, S., (2010) Commercialization potential of microalgae for biofuel production, *Renewable and Sustainable Energy Reviews* **14**, 2596-2610.

6) Batan, L., Quinn, J., Willson, B. & Bradley, T. (2010) Net energy and greenhouse gas emission of biodiesel derived from microalgae. *Environmental Science & Technology* **44**, 7975-7980.

7) Greenwell, H. C., Laurens, L. M. L., Shields, R. J., Lovitt, R. W. & Flynn, K. J. (2010) Placing microalgae on the biofuels priority list: a review of the technological challenges. *Journal of the Royal Society Interface* **7**, 703-726.

8) Yang, j., Xu, M., Zhang, X., Hu, Q., Sommerfeld, M. & Chen, Y. (2011) Life-cycle analysis on biodiesel production from microalgae: Water footprint and nutrient balance. *Bioresource Technology* **102**, 159-165.

9) Pittman, J. K., Dean, A. P. & Osundeko, O. (2011) The potential of sustainable algal biofuel production using wastewater resources. *Bioresource Technology* **102**, 17-25.

10) Rodolfi, L., Zittelli, G. C., Bassi, N., Padovani, G., Biondi, N., Bonini, G. & Tredici, M. R. (2009) Microalgae for oil: strain selection, induction of lipid synthesis and outdoor mass cultivation in a low-cost photobioreactor. *Biotechnology and Bioengineering* **102**, 100-112.

11) Chen, C.-Y., Yeh, K.-L., Aisyah, R., Lee, D.-J. & Chang, J-S. (2011) Cultivation, photobioreactor design and harvesting of microalgae for biodiesel production: A critical review. *Bioresource Technology* **102**, 71-81.

12) Pruvoat, J., Van Vooren, G., Le Gouic, B., Couzinet-Mossion, A. & Legrand, J. (2011) Systematic investigation of biomass and lipid productivity by microalgae in photobioreactors for biosel application. *Bioresource Technology* **102**, 150-158.

13) Park, J. B. K., Craggs, R. J. & Shilton, A. N. (2011) Wastewater treatment high rate algal ponds for biofuel production. *Bioresource Technology* **102**, 35-42.

14) Mutandaa, T., Ramesha, D., Karthikeyanb. S., Kumaria, S., Anandrajc, A. & Buxa, F. (2011) Bioprospecting for hyper-lipid producing microalgal strains for sustainable biofuel production. *Bioresource Technology* **102**, 57-70.

15) García, V., Päkkilä, J., Ojamo, H., Muuurinen, E. & Keiski, R. L. (2011) Challenges in biobutanol production: how to improve the efficiency. *Renewable and Sustainable Energy Reviews* **15**, 964-980.

16) Zhang, Y.-N., Li, L.-Z., Xian, M., Ma, Y.-J., Yang, J.-M., Xu, X. & He, D.-Z. (2009) Problems with the microbial production of butanol. *Journal of Industrial Microbiology and Biotechnology* **36**, 1127-1138.