

University of South Florida ***Sustainable Algal Biofuel Production***

PI: Sarina J. Ergas **Co-PI:** Qiong Zhang, James R. Mihelcic, John Wolan (deceased)

Students: Angela Chapman, PhD Secondary Ed. in progress; Matthew Gaston, MS Environmental Engineering in progress; Benjamin Gillie, BS Chemical Engineering; Trina Halfhide, PhD Engineering Science in progress; Mehregan Jalalizadeh, MS Environmental Engineering; Ruben Jean, BS Environmental Engineering (UF) in progress; Eunyong Lee, PhD Environmental Engineering in progress; Maria Pinilla, MS Environmental Engineering; John Trimmer, MS Environmental Engineering in progress; Innocent Udom, PhD Chemical Engineering in progress; Sarah Watson, MS Environmental Engineering in progress.

Description: Microalgae are productive at utilizing CO₂ and can generate biomass for production of biodiesel, methane, or other fuels as well as valuable co-products (e.g. animal feeds, polymers). Algal biofuel production can be more profitable and sustainable when combined with wastewater treatment and CO₂ utilization from electric power generation facilities. A number of research gaps exist for full scale algal biofuel production including: 1) improvement of algal growth and nutrient uptake rates, 2) integration of systems with waste gas, wastewater, and water reclamation systems, 3) improved gas transfer and mixing, 4) improved algal harvesting and dewatering and 5) life cycle assessment (LCA) and economic analysis. In addition, little attention has been given to the potential use of algal biofuel systems to treat wastewater and produce heating and cooking fuels in developing countries. The overall objective of this project is to develop an interdisciplinary multi-investigator research program that integrates microalgal biofuel production with wastewater treatment and carbon recycling. An algal biofuels lab will be established at the USF Botanical Gardens, which will house several bench-scale algal photo-bioreactors. Initial experiments will focus on optimizing CO₂ uptake from combustion gases, wastewater nutrient removal and production of algal biomass under varying operating conditions. Both oil rich algal species and algae that grow well on wastewater will be investigated. LCA methods will be used to provide insight into the environmental impacts of the process under varying conditions and enable system evaluation based on both technical performance and life cycle impacts. This project is designed to develop PI expertise and collaborations and train graduate students in a new field of research that is critical in establishing Florida as center of algal biofuels production. Future research directions include: 1) integration of algal biofuel production with domestic, agricultural and industrial wastewater, 2) sustainable aquaculture system development, 3) production of jet fuel from algae cake, 4) application of algal biofuels technology in developing countries, 5) development of integrated LCA-economic assessment tools to assist in algal biofuel system decision making.

Budget: \$50,000

Project Time Period: 2/1/2010-6/30/2012

Universities: USF

External Collaborators: Mote Marine Laboratories

Research Summary:

Algae culturing studies were carried out using bag photo-bioreactors (PBRs) in a greenhouse at the USF Botanical Gardens. The reactors were fed anaerobic digester centrate from the Howard F. Curren Advanced Wastewater Treatment Plant in Tampa Florida and synthetic aquaculture wastewater based on Mote Aquaculture Research Park sturgeon program wastewater. The PBRs were operated over a nine month period under natural light conditions. Gas transfer tests were used to determine CO₂ uptake rates

at varying influent CO₂ concentrations (2% and 5%). The results show that wild algae adapted to wastewater nutrients are able to have high productivity, CO₂ and nutrient uptake rates despite high ammonia levels. Life Cycle Assessment (LCA) results showed that the pathway with algae biomass (feedstock), anaerobic digestion and combined heat and power (conversion process), and electricity and heat (energy products) has lower environmental impacts if wastewater and flue gas is used in algae cultivation. Algae harvested from the PBRs were used in algae coagulation tests. Coagulants tested included: alum, ferric chloride, cationic and non-ionic polymers, and natural materials (moringa seeds and cactus mucilage). Results showed that high algae harvesting efficiencies could be obtained with alum (91%), ferric chloride (93%), and cationic polymer (98%). Cationic polymer also resulted in the lowest cost alternative. LCA results; however, showed that the highest energy use and greenhouse gas (GHG) emissions were obtained for the cationic polymer, while ferric chloride resulted in the lowest energy use and GHG emissions. LCA results also showed that the pathway that converts algae biomass through anaerobic digestion and combined heat and power (CHP) process to electricity and heat had a higher overall environmental impact than the pathway that convert algae biomass to liquid biofuels through thermochemical gasification and Fischer-Tropsch Synthesis (FTS) process.

Student research:

PhD: Angela Chapman (Secondary Ed.), Trina Halfhide (Engr. Science), Eunyoung Lee (Environ. Engr.), Innocent Udom (Chemical Engr.); Master's (all from Environ. Engr.): Maria Pinilla John Trimmer, Mehregan Jalalizadeh, Matthew Gaston, Sarah Watson; Undergraduates: Benjamin Gillie (Chemical Engr.), Ruben Jean (Environ. Engr. UF).

Outreach:

The project team participated in Earth Day Events at the USF Botanical Gardens in 2011 and 2012. We are currently working with Middleton Magnet School for Science and Technology, an economically challenged high school in East Tampa, on a project that looks at the effect of participation in authentic science research on students understanding of science.

Grant proposals:

Three proposals were funded that tied to this seed funding: PIRE: Context Sensitive Implementation of Synergistic Water-Energy Systems, PI: J. Mihelcic, Faculty Participants: S.J. Ergas and Q. Zhang, NSF, \$3,912,276, 5 years; REU Site: Tampa Environmental Interdisciplinary Research, co-PI: S.J. Ergas, NSF, \$392,000, 3 yrs; Advanced Biological Waste-to-Energy Technologies, co-PI S.J. Ergas, European Commission, student stipends and lab fees, 4 yrs. Two doctoral students obtained fellowships (NASA and Fulbright) based on their research related to this project.

Partnerships:

A. F. Hepp, NASA Glenn Res. Center; J. Bartacek, Asst. Prof., Water Technol. & Environ. Engr., ICT Prague; C. Butler, Asst. Prof., Civil & Environ. Engr. UMass; P. Jenicek Prof. & Head, Water Technol. & Environ. Engr., ICT Prague; P. Lens Prof., Environ. Biotechnol., UNESCO-IHE, Delft; J. Love, Sen. Lec., Plant Mol. Bio., U. Exeter; G. Philippidis Assoc. Prof., Chem. Engr., USF; A.C. Wilkie, Assoc. Prof., Soil & Water Sci. UF-IFAS.

Funded Research Grants and Fellowships:

PIRE: Context Sensitive Implementation of Synergistic Water-Energy Systems, PI: J. Mihelcic, co-PIs: M. Trotz, E.C. Wells, C. McKayle (UVI), **Senior Personnel: S.J. Ergas, Q. Zhang, D. Yeh, L. Whiteford, R. Zarger, F. Muller-Karger, Y. Goswami, D. Durham, A. Feldman, B. Batson, K. Alexandridis, M. Brandt (UVI), National Science Foundation, \$3,912,276, 5 yr.**

REU Site: Tampa Environmental Interdisciplinary Research, PI: M. Trotz, co-PI: S. Ergas, National Science Foundation, \$392,816, 3 yr.

BioWET—Advanced Biological Waste-to-Energy Technologies, European Commission, PI: D. Yeh, co-PI: S. J. Ergas, European PIs: J. Bartacek (ICT –Prague), P. Lens (UNESCO IHE), \$330,000 to EU Partners, Student stipends and laboratory bench fees to USF.

Green Aviation Fuels from Microalgae, NASA-Harriett G. Jenkins Pre-doctoral Fellowship Project (JFPF) award, \$121,500 for support of Innocent Udom, Collaboration with NASA Glenn Research Center.

Examining the Use of Algal Photobioreactor Production Systems for the Dual Purpose of Bioremediation and Biofuel Production under Different Climatic Conditions, Fulbright Fellowship awarded to Trina Halfhide (PhD candidate USF) for 1-year study at the Life Sciences University, Oslo Norway.

Not Funded:

EFRI-PSBR: Sustainable Microalgae Bioenergy Systems, PI: S. Ergas, co-PIs: Q. Zhang, B. Joseph, A.C. Wilkie (UF), G. Phillipidis (USF Polytech), National Science Foundation, \$1,999,361, 4 yr.

Sustainable Microalgal Bioenergy System, PI: Q. Zhang, Co-PIs: S. Ergas, J. Mihelcic, National Science Foundation, \$3299,968, 3 yr.

New Partnerships:

Aloysius F. Hepp is a Ph.D. Senior Chemist in the Bio Science and Technology Branch Space Processes and Experiments Division of NASA John H. Glenn Research Center in Ohio. Dr. Hepp co-supervises ongoing research being conducted by Innocent Udom as part of the NASA-Harriett G. Jenkins Pre-doctoral Fellowship program. Drs. Ergas and Goswami traveled to NASA Glenn Research Center in December, 2011 to meet with Dr. Hepp and discuss our collaboration.

Middleton Magnet School for Science and Technology is located in East Tampa and has more than 1,300 students (~70% African American, 12% Hispanic, 78% receive free or reduced price lunches). We are currently collaborating with agricultural biotechnology and marine science teachers at Middleton. Students construct simple algal photo-bioreactors using soda bottles and aquarium pumps. The reactors are inoculated with algal consortia from our laboratory and set up in greenhouses at Middleton. Experiments are designed to investigate algal growth rates under varying conditions. Experimental data are used to teach science and engineering principles, statistics, and use of data analysis tools.

Jan Bartacek is an Assistant Professor in the Dept. of Water Technology and Environmental Engineering at the Institute of Chemical Technology (ICT) Prague. Dr. Bartacek is the PI of the BioWET grant and participated in the BioWET summer school at USF.

Caitlyn Butler is an Assistant Professor in the Dept. of Civil and Environmental Engineering at the University of Massachusetts, Amherst. Dr. Butler's research focuses on energy and resource recovery in wastewater treatment, emphasizing the use of bioelectrochemical systems. Dr. Butler participated in the BioWET summer school at USF.

Pavel Jenicek is a Professor and Head of the Department of Water Technology and Environmental Engineering at ICT Prague. He is active in the research of anaerobic wastewater treatment, biogas treatment, anaerobic digestion of sludge, minimisation of sludge production, biological nutrient removal and recovery. Dr. Jenicek is a co-PI of the BioWET grant and participated in the BioWET summer school at USF.

Piet Lens is Professor of Environmental Biotechnology at the Pollution Prevention and Control core of the Department of Environmental Resources of UNESCO-IHE Water Research and Education Institute in Delft, the Netherlands. Dr. Lens is a co-PI of the BioWET grant and participated in the BioWET summer

school at USF. Dr. Ergas spent two weeks at UNESCO-IHE during the summer of 2012 as a visiting scholar.

Dr John Love, Senior Lecturer in Plant Molecular Biology at the University of Exeter. Dr. Love's research group has a productive collaboration with Shell Global Solutions, investigating the molecular and cell biology of hydrocarbon production in algae.

George Philippidis is Associate Professor of Chemical Engineering at USF. He has over 20 years of experience in leading strategic business units in advanced biofuels and renewable energy. Dr. Phillipidis was a participant in the BioWET summer school and a co-PI on the EFRI proposal.

Peter van der Stein is a lecturer in the Pollution Prevention and Control core of the Department of Environmental Resources of UNESCO-IHE Water Research and Education Institute in Delft, the Netherlands. Dr. Van der Stein is a co-PI on the BioWET grant.

Ann C. Wilkie is an Associate Professor of Bioenergy and Sustainable Technology in the Soil and Water Science Department at the University of Florida-Institute of Food and Agricultural Sciences (UF-IFAS). Her specialty is environmental microbiology, with particular emphasis on anaerobic technology and algal biofuels. Dr. Wilkie was a participant in the BioWET summer school at USF and a co-PI on the EFRI proposal. She is currently collaborating with Dr. Ergas on research related to bioprospecting wild algae species.

Annual Progress Report

Algae Culturing Studies:

Algal growth experiments were conducted under natural illumination in a temperature controlled (25-32°C) greenhouse at the University of South Florida Botanical Gardens in Tampa Florida. Three vertical hanging tubular plastic bag photobioreactors (PBRs) were obtained from the Norwegian Life Sciences University in Oslo. Two PBR cells were fed an anaerobic digester centrate feed and the third cell was operated with a 50:50 mixture of centrate and synthetic aquaculture wastewater. Centrate was collected from the Howard F. Curren Advanced Wastewater Treatment Facility (HFCAWTF) in Tampa, FL. The aquaculture-centrate mixture (ACM) was based on research by Watson et al., (2011) showing that incorporation of algae and anaerobic digestion could reduce the nutrient impacts of land based recirculating aquaculture systems. Wild algae harvested from the surface of a secondary clarifier at the HFCAWTF were used to inoculate the reactors. The reactors were maintained at a mean cell residence time of 7-days for approximately nine months. A summary of the biomass production and nutrient removal data from the algal PBR studies is given in Table 1. More detailed results have been presented elsewhere (Dalrymple et al., 2011). The low productivity observed in this study may have been due to the low natural ambient light intensity (2.3-9.4 mol/m²/day) during the wet season in Southwest Florida compared with the light intensity used by other authors. Average algal productivity was higher for the 100% centrate reactor than for the ACM reactor, most likely due to the algae being adapted to centrate during a long acclimatization phase (March-October, 2011), during which the reactor was only fed centrate. Both reactors achieved good overall removal efficiencies for nitrogen and phosphorous.

Table 1: Mean and maximum algae growth and nutrient removal data from the centrate and ACM PBRs. Standard deviations shown in parentheses.

Parameter	Centrate		ACM	
	Mean	Max	Mean	Max
Productivity (g/m ² /day)	2.3 (0.77)	3.6	1.7 (0.89)	4.0
Biomass density (g/L as TSS)	664.4 (222)	1030	478.2 (259)	1,140
TN removal efficiency (%)	65	92	64	91
Effluent TN (mg/L)	76.2 (71.1)	180	60.3 (60.5)	130
NH ₄ ⁺ removal efficiency (%)	95	77	86	100
Effluent NH ₄ ⁺ -N (mg/L)	50 (38.0)	92	5.0 (8.6)	15
TP removal efficiency (%)	72	79	73	93
Effluent TP (mg/L)	12.5 (23.6)	54.6	20.1 (34.2)	71.3

Algal growth kinetics:

Current research is focused on characterizing wild algae in batch photoreactors to provide data for modeling efforts. Algae are cultured in batch systems using anaerobic digester feed under artificial light. Environmental conditions are systematically altered to examine the effects of the following on algal growth rates, lipid production, and nutrient removal: 1) nutrient concentration, 2) CO₂ partial pressure, 3) mixing and 4) light intensity. Kinetic parameters are determined by fitting the experimental data to the Monod-type kinetic equations.

Life Cycle Assessment:

A preliminary LCA was conducted using our algae cultivation and digestion data to evaluate the environmental impacts associated with the energy pathways shown in Figure 1. The results showed that the pathway with algae biomass (feedstock), anaerobic digestion and combined heat and power (conversion process), and electricity and heat (energy products) has lower environmental impacts if wastewater and flue gas is used in algae cultivation, as shown in Figure 2.

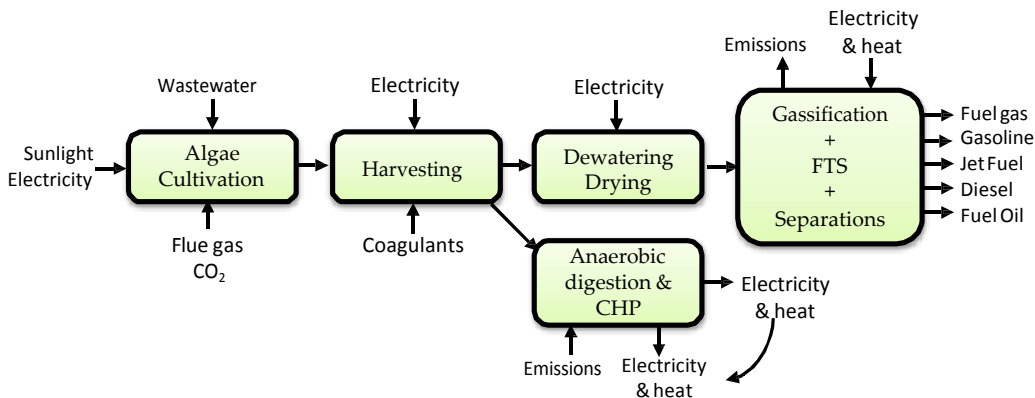


Fig. 1. Schematic of algae bioenergy pathways evaluated.

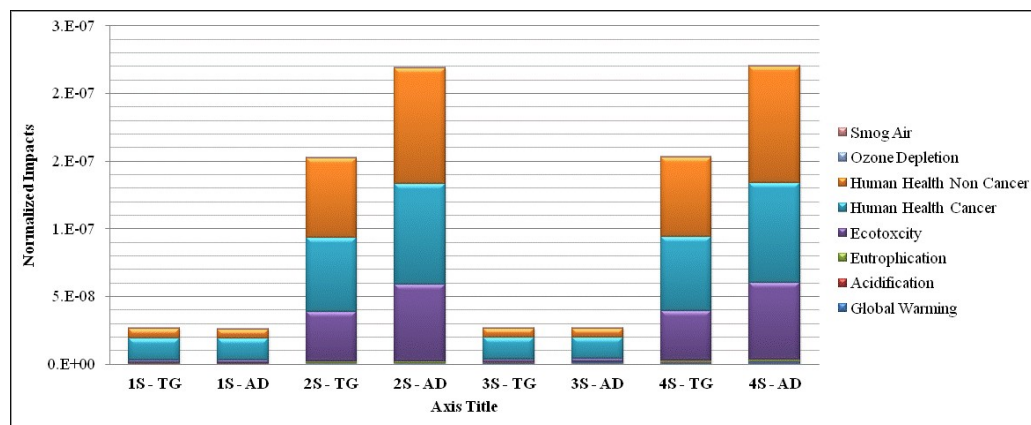


Fig. 2. Overall results from the comparative algae biomass conversion LCA. (TG: thermochemical gasification; AD: anaerobic digestion; 1S: Wastewater is used as a source of nutrients and flue gas is used as a source of CO₂, assuming the process is co-located with a power plant; 2S: Fertilizers are used as a source of nutrients, and Flue Gas is used as a source of CO₂; 3S: Wastewater is used as a source of nutrients, and chemical CO₂ is used as the carbon source; 4S: Fertilizers are used as a source of nutrients, and chemical CO₂ is used as the carbon source.).

Harvesting studies:

Jar tests were conducted using reagent grade (Fisher, Pittsburgh, PA) alum (Al₂(SO₄)₃·12H₂O) and ferric chloride (FeCl₃·6H₂O), Zetag (BASF Chemical Company, Suffolk, VA) 8800 series cationic polymers (8814, 8816, 8818, and 8819), two anionic polymers (Magnafloc E-38 and E-34; BASF Chemical Company, Suffolk, VA) and two natural flocculants (*Moringa Oleifera* and *Opuntia ficus-indica* cactus). Coagulation tests were conducted on suspensions collected from the algal PBRs using a Phipps & Bird Jar Test Apparatus (Richmond, Virginia). An economic and LCA study was conducted to assess the cost and energy and GHG emissions associated with algae harvesting. Costs of coagulants were obtained from bulk vendors of industrial chemicals. The functional unit was production of one metric ton (MT) of dried algae.

Cumulative Energy Demand Analysis (CEDA) in Simapro 7.2 software was used to estimate the life cycle energy (direct energy use as well as the upstream energy consumption) required to produce the amount of alum or ferric chloride needed to harvest one MT of dry algae. The Tool for the Reduction and Assessment of Chemical and Other Environmental Impacts (TRACI) developed by U.S. Environmental Protection Agency (EPA) was used to estimate the GHG emission resulting from alum or ferric chloride production.

An overall summary of the jar test results for all coagulants tested is shown in Table 2. Among the metal salts tested, FeCl₃ had a lower optimal dose (122 mg/L) and slightly higher recovery (93%) than alum. When using FeCl₃, the color of culture changed from green to brown-yellow. This could have a negative impact on the final product if the algae are to be used for pigments. An initial screening study was conducted with the Zetag 8000 series cationic polymers (8814, 8816, 8819, 8846, 8848) to determine the best cationic polymer for further study (data not shown). Although excellent algae recovery (92-98%) was observed at low polymer concentration (34-41 mg/L) for all of the cationic polymers tested, Zetag 8819 was selected for further study because it provided the highest harvesting efficiency (98%) at the lowest optimal dose (34 mg/L). Zetag 8819 also had the overall best performance, in terms of coagulant dose required and algae recovery, of all of the coagulants tested. Poor performance was observed with the non-ionic and anionic polymers and the natural coagulants.

Table 2: Summary of harvesting experiments, showing optimal dose for each coagulant

Coagulant	Optimal Dose (mg/L)	Supernatant Turbidity (NTU)	Supernatant TSS (mg/L)	Solids Recovered (%)
Ferric Chloride	122	7.65	15	93
Alum	140	5.40	30	91
Zetag 8819	34	6.05	20	98
Magnafloc E-38	NA	760	>500	~0
<i>Moringa oleifera</i>	4,670	20.0	25	85
<i>Opuntia ficus-indica</i> cactus	NA	740	>500	~0

Economic and LCA were carried out to compare costs, energy use and GHG emissions associated with each coagulant (Figure 3). Ferric chloride was the most expensive option (\$130/MT of algae), followed by alum (\$65/MT of algae) and cationic polymer (\$50/MT of algae). However, it can be seen that using cationic polymer (Zetag 8819) for harvesting will result in the highest GHG emissions, with 92 kg CO₂ eq/MT Algae, followed by alum and ferric chloride (51 kg CO₂ eq/MT algae and 13 kg CO₂ eq/MT algae, respectively). The energy consumption results are in line with the GHG emissions, showing that polymer has the highest energy consumption (1983.9 MJ/MT of algae), while ferric chloride has the lowest (204 MJ/MT of algae) among three coagulants evaluated.

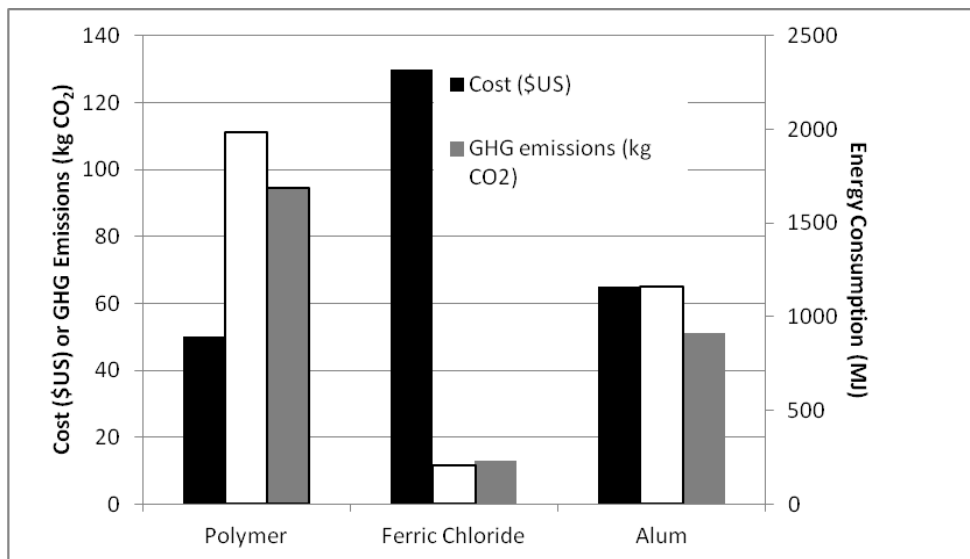


Figure 3: Cost (\$ US), GHG emissions and energy consumption for cationic polymer, ferric chloride and alum.

Gas Transfer Tests:

Initial tests were conducted in one of the bag reactor cells to determine CO₂ gas mass transfer coefficients at varying gas feed flow rates. The concentration of dissolved CO₂ was measured using an Oxyguard CO₂ analyzer (Model # G02C2P, Birkerød, Denmark) and the pH of the reactor was measured using a Teledyne Isco 701 pH/Temperature Module on a 6712 portable Sampler (Lincoln, Nebraska). CO₂ utilization rates in the photo-bioreactors with respect to time are shown in Figure 4. The utilization rates

ranged from 5.56×10^{-3} to 1.5×10^{-2} $\text{mols m}^{-3} \text{min}^{-1}$ for the reactor with a 2% CO_2 gas feed. The results were in agreement with the growth rate data.

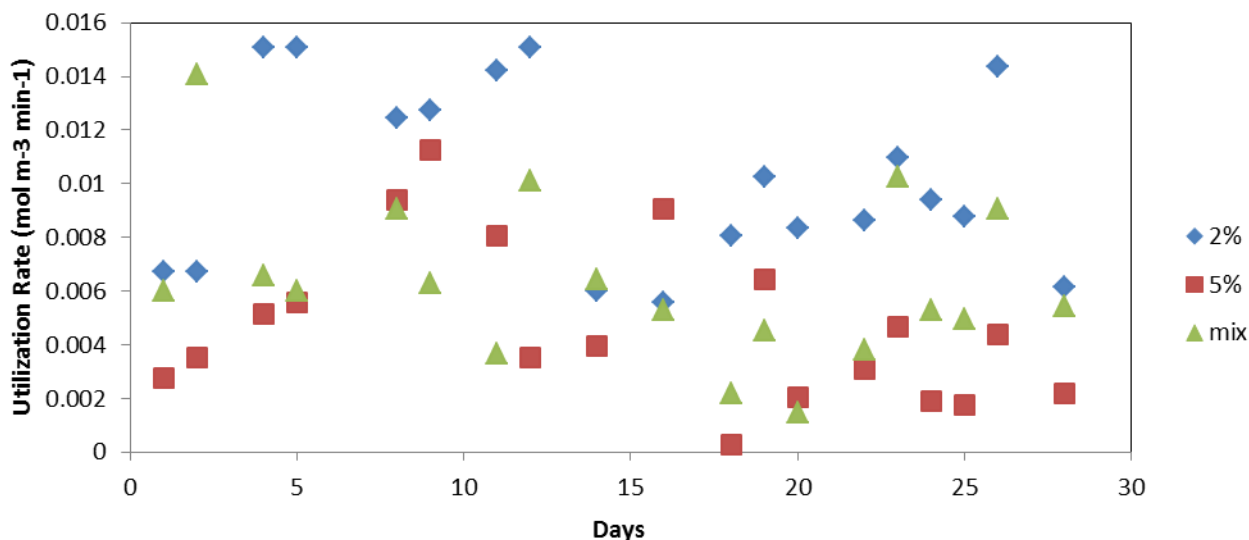


Figure 4: CO_2 Utilization Rates in each reactor vs. time.

Peer Reviewed Journal Publications:

1. Yuan, X., Wang, M., Park, C., Sahu, A.K., Ergas, S.J. (2012) Microalgae Growth Using High Strength Wastewater Followed by Anaerobic Co-digestion, *Water Environ. Research*, 84(5):396-404.
2. Yuan, X., Kumar, A., Sahu, A.K., Ergas, S.J. (2011) Impact of Ammonia Concentration on *Spirulina platensis* Growth in an Airlift Photobioreactor, *Bioresource Technology*, 102(3): 3234-3239.
3. Kumar, A., Yuan, X., Sahu, A.K., Zhang, Q., Ergas, S.J., Malcata, F. X., Van Langenhove, H. (2010) Strategies for CO_2 sequestration using microalgae and cyanobacteria: Recent developments and future directions, *Trends in Biotechnology*, 28(7): 371-380.
4. Dalrymple, O.K., Halfhide, T., Udom, I., Gilles, B., Wolan, J., Zhang, Q., Ergas, S.J. (accepted) A preliminary estimation of the algal feedstock production potential of Tampa Bay utilizing CO_2 emissions and wastewater effluent, *J. Aquatic Biosystems*.

Conference Presentations and Posters:

1. Udom, I., Halfhide, T., Gillie, B., Dalrymple, O., Zaribaf, B. H., Zhang, Q., Ergas, S.J. (2012) Harvesting algae grown on wastewater, *Proc. 85th Annual Meeting of the Water Environment Federation (WEFTEC 12)*, Sept. 29-Oct. 3., New Orleans, LA.
2. Dalrymple, O.K., Halfhide, T., Udom, I., Gilles, B., Wolan, J., Zhang, Q., Ergas, S.J. (2011) A preliminary estimation of the algal feedstock production potential of Tampa Bay utilizing CO_2 emissions and wastewater effluent, *Proc. Florida Energy Systems Consortium (FESC) Summit*, Sept. 26-27, 2011, Gainesville, FL.
3. Halfhide, T., Trimmer, J., Pinilla, M., Bosshart, W., Zhang, Q., Wolan, J., Main, K., Ergas, S. J. (2011) Reducing Carbon and Nutrient Impacts of Aquaculture Using an Algal Photo-bioreactor Production System, *Proc. International Water Association Leading Edge Technologies Conference*, June 6-10, Amsterdam, The Netherlands.
4. Watson, S., Halfhide, T., Trimmer, J., Zhang, Q., Wolan, J., Main, K., Ergas, S.J. (2011) Reducing the Nutrient Impacts of Aquaculture Through the Use of an Algal Photobioreactor Production System, *Proc. 2011 WEF Nutrient Recovery and Management Conference*, Jan. 9-12, 2011, Miami, FL.