Enhanced waste to fuel conversion with a bioelectrochemically controlled autotrophic bioreactor

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1. Introduction:

Photobioreactors (PBRs) are increasingly being used to grow algae for production of biofuel and other products. Light and CO_2 are two major limiting resources that can restrict algal growth in PBRs. Atmospheric CO_2 is only 0.04% and to overcome this limitation researchers have started to couple biogas released from the anaerobic digestion (AD) of organic waste with PBRs since biogas consists of mostly CO_2 and CH_4. The CO_2 component of biogas is normally considered a nuisance and its abiotic removal can be expensive. Unfortunately, the conventional mesophilic AD is a slow process. Recently, bioelectrochemical systems (BES) have been described that accelerate the anaerobic breakdown of organic waste into biogas (Booth, 2009, Cheng et al., 2009). BESs accomplish this via anaerobic cultures of red pigmented, exoeletrogenic *Geobacter* species grown on the anode with poorly understood electroactive methanogens that grow in association with biocathodes (Cheng et al, 2009, Pisciotta et al, 2012, Zaybak et al., 2013).

2. Aim of Investigation:

In this report, BlueSens CO_2, CH_4 and O_2 Gas Sensors were integrated into a new type of BES-fed PBR built to test if bioelectrochemically produced biogas can be used to grow algae. If successful, this could provide a method for more rapidly treating organic waste streams while biologically processing bioelectrochemically-generated biogas via potentially enhanced growth of photoautotrophs. This gas sensor integrated arrangement should provide real time characterization of the overall metabolic activity inside a hybrid BES-PBR linked system. Two PBR systems were built and tested head-to-head to study *Chlorella vulgaris* growth in BG-11 media on standard AD derived biogas versus BES derived biogas (**Fig 1**).

3. Materials and Methods

3.1 Photobioreactors:

Two rectangular 18.9 L glass aquaria were converted into biogas-fed, closed-loop photobioreactors. Each aquarium was sealed with a custom cut sheet of glass epoxyed into place over the open top of each tank. Next, 0.5 cm holes were drilled into both ends of the glass covers using diamond tipped drill bits. Gas incurrent and out-current silicone tubing lines were passed through these holes and sealed to prevent gas leaks. The input gas line was connected to an airstone affixed to the bottom of each tank. This provided for even gas dispersion and bubble mediated mixing of the media. Gas bubbling up into the headspace then exited PBR via the out-current gas line connected to an air pump sealed inside a hollow plastic reservoir. The pump set

up provides for constant gas flow through the closed loop PBR systems. Prior to reentering the PBRs gas was pumped through in-line O_2, CO_2 and CH_4 sensors via gas line tubing. For each PBR, a biogas delivery line tied into the circulating gas just upstream of the pump and was fitted with a one way valve. The one way valves prevent oxygen from entering the BES or AD chambers and inhibiting anaerobes therein. Log grown *C. vulgaris* was inoculated into 5 L of illuminated (12hr light/12hr dark) BG-11 per PBR at an initial density of OD 0.025 (**Fig 2**).

3.2. Anaerobic Digesters and Bioelectrochemical Cells.

Triplicate gas-linked anaerobic digesters were constructed from 100 ml serum vials containing 75 mls of synthetic wastewater; *ie* 50 mM Phosphate Buffered Saline (PBS) amended to contain 1 g/l sodium acetate plus Wolfe's vitamins and minerals. All vials were degassed with nitrogen and crimp sealed prior to injection of 0.5 mls of starter culture from a 5 week old, acetate fed, biogas producing digester. Headspace of the three anaerobic digesters was connected by needles passing through the rubbers stoppers and connected by tubing. Passive pressurization of biogas formation forced the biogas through a one way valve just prior to biogas entry into the recirculated PBR gas stream. Triplicate BESs were similarly produced by inserting two 10 cm graphite electrodes into each BES vial and a potential of 0.7 V was applied using a Biologic MPG-2 that also monitored current over time; an indicator of bioelectrochemical activity and acetate consumption. Cell density was periodically measured at OD 600 nm via spectrophotometer.

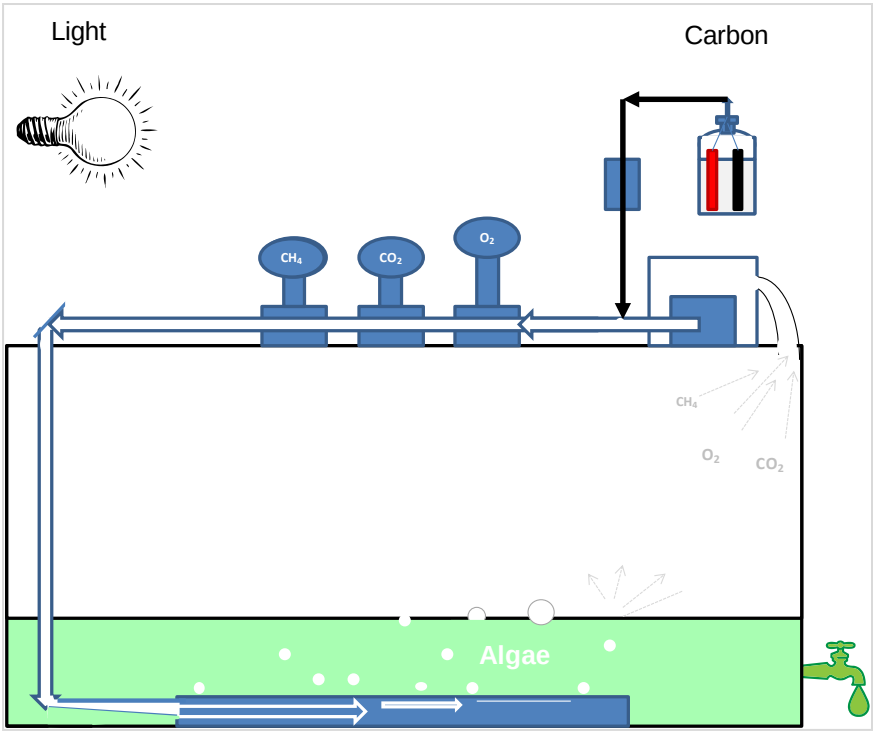

Fig 1. Simplified schematic overview of BES biogas fed PBR fitted with in-line BlueSens O_2, CO_2 and CH_4 gas sensors.

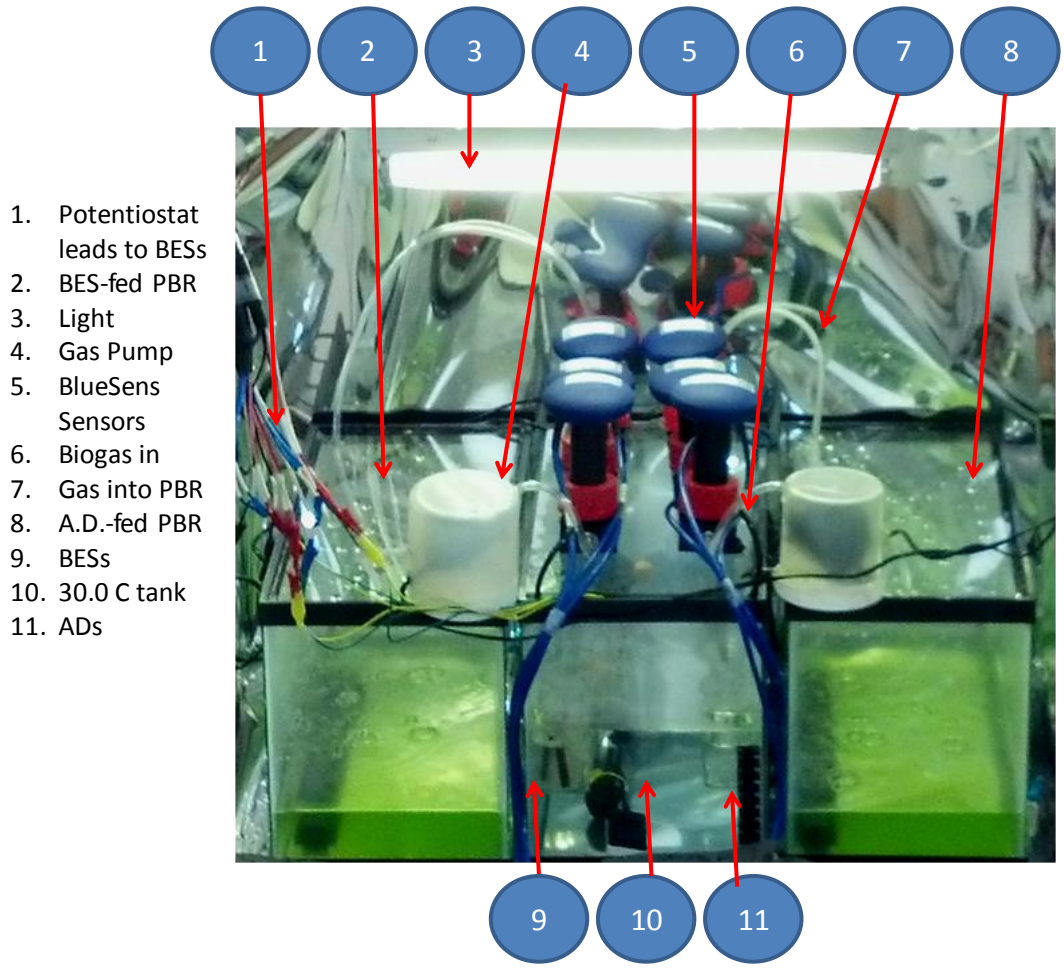

Fig 2. Side-view photograph detailing Chlorella containing PBRs fed by either BES (left) or AD biogas (right). Components of the system including pumps and sensor indicated by numbered arrows on side bar legend.

4. Results and Conclusions

In-line BlueSens CH_4 Sensors revealed a considerably more rapid accumulation of methane in the BES-PBR headspace compared to the conventional control AD reactor (**Fig 3A**). Potentiostat measured current peaked within four days for all three BESs indicating rapid catabolism of acetate (**Fig 3B**). The optical cell density of both *C. vulgaris* cultures increased from the initial inoculum baseline level (0.025) during the course of the incubation indicating algal growth in BG-11 media in both PBRs (**Fig 4A**). The control AD linked PBR increased more rapidly but the BES-PBR system reached a higher final cell density. Oxygen sensor also indicated higher algal photosynthesis in the late stage BES-PBR (**Fig 4B**).

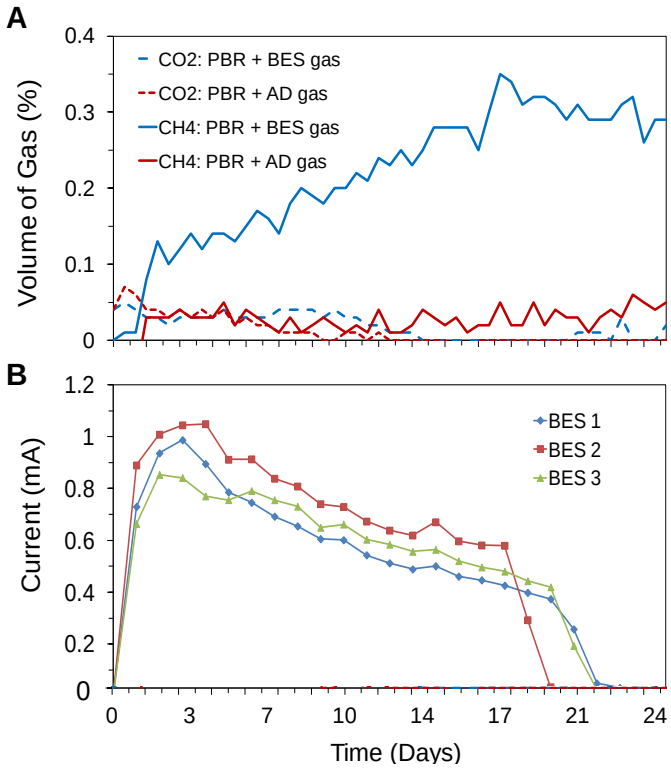

Fig 3. Methane accumulation and CO_2 consumption in PBRs (A). Current generation in triplicate BESs attached to the BES-PBR (B).

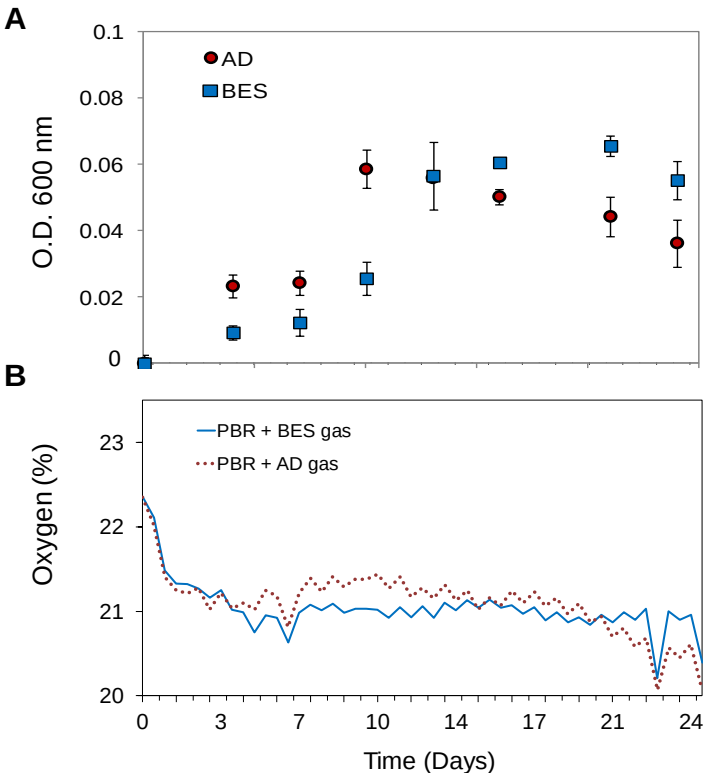

Fig 4. *C. vulgaris* optical cell density in PBRs (A) and sustained O_2 detected in PBRs (B).

For both reactors CO_2 remained near baseline levels of detection; likely a function of the relatively high headspace volume and photosynthetic demand of the cultures. Increasing the volume of the BESs and ADs relative to the PBR culture volume could further improve productivity. These results suggest integration of BlueSens Sensors with photobioreactors can enable optimized biofuel production from renewable resources like sunlight and bioelectochemically treated organic waste.

References:

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Acknowledgments:

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