Optimising *Isochrysis* sp. carbon fixation: a step towards the greening of fossil fuel

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Abstract

As concerns over global warming are increasing, a great deal of research on carbon capture has been carried out. Among various CO_2 utilization technologies, biological methods – particularly ones using microalgae photosynthesis – have several advantages. This natural means of capturing emitted CO_2 through photosynthetic microalgae, while producing some value-added by-products, can be regarded as economically viable and more sustainable. In this study, marine microalgae *Isochrysis* sp. was cultured using a 2x10-L customized bubble column photobioreactor skid to determine the optimum operating condition (pH, temperature, gas flow rate and luminance) towards highest carbon fixation rate. Twenty-one experimental runs were carried out in a lab-scaled photobioreactor supplied with simulated flue gas, containing O_2, SO_2, NO_2, CO_2 and CO. Illuminance is available through units of Philip's T5 type fluorescent bulb with timer-control. Automatic data logging was made possible through Dewetron's DEWE 43 data acquisition system. A carbon fixation ability calculation was made by comparing the gained dry weight between first and last days, taking into account the approximate biomass molecular weight in the photosynthesis reaction. Design of the experiment and optimization analysis was carried out using Design Expert Software 8.0. Results indicate that experimental simulations with temperatures of 300°C, pH 7.5, luminance of 1500 lux and a 0.15 liter/min gas flow rate give the highest fixation rate of 3.68 g CO_2/day. Thus, this finding shall be useful in utilizing the capabilities of *Isochrysis* sp. as the biological carbon fixer in neutralizing carbon emissions from power plants.

Keywords: CO_2 emission, carbon capture, flue gas, microalgae, optimization.

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1 Introduction

Coal, being an abundant and cheap fuel source, coupled with its proven and matured technology remains a dominant energy source to be utilized. Pertinent emission statistics, as published by the International Energy Agency (IEA) [6], states that coal accounts for 42% of the global CO_2 emissions, due to its high carbon density. Owing to growing energy demand from developing countries, coal supply will grow from 3,184 million tonnes of oil equivalent (Mtoe) in 2007 to 4,887 Mtoe in 2030, and estimated CO_2 emissions from fossil fuel combustions at this time is 40.2 Gt.

In a gesture to curb this CO_2 emission, development on Carbon Capture and Storage (CCS) technologies are actively implemented globally. These CCS initiatives include separating, capturing, delivering and storing, or its combination thereof, of emitted CO_2 from utilities or industries. The works can be categorized into three major approaches – pre-combustion, during combustion and post combustion, denoting where the initiatives are taken place in a typical power generation process. Some common technologies include gasification, oxy-fuel combustion and amine-based absorption, for pre-combustion, during combustion and post-combustion stages respectively [7]. To date, the total number of projects operating or executed are 17 and their total capture capacity are more than 37 million tonnes of CO_2 per annum (Mtpa) [3]. There are still much to be achieved however, for these technologies to be satisfactorily mature and well adopted, as issues like material degradation, loss of efficiency, larger plant footprint, high capital cost and high auxiliary power are still pressing issues [7].

An alternative means in post-combustion CCS, which receives much attention lately is through biological fixation, by the virtue of natural photosynthesis process by autotrophic microalgae. Principally, the process fixes CO_2 into O_2 and organic matter in the form of C_x-H_y-O_z chain. This organic matter provides multitudes of potential value-added downstream products, which, with additional processes, can be converted into biomass, biofuel, nutritional diets, aquaculture food and fertilizers, to name a few [9, 10]. Thus, this biological mitigation technique can be regarded to be economically viable and environmentally sustainable in the long term. With the variety of potential valuable products it can raise, this method can function as a catalyst towards the widespread implementation of industrial symbiosis, where several industries co-exist and interdependent with each other. Where *"the consumption of energy and materials is optimized and the effluents of one process serve as the raw material for another process"* [1].

However, vital challenges do lie in the path. Of utmost concern is the method's ability to significantly reduce CO_2 emissions from a coal-fired power plant. A modern-day coal-fired power plant is estimated to emit between 6–8Mt CO_2 per year [16]. If we compare this with some species' best available carbon fixation rate, the requirements for land area needed and capitals for a fully functional photobioreactor farm, are still leading to a staggering figure, even though to fix 1% of annual CO_2. Yet, this comparison is not a fair one since this biological method does not store permanently whatever amount of CO_2 captured, like what typical CCS technologies do. Rather, this algae recycles the fixed CO_2 molecules after it has been decayed or re-utilized from downstream products they are turned into. Carbon recycling is exactly how mother earth manages CO_2 emission by design and it works just well until the balance is disturbed by human activities [12, 15].

It would be a noble effort to reinstate such a balance, simply through wider acceptance and utilization of microalgae based carbon fixation. One basic step, as recommended in [13] is to increase the yield (growth rate) of the species used through process optimization.

2 Material and methods

2.1 Microalgae species

Isochrysis sp. marine microalgae species was selected for this study. This microalgae species was isolated from the seawater surrounding TNB Janamanjung power station, Manjung, Perak, Malaysia.

2.2 Photobioreactor

A customized laboratory scale photobioreactor (PBR) was used in the experiment. The PBR is of bubbling column type, having double cylindrical column of 10-L each as the reactor vessel. The material of the reactor column is of polycarbonate having dimension 3 mm thick, 200 mm diameter and 800 mm high. Polycarbonate was chosen as the material due to its good impact resistance and optical properties having at least 92% transparency [2]. The PBR has the capability to vary and maintain temperature from -10°C to 100°C using a chiller/heater. Other instrumentations equipped are pH sensor, type K thermocouple and dissolved O_2 sensor. Illuminance is available through five units of T5 type fluorescent bulb with timer-controlled. Automatic data logging of temperature, pH and dissolved O_2 was made possible through a data acquisition system. Photograph of the PBR is as shown in Figure 1.

2.3 Culturing conditions

The culturing was done using the f/2 media mixed with artificial sea water. The microalgae were cultured in 10-L PBR, where the carbon fixation experiment conducted. The operating conditions (temperature, pH, gas flow rate and luminance) were varies depending on optimization runs which been suggested by the Design Expert Software. The initial optical density for the species was maintained at 0.20 abs for each experimental run.

Figure 1: A 2x10L photobioreactor used in the experiment.

2.4 Design of experiment

A statistical optimization study was done for photobioreactor operating parameters to obtain maximum carbon fixation rate of *Isochrysis* sp. Four photobioreactor operating parameters involved include temperature, pH, gas flow rate and luminance. Table 1 indicates the experimental range of independent variables studied to optimize the maximum carbon fixation rate.

Table 1: Experimental range of independent process variables.

Independent variables	Levels of independent variables				
	-2	-2	0	+1	+2
Temperature (ºC)	20	25	30	35	40
pH	4	5	6	7	8
Gas flow rate (L/m)	0.05	0.10	0.15	0.20	0.25
Luminance (lux)	500	1000	1500	2000	2500

Central Composite Design (CCD) by Response Surface Methodology (RSM) using Design Expert 8.0 (Stat Ease Inc. Minneapolis) was used to determine the maximum carbon fixation rate of *Isochrysis* sp. Table 2 indicates the experimental design obtained from the Design Expert Software.

Each run will be conducted up to 10 days using simulated flue gas containing 4% CO_2, 3% O_2, 105 ppm CO, 272 ppm NO_2, 121 ppm SO_2 and N_2 (balance). The gas in use is a replication of a coal-fired power plant's flue gas. 30 mL samples were withdrawn daily for growth rate and carbon fixation measurements.

Table 2: Experimental runs designed by the software.

Run	Factor 1 Temperature (°C)	Factor 2 pH	Factor 3 Gas flow rate (L/m)	Factor 4 Luminance (lux)	RESPONSE CO_2 fixation rate (gCO_2/day)
1	30.00	6.00	0.15	2500.00	
2	30.00	6.00	0.25	1500.00	
3	30.00	6.00	0.15	1500.00	
4	35.00	5.00	0.10	2000.00	
5	20.00	6.00	0.15	1500.00	
6	30.00	6.00	0.15	1500.00	
7	35.00	7.00	0.10	1000.00	
8	35.00	7.00	0.20	1000.00	
9	30.00	8.00	0.15	1500.00	
10	25.00	7.00	0.20	2000.00	
11	30.00	6.00	0.15	1500.00	
12	30.00	4.00	0.15	1500.00	
13	30.00	6.00	0.15	1500.00	
14	30.00	6.00	0.15	500.00	
15	35.00	5.00	0.20	2000.00	
16	25.00	5.00	0.10	1000.00	
17	30.00	6.00	0.15	1500.00	
18	40.00	6.00	0.15	1500.00	
19	25.00	7.00	0.10	2000.00	
20	25.00	5.00	0.20	1000.00	
21	30.00	6.00	0.05	1500.00	

2.5 Carbon fixation experiment

The carbon fixation calculation was adopted by taking into considerations of the ratio between the moles of CO_2 and the moles of typical molecular formula of biomass [5, 20]. Thus by taking into considerations of a balanced photosynthesis formula as in Eqn (1) below:

$$4CO_2 + \textit{nutrient} + H_2O + \textit{light} \rightarrow 4CO_{0.48}H_{1.83}N_{0.11}P_{0.01} + 3(1/2)\ O_2 \qquad (1)$$

The ratio of molecular weight of CO_2 and biomass, by the above equation is 1.882, which is within the range of various elemental analyses performed – 1.81 to 2.37 [14].

2.6 Optimization analysis

The optimum conditions for the highest carbon fixation rate will be determined from the results of experimental runs done based on varies operating conditions from Table 2. The Design Expert Software will then calculated these quantitative results to perform a desired response by function of smooth curve that indicate the optimum point for each parameter to obtain highest carbon fixation rate.

3 Results and discussion

3.1 Experimental results and suggestion of optimized parameters

Total of 21 runs of experiments were conducted consisting the interaction of four parameters. Table 3 showed the results for experimental runs performed based on the tabulated data by the Design Expert Software.

Table 3: Experimental results for suggestion of optimized parameters.

Run	Factor 1 Temperature (°C)	Factor 2 pH	Factor 3 Gas flow rate (L/m)	Factor 4 Luminance (lux)	RESPONSE CO_2 fixation rate (gCO_2/day)
1	30.00	6.00	0.15	2500.00	0.8783
2	30.00	6.00	0.25	1500.00	0.3137
3	30.00	6.00	0.15	1500.00	1.4429
4	35.00	5.00	0.10	2000.00	0
5	20.00	6.00	0.15	1500.00	0.3485
6	30.00	6.00	0.15	1500.00	1.5056
7	35.00	7.00	0.10	1000.00	3.5041
8	35.00	7.00	0.20	1000.00	3.2084
9	30.00	8.00	0.15	1500.00	2.599
10	25.00	7.00	0.20	2000.00	2.4914
11	30.00	6.00	0.15	1500.00	1.3174
12	30.00	4.00	0.15	1500.00	0
13	30.00	6.00	0.15	1500.00	1.2064
14	30.00	6.00	0.15	500.00	0.4929
15	35.00	5.00	0.20	2000.00	0.3674
16	25.00	5.00	0.10	1000.00	0
17	30.00	6.00	0.15	1500.00	1.2547
18	40.00	6.00	0.15	1500.00	0
19	25.00	7.00	0.10	2000.00	3.4683
20	25.00	5.00	0.20	1000.00	0
21	30.00	6.00	0.05	1500.00	1.2547

From the results, the highest carbon fixation rate was predicted from Run 7 with fixation rate of 3.5041 gCO_2/day at temperature of 35°C, pH 7, gas flow rate of 0.10 L/m and luminance of 1000 lux. These results were then analysed by Response Surface Methodology (RSM) using Design Expert Software to obtain the actual optimum parameters for highest carbon fixation rate of *Isochrysis* sp. The 3D response surface and 2D contour plots are graphical representation of the regression equation in order to determine the optimum values of the variables [17, 18]. The 3D response surface and 2D contour plots of the interaction are represented in Figure 2 showing the interaction effect of temperature, pH, gas flow rate and lighting on carbon fixation rate of *Isochrysis* sp.

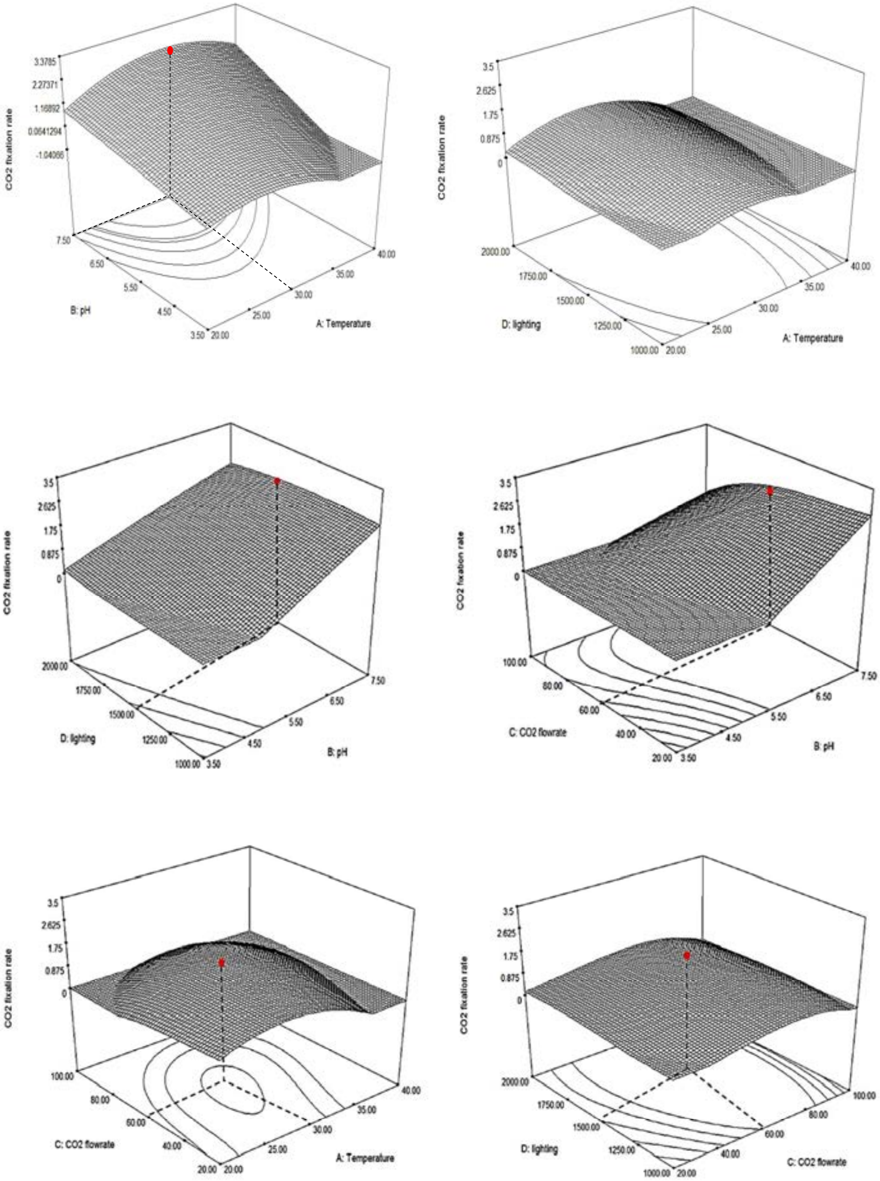

Figure 2: Highest CO_2 fixation rate achieved in correlation between optimum parameters.

These graphs showed the optimum interaction between all the parameters involved. Thus from these graph, the optimum point of PBR operating conditions for highest CO_2 fixation rate was achieved at temperature 30°C, pH 7.5, gas flow rate 0.15L/min and luminance 1500lux. A verification run was conducted using these optimum conditions and the carbon fixation rate of 3.6833g CO_2/day was obtained. This showed that optimization analysis of operating conditions lead to 5.1% improvement of CO_2 fixation rate from the suggested run in Table 3.

3.2 Effects of operating parameters

From the optimization results obtained in Figure 2, the preferable range for *Isochrysis* sp. growth is at 30°C. The growth rate was found slightly slower at lower temperature and inhibited at higher temperature. As stated in many technical publication, the optimum temperature for microalgae growth is highly dependent on the microalgae species being used, normally 28–30°C [5]. pH is the other most critical parameter to sustain microalgae growth and from the optimization analysis, *Isochrysis* sp. growth was preferable at pH 7–8. pH was directly affected by the gas composition especially CO_2 and SO_2 solubility.

Simulated flue gas flow rate gave high impact on algae growth rate where higher exposure of CO_2 and SO_2 from the flue gas could provide the acidic environment needed for the microalgae growth and it may delay or inhibit the microalgae growth [8]. Thus it is crucial to optimize the amount of gas flow rate for highest CO_2 fixation rate. Optimization of gas flow rate on flue gas had never been done before, previous studies only concern on the CO_2 concentration [11]. From the optimization 3D response surface and 2D contour plots, the optimum condition was achieved at 0.15L/min. This explains that at higher gas flow rate, higher gas composition will increase the rate of pH drop and produce hydrodynamic stress to the algae thus not preferable for microalgae growth. Thus, optimum gas flow rate will also ensure a better gas transfer rate which will sustain the optimum pH level at a longer time, and ensure proper mixing of algae and nutrient. The illuminance was set for 12 hour light–12 hours dark, according to normal sunlight. From the results obtained, the effect was not so obvious due to the small gap of artificial light however the range of luminosity can be adapted by the microalgae under sunlight range was reported to be from 5000–10,000 klux [4, 5, 19].

4 Conclusion

Response Surface Methodology (RSM) has been used in this work to perform optimization routine and validate the result. The variables consist of the operating parameters for a 10-litre lab-scaled bubbling column photobioreactor, which include temperature, pH, gas flow rate and luminance. The species used was *Isochrysis* sp. which was isolated from the coastal coal-fired power station. The maximum CO_2 fixation rate obtained was 3.683g CO_2/day at the following optimum culture conditions: 30°C culture temperature, pH 7.5, gas flow rate of 0.15 L/m and at luminance intensity of 1500 lux.

The optimum culture temperature of 30°C proves to be close to ambient conditions of most of South East Asia countries suggesting this region's ambient temperature is conducive for optimum growth of microalgae. The optimum pH 7.5 however suggests a pH control challenge to be tackled for cultures receiving actual flue gas from coal-fired power plants as the flue gas contains acidic components, especially that of SO_2. Fortunately, the almost neutral pH environment offers simpler and safer liquid handling procedure, especially during sub-culturing or disposal.

Future works that can further enrich the knowledge gained in this work could enhance the growth rate of algae used not just in coal-fired power plant's environment but in other industries as well. The anticipated industrial symbiosis will emerge, with microalgae be the focal agent, and would justify coal as a 'green' fuel.

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