

Video Article

Optimize Flue Gas Settings to Promote Microalgae Growth in Photobioreactors via Computer Simulations

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Abstract

Flue gas from power plants can promote algal cultivation and reduce greenhouse gas emissions¹. Microalgae not only capture solar energy more efficiently than plants³, but also synthesize advanced biofuels^{2,4}. Generally, atmospheric CO₂ is not a sufficient source for supporting maximal algal growth⁵. On the other hand, the high concentrations of CO₂ in industrial exhaust gases have adverse effects on algal physiology. Consequently, both cultivation conditions (such as nutrients and light) and the control of the flue gas flow into the photo-bioreactors are important to develop an efficient “flue gas to algae” system. Researchers have proposed different photobioreactor configurations^{4,6} and cultivation strategies^{7,8} with flue gas. Here, we present a protocol that demonstrates how to use models to predict the microalgal growth in response to flue gas settings. We perform both experimental illustration and model simulations to determine the favorable conditions for algal growth with flue gas. We develop a Monod-based model coupled with mass transfer and light intensity equations to simulate the microalgal growth in a homogenous photo-bioreactor. The model simulation compares algal growth and flue gas consumptions under different flue-gas settings. The model illustrates: 1) how algal growth is influenced by different volumetric mass transfer coefficients of CO₂; 2) how we can find optimal CO₂ concentration for algal growth via the dynamic optimization approach (DOA); 3) how we can design a rectangular on-off flue gas pulse to promote algal biomass growth and to reduce the usage of flue gas. On the experimental side, we present a protocol for growing *Chlorella* under the flue gas (generated by natural gas combustion). The experimental results qualitatively validate the model predictions that the high frequency flue gas pulses can significantly improve algal cultivation.

Video Link

The video component of this article can be found at <https://www.jove.com/video/50718/>

Protocol

1. Algal Cultivation and Scale-up

1. Prepare the culture medium using deionized water containing 0.55 g/L⁻¹ urea, 0.1185 g/L⁻¹ KH₂PO₄, 0.102 g/L⁻¹ MgSO₄·7H₂O, 0.015 g/L⁻¹ FeSO₄·7H₂O and 22.5 µl microelements (18.5 g/L⁻¹ H₃BO₃, 21.0 g/L⁻¹ CuSO₄·5H₂O, 73.2 g/L⁻¹ MnCl₂·4H₂O, 13.7 g/L⁻¹ CoSO₄·7H₂O, 59.5 g/L⁻¹ ZnSO₄·5H₂O, 3.8 g/L⁻¹ (NH₄)₆Mo₇O₂₄·4H₂O, 0.31 g/L⁻¹ NH₄VO₃). Adjust medium pH to 7-8. Sterilize culture medium via 0.22 µm syringe filter.
2. Inoculate *Chlorella* sp. from a single colony on a fresh agar plate into a shake flask containing 50 mL medium with a sterile inoculating loop. Culture algae under 150 rpm and 30 °C for six days (continuous light condition, photon flux = 40-50 µmol m⁻² sec⁻¹). Monitor cell density by a spectrophotometer (OD₇₃₀).
3. Transfer 50 mL algal culture (middle-log growth phase, OD₇₃₀ > 1) into a 2-L glass flask (with ~1 L sterilized culture medium). Pump filtered air (or CO₂) into the culture during the incubation (for 5 days).
4. Transfer 1 L algal culture into a 20-L glass carboy containing 15 L non-sterilized culture medium (at this stage, risk of microbial contamination is small), then culture algae under same condition as stated in step 1.3.
5. Place 15 L fresh algal culture (OD₇₃₀ = 2) and 85 L non-sterilized medium into a flat plate photobioreactor (equipped with light-emitting diodes, computer controller, gas mixture, analyzers for cell optical density, pH, dissolved oxygen, temperature and dissolved CO₂). Pump the flue gas/air mixture into the bioreactor.
6. Thoroughly dry-clean the photobioreactor using 70% ethanol after biomass harvest (OD₇₃₀ > 20).

2. Laboratory Demonstration of Flue Gas Treatment Using Small Photobioreactors

1. Inoculate algal cultures in glass bottles (200 ml/min medium/bottle, initial OD₇₃₀ ~0.3).
2. Burn natural gas and pump the flue gas (~250 cm³ min⁻¹) through a funnel, a condenser tube, and a 0.5 L washing bottle (containing water/limestone slurry).
3. The mass flow controllers control the flue gas flow into algal culture (**Figure 1**). Flue gas pulses include two modes: flue gas-on and flue gas-off (pump air instead).

3. Kinetic Model Development

The kinetic model assumes: (1) the cultures are homogeneous systems. (2) CO₂ concentration and light intensity in the cultures are the limiting factors for algal growth. (3) CO₂ partial pressure and its liquid phase equilibrium with H₂CO₃, HCO₃⁻, and CO₃²⁻ is simplified with Henry's Law). The model equations are:

$$\frac{dX}{dt} = \frac{S}{S+K_s+S^2/K_i} \cdot \frac{I}{I+K} \cdot \prod_{i=1}^n \left(1 - \frac{P_i}{P_{max,i}}\right)^{\eta_i} \cdot \mu_{max} \cdot X - k_d \cdot X \quad (1)$$

$$\frac{dS}{dt} = K_{La} (P/H - S) - Y_{S/X} \cdot \left(\frac{S}{S+K_s+S^2/K_i} \cdot \frac{I}{I+K} \cdot \prod_{i=1}^n \left(1 - \frac{P_i}{P_{max,i}}\right)^{\eta_i} \cdot \mu_{max} \cdot X \right) \quad (2)$$

X is the biomass (kg·m⁻³). S is the dissolved CO₂ (mol·m⁻³). P is the CO₂ partial pressure in the gas phase (Pa). p_i is the partial pressure of *i*th toxic compound in the gas (such as NO_x and SO_x). P_{max,i} is the partial pressure of toxic gas to have full inhibition on biomass growth. η_i is the empirical coefficient. K_s is the Michaelis-Menten constant of CO₂ (mol·m⁻³). K_i is the inhibition constant of CO₂ (mol·m⁻³). K is the Michaelis-Menten constant of light intensity (μmol·m⁻²·sec⁻¹). H is the Henry's constant for CO₂ (Pa·m³·mol⁻¹). K_{La} is the mass transfer rate of CO₂ (hr⁻¹). I is the average light intensity, μmol·m⁻²·sec⁻¹, which can be calculated as follows (Eq. (3))⁹.

$$I = \frac{I_0}{A \cdot X} (1 - e^{-A \cdot X}) \quad (3)$$

The definition of model parameters is in **Table 1**. The initial conditions assume that biomass and dissolved CO₂ concentrations are 100 mg/L and 13 μmol/L, respectively. The volumetric mass transfer coefficient can be estimated by empirical correlation to bioreactor parameters¹⁰:

$$K_{La} = \alpha \left(\frac{P_g}{V} \right)^{\beta} (u_{gs})^{\gamma} \quad (4)$$

P_g/V is the power consumption of the aerated system in the bioreactor (W/m³). u_{gs} is the superficial velocity of the gas flow through the bioreactor (m/sec). α, β, and γ are constants related to mixing conditions.

1. Construct a Simulink file for the model simulation (Screen shots are given in the Supporting Material I).
 1. Choose File/New/Model on the MATLAB interface to create a Simulink model, and open "Library Browser" (screen shot 1).
 2. Choose 'Subsystem' block in the library browser to create the Subsystems for Equation 1 and 2. Drag one subsystem block to the Simulink model file, change its name to 'Equation 1', and then repeat the same steps for Equation 2.
 3. Create appropriate blocks and parameters in each subsystem (screen shot 3). Double click the 'Equation 1' block, choose appropriate blocks from the library browser and connect them with arrows that denote the calculation sequence, double click the blocks to set up the parameters, and repeat these steps for the other subsystem.
Note: 1) The sequence should start with input blocks and conclude with output blocks; 2) The operator blocks for addition, subtraction, multiplication, division and integration can be all found in the library browser, and we suggest users explore the help files of the Simulink to understand how to use them; 3) The optimization solver can be set through the pathway Simulation/Configuration parameters on the toolbar.
 4. Link the two subsystems to represent model equations (1 and 2). Connect the output of one subsystem to the input of the other subsystem by arrow if necessary. For example, the dissolved CO₂ concentration is the output in the Equation 2 subsystem, and also the input of the Equation 1 subsystem.
 5. Use 'Pulse Generator' block as the inputs for 'Equation 2' to simulate the on-off CO₂ pulses; use 'Constant' block as the surface light input value. Double click the blocks to change the parameters such as the period time and amplitude.
 6. Choose 'Mux' block in the library browser. Connect all the outputs to 'Mux' and then connect it to 'To Workspace' block that stores the simulated results.
 7. Define the 'Simulation stop time' on the top toolbar, click the button "▶" to start the simulation, and the results will be shown in the MATLAB workspace (screen shot 4).
2. Apply dynamic optimization approach to profile optimal CO₂ conditions.

To find the changes of inlet inflow CO₂ profile (P_{opt}) that maximize biomass production¹¹, MATLAB 'fmincon' function and CVP (control vector parameterization)¹² are used. **Figure 2** illustrates the optimization algorithm (see MATLAB programming codes in the Supporting Material II).

Representative Results

Our previous experimental analysis indicates that continuous flue gas exposure adversely affects the *Chlorella* growth, while decreasing CO₂ exposure time is able to alleviate this inhibition¹³. To better understand the flue gas inflow and algal growth relationship, we develop an empirical model to simulate the biomass growth in the presence of flue gas. We assume that the flue gas contains 15% CO₂ (note: The typical CO₂ concentration from coal combustion is 10-15%, while flue gas from oxy-combustion power plant has CO₂ >15%). The mass transfer and algal growth parameters are based on **Table 1**. The model simulation tests three methods to avoid growth inhibition by flue gas: 1. Keep low flow rate into the culture to reduce the mass transfer condition. 2. On-off pulses of flue gas into the culture. 3. Control the inflow CO₂ compositions at the optimal level.

Firstly, we test the influence of mass transfer rate on the algal growth (**Figure 3a**), which indicates that optimal mass transfer rate ($K_{La} = 0.17\text{--}0.18\text{ hr}^{-1}$) is able to reduce the flue gas inhibition to algal growth. If K_{La} is lower or higher than the optimal value, the algal growth will be reduced. Equation 4 suggests the decrease of aeration and gas flow through the culture can reduce the mass transfer coefficient. **Table 2** shows how the flow rate (i.e., superficial velocity) affects the algal growth. Generally, low flow rate reduces K_{La} and prevents CO₂ inhibition to algal growth as the same trend shown in **Figure 3**. Further reducing flow rate through bioreactor will cause the mass transfer coefficient too small to provide enough CO₂ for algal growth (**Figure 3b**).

Secondly, we introduce an on-off flue-gas pulse mode to overcome growth inhibition if flue gas mass transfer K_{La} is high in the photobioreactor (i.e. $K_{La} = 17\text{ hr}^{-1}$). In the simulation, we assume the algal cultures are pulsed with two different CO₂ concentrations (15% for flue-gas-on and 0.04% with atmospheric CO₂ for flue-gas-off). To optimize the flue-gas pulse mode, different on-off frequencies are tested (**Figure 4**). The simulation shows that high frequency flue gas pulses (on-off control of flue gases) are able to promote algal growth. **Table 2** also indicates that on-off control mode uses less flue gas comparing to continuous feeding of flue gas into the bioreactor.

Thirdly, we calculate CO₂ concentration profiles for maximal algal growth. Using model parameters in **Table 1**, the dynamic optimization approach shows the optimal CO₂ concentrations in the gas phase should be continuously increased during algal growth. Model simulation also shows that both the on-off CO₂ pulses (Method 2) and the control of optimal CO₂ input (method 3) are equally good to promote the algal growth with flue gas (**Figure 5**).

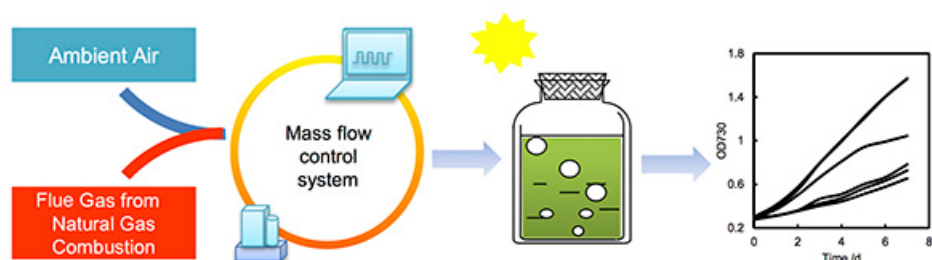


Figure 1. Diagram of the gas on-off control system at laboratory scale. The flow rates of flue gas generated by natural combustion are controlled by the mass flow control system before introduced into the algal system. [Click here to view larger image.](#)

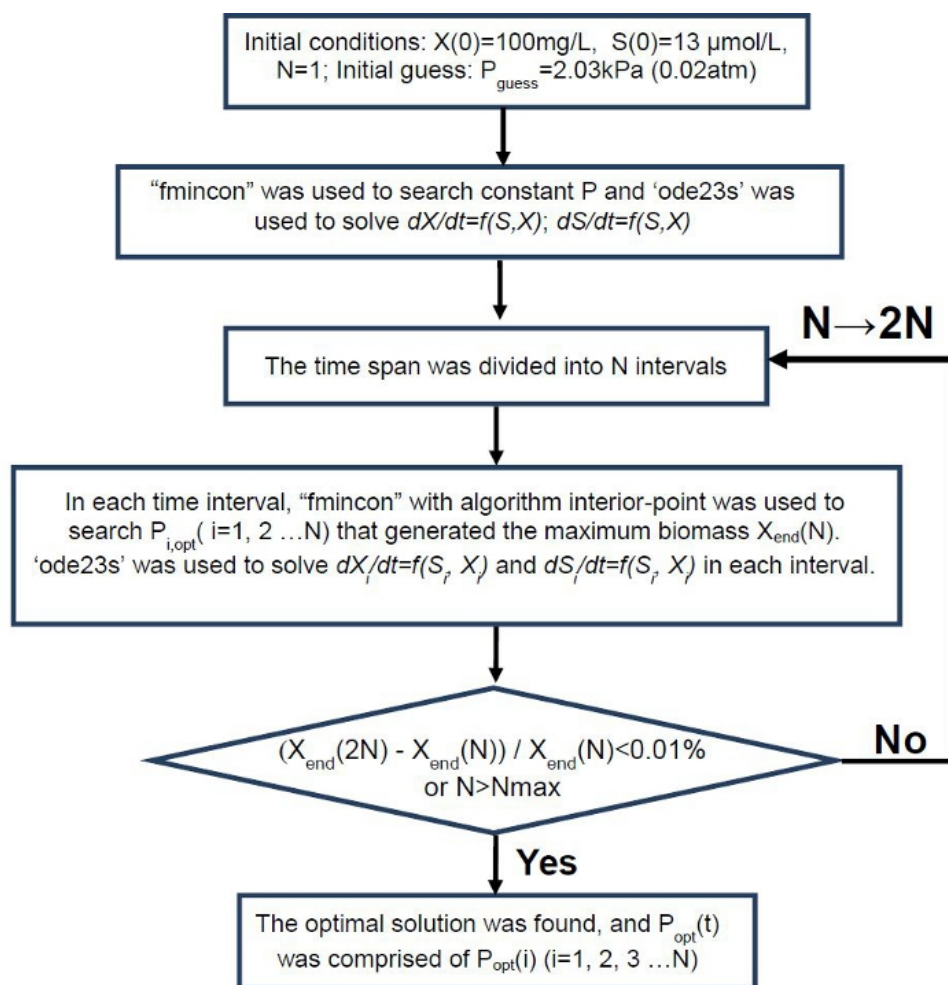


Figure 2. Flow chart of dynamic optimization procedures. [Click here to view larger image.](#)

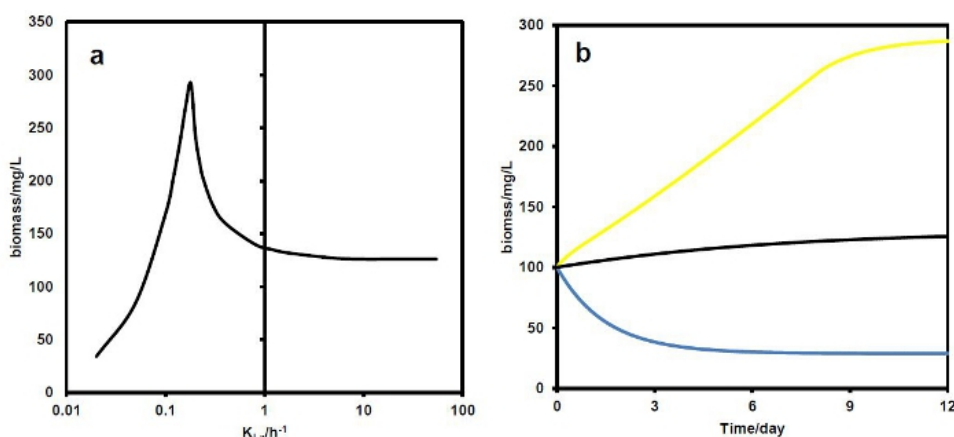


Figure 3. Final biomass concentration at day 12 as a function of K_{La} under continuous flue gas treatment (CO_2 , 15% v/v) (a), and the comparison of biomass growth with different K_{La} : 0.017 h^{-1} (blue line), 0.17 h^{-1} (yellow line), and 17 h^{-1} (black line) under continuous flue gas treatment (CO_2 , 15% v/v) (b). [Click here to view larger image.](#)

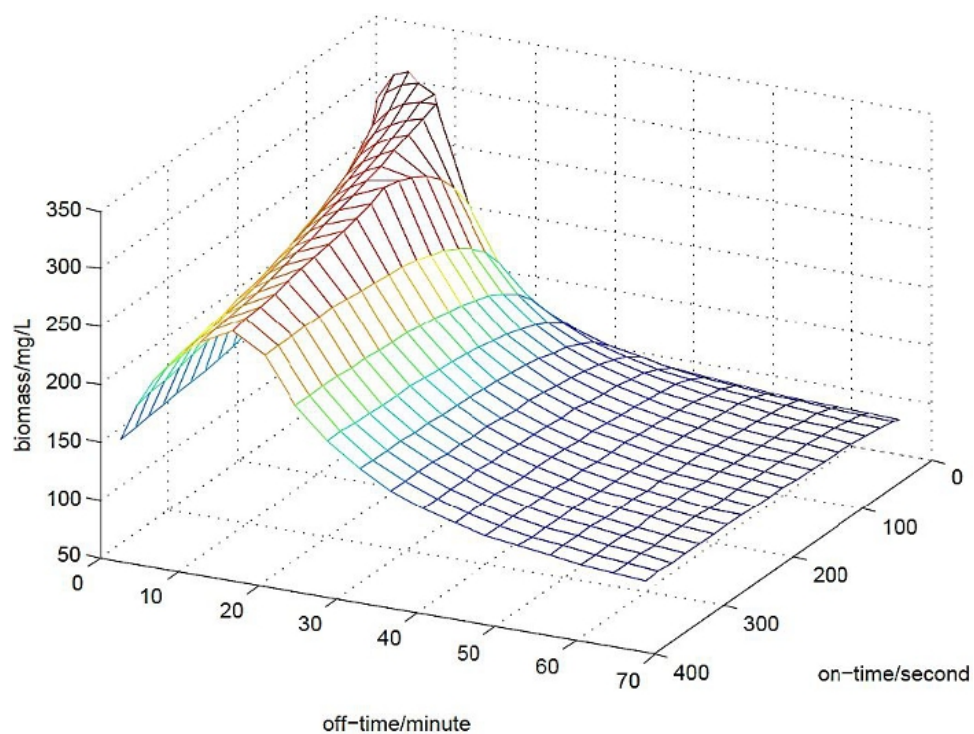


Figure 4. Effect of gas-on/gas-off frequency on biomass production in 12 days. The model assumes the microalgae are exposed to CO₂ (15% v/v) pulses at different tested frequencies. [Click here to view larger image.](#)

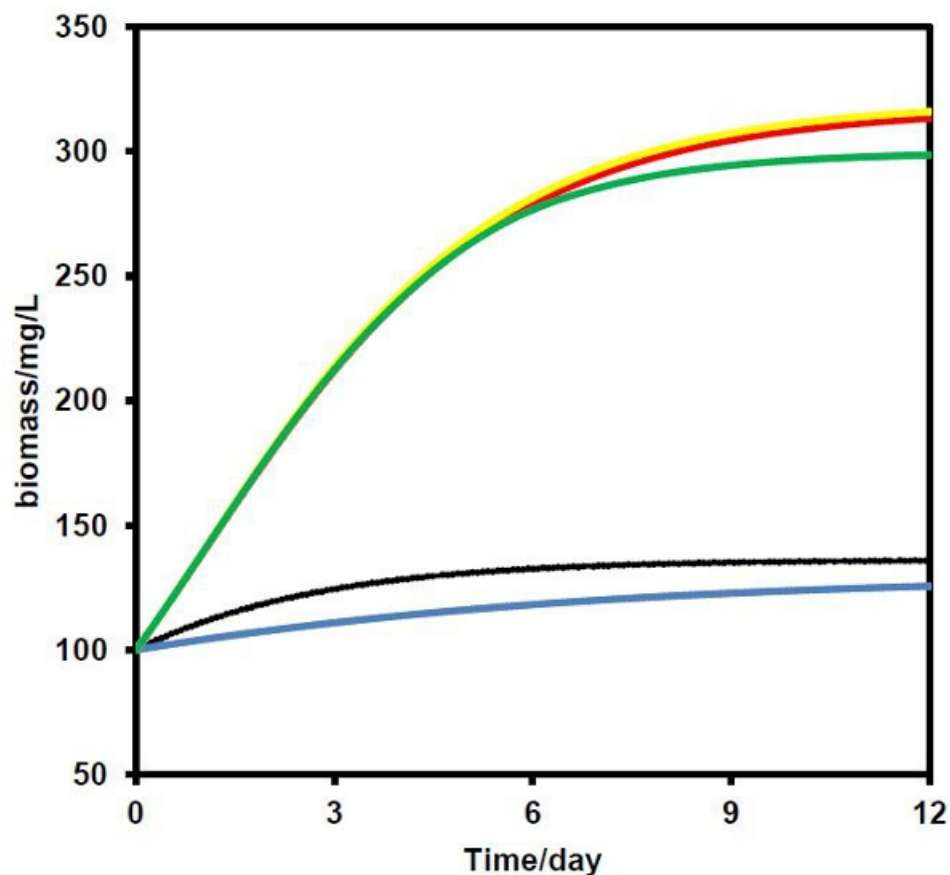


Figure 5. Comparison of biomass growth under optimal CO₂ profile (yellow line), the on-off frequency of 10 sec gas-on/5 min gas-off (red line), on-off control at a frequency of 10 sec gas-on/7 min gas-off (green line), on-off control at a frequency of 1 min gas-on/29 min gas-off (black line), and the continuous treatment with flue gas containing 15% (v/v) CO₂ conditions (blue line). [Click here to view larger image.](#)

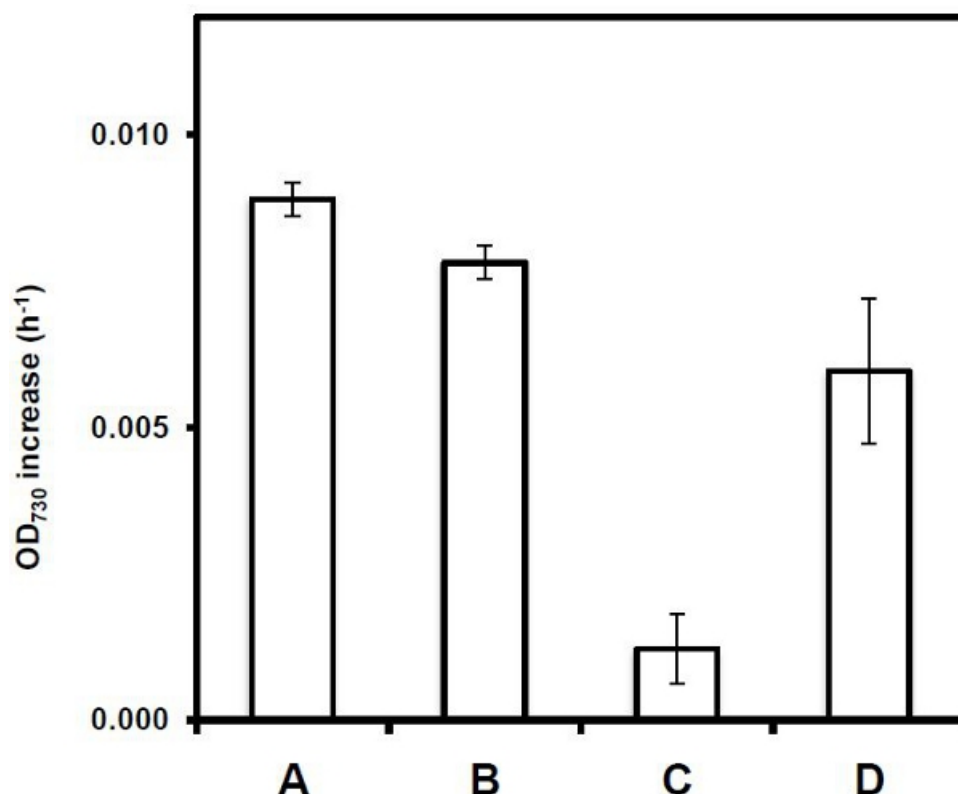


Figure 6. Experimental results from our previous paper¹³ to show effect of flue gas pulses on *Chlorella* growth. Gas-on (flue gas treatment); gas-off (air treatment). A: 10 sec gas-on/7 min gas-off; B: 30 min gas-on/30 min gas-off; C: 5 hr gas-on/7 hr gas-off; D: cultivation in shaking flasks. The culture preparation was detailed in the protocol part, and the experiments were conducted under the room temperature. [Click here to view larger image.](#)

Parameters	Descriptions	Values	Units	References/Notes
μ_{\max}	maximum specific growth rate	0.070	hr ⁻¹	14
k_d	mortality rate	0.028	hr ⁻¹	15
K_s	Michaelis-Menten constant of CO ₂	0.00021	mol·m ⁻³	14
K_i	inhibition constant of CO ₂	10 ^a	mol·m ⁻³	16
K	Michaelis-Menten constant of light intensity	14 ^b	μmol·m ⁻² ·sec ⁻¹	9
K_{La}	mass transfer rate of CO ₂	17	hr ⁻¹	17
H	henry's constant of CO ₂	3202 ^c	Pa·m ³ ·mol ⁻¹	18
$Y_{S/X}$	yield coefficient	100 ^d	(mol CO ₂)/(kg biomass)	19
A	Constant	14.7	m ³ ·kg ⁻¹	9
I_0	surface light intensity	45 ^e	μmol photons·m ⁻² ·sec ⁻¹	measured
Atmospheric CO ₂	atmospheric CO ₂ concentration	0.04%	volume fraction	
CO ₂ in flue gas	CO ₂ concentration in the flue gas	15%	volume fraction	assumed
$X(0)$	initial biomass concentration	0.1	kg·m ⁻³	assumed
$S(0)$	initial dissolved CO ₂ concentration	0.013	mol·m ⁻³	assumed

Table 1. Parameters used in the model.

$K_L=10$ mM, and the test range in this study is $0.5\text{--}10\text{ mol}\cdot\text{m}^{-3}$;
^b $K=1011$ lux, which is $\sim 14\text{ }\mu\text{mol}\cdot\text{m}^{-2}\cdot\text{sec}^{-1\ 20}$;
^c $H=31.6\text{ atm}\cdot\text{M}^{-1}$;
^d 4.4 kg CO_2 is needed for production of 1 kg (dry weight) of biomass;
^e The measured light intensity is $40\text{--}50\text{ }\mu\text{mol}\cdot\text{m}^{-2}\cdot\text{sec}^{-1}$;

Superficial velocity /m/s	Initial biomass /mg/L	$\gamma=0.2$		$\gamma=0.5$		$\gamma=0.8$		Total flue gas used in 12 days (m^3/m^2)
		Final biomass /mg/L	K_{La} /m/s	Final biomass /mg/L	K_{La} /m/s	Final biomass /mg/L		
0.001^*	100	4.3	128	0.54	149	0.068	115	1.0×10^3
0.01^*	100	6.8	127	1.7	132	0.43	160	1.0×10^4
0.1^*	100	11	126	5.4	127	2.7	129	1.0×10^5
1^*	100	17	126	17	126	17	126	1.0×10^6
10^*	100	27	126	54	126	107	125	1.0×10^7
10s/5min frequency	100	17	313	17	313	17	313	3.3×10^4

Table 2. Biomass growth with 15% (v/v) flue gas at day 12 under different superficial gas flow velocities. In this model, we assume that $K_{La}=17(u_{gs})^Y$

*: Assuming that CO_2 is continuously pumped into bioreactor at the constant flow rate.

Discussion

In this study, we demonstrate the experimental protocol for scaling up algal cultivations in photobioreactors. We also examine several methods for flue gas inputs to promote algal growth. Using a mass transfer and bio-reaction model, we demonstrate that the CO_2 mass transfer coefficient K_{La} (determined by bioreactor mixing condition and CO_2 superficial velocity) strongly influences algal growth. The model simulation indicates continuous on-off flue gas pulses with short pulse width and high on-off frequencies can improve *Chlorella* growth (*i.e.* high frequency on-off flue-gas pulses can support biomass growth almost as well as optimal CO_2 conditions, **Figure 5**). Meanwhile, on-off mode can significantly reduce the total amount of flue-gas that has to be pumped through the bioreactor (**Table 2**), which saves the energy for transporting the amount of flue gas for algal cultivation. The on-off gas pulse mode can be used in photo-bioreactors or algal ponds, considering that the mode of constant flue gas pulses is much easier to operate than dynamic control of the inflow CO_2 concentration. On the other hand, we have performed the algal culture experiments using flue gases. Flue gas are pulsed into the photobioreactors at a specific on/off frequency, which clearly minimizes the inhibitory effect of the flue gas and improves biomass production comparing to cultures using atmospheric CO_2 (**Figure 6**)¹³. The experimental results have qualitatively verified our model and confirmed that the on-off control of the flue gas is effective for increasing *Chlorella* growth.

Finally, this model study is subject to several limitations. First, the model does not directly consider the effects from toxic compounds such as the SO_x and NO_x in the flue gas. Second, the chemical reactions and equilibriums in the culture medium (include CO_2 , H^+ , OH^- , NH_3 , *etc.*) are simplified. Third, the model does not take into account CO_2 fluid dynamics, where the actual gaseous mass transfer is not instantaneous or homogeneous in culture medium. However, the simplified model approaches still have practical applications for providing guidelines for optimizing algal growth.

Disclosures

These authors have nothing to disclose.

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