Optimizing the gas distributor based on CO₂ bubble dynamic behaviors to improve microalgal biomass production in an air-lift photo-bioreactor

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Abstract:

Dynamic behavior of bubbles would significantly affect CO₂ mass transfer and may cause microalgae cells uneven distribution due to the bubble carrying effect. To improve microalgae growth, the gas distributor and aeration conditions was optimized according to the bubble rising behavior. The CO₂ bubble rising trajectory is similar to Zigzag. The amplitude and wavelength of the Zigzag, which reflected the influenced zone of microalgae suspension in horizontal direction and disturbance intensity on culture, respectively, was controlled by the structure of gas distributor and aeration conditions. An optimized round gas distributor that full of holes with a inner diameter of 0.5 mm and spacing of 1.5 mm was designed. When cultivated with the optimized gas distributor aerating 5% CO₂ gas at 0.250 vvm, the maximum biomass of *Chlorella pyrenoidosa* achieved 2.88 g L⁻¹, increased by 83.44% compared to that 1.57 g L⁻¹ cultivated with the commercial micro-bubbles aerator.

Keywords: CO₂; Bubbles behaviors; Gas distributor, Microalgae; Photo-bioreactor

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1. Introduction

Greenhouse warming has made the development of efficient and eco-friendly CO₂ capture utilization technology (CCUS) becoming an urgent task. Among the CCUS methods, biochemical conversion by microalgae photosynthesis has been becoming a main trend because of its high CO₂ conversion efficiency, high lipid productivity and low cost (Chang et al., 2017; Ugwu et al., 2008; Yang et al., 2014).

CO₂ is one of the necessary elements for microalgae photosynthesis (Chang et al., 2017), the gas containing CO₂ is usually pumped into culture through a gas distributor in the form of bubbles. During the rising of bubbles, CO₂ molecules gradually diffuse through a gas-liquid interface into the culture and then into microalgae cell, where it is captured by the microalgal cells and converted to carbohydrates (Carvalho et al., 2006; Singha et al., 2016). Therefore, bubbles dynamic behavior in microalgal suspension would seriously affect the transformation process CO₂, thereby significantly influencing on the microalgae growth and CO₂ fixation. Furthermore, in the process of bubble formation and rising, a mass of microalgal cells adsorb on the gas-liquid interface due to microalgae cells collisions on the interfaces and biological chemotaxis, leading to the uneven distribution of cells in photo-bioreactor (Chen et al., 1998). Although bubble carrying has been applied to microalgae harvesting (Barrut et al., 2013; Kim & Kwak, 2014), it may cause a large number of cells gather on the gas-liquid interface. On the one hand, it hindered the transmission. On the other hand, the cells that gather onto the top of suspension may slowly die due to insufficient water supply. Therefore, to keep an appropriate CO₂ bubble rising and mixing state is a key issue for microalgae growth and carbon

optimization.

In 1988, Weissman et al. (1988) reported that turbulent mixing of bubble flow in the photo-bioreactor could affect microalgae growth. Yang et al. (2014) tested air bubbles distribution in the water using high-resolution particle image velocimetry and studied the effect of turbulence mixing on microalgae growth in the plate photo-bioreactor. However, they fail to measure the bubble behavior in culture with microalgae cells due to opacity of microalgae suspension. Besides, the current studies mainly focused on the influence of turbulence mixing on reinforcement of CO₂ and light transmission, few studies on the microalgal cells uneven distribution caused by bubble carrying and its effect on microalgae growth. Although a few studies that the dynamic behavior of bubbles greatly affects microalgae cells distribution et al., 2016), study on how to reduce the inhibition of bubble carrying on microalgal growth has yet been reported. Although the mechanical mixing could make uniformity of microalgal cells distribution in the bioreactor, it consumed lots of energy. some previous studies on CO₂ bubbles dynamic behavior have indicated that the rising trajectory wave was similar to Zigzag, and two vortexes were emerged due to bubble' bending rising. The horizontal offset width determined the turbulent mixing region, which depends not only on airflow rate, but also on the structure of gas distributor and the geometric positioning of the aeration inlets (Ding et al., 2016; Zhao et al., 2015b). While bubble itself could cause turbulence and mixing besides cells carrying effect. In other words, we can control turbulent mixing and bubble carrying regulating structure of gas distributor and the aeration conditions base on CO₂ bubbles

dynamic behavior.

Therefore, with CO₂ bubbles dynamic behavior as theoretical basis, a porous gas distributor was proposed for air-lift photo-bioreactor. The pore size, spacing, and arrangement mode are optimized. Also, their effect on microalgae cells distribution and carbon fixation efficiency were compared to obtain the gas distributor that could efficiently improve microalgal growth.

2. Methods and Materials

2.1. Microalgae species and culture

Microalgae *Chlorella pyrenoidosa* was purchased from Institute of Hydrobiology, Chinese Academy of Sciences (Wuhan, China). *C. pyrenoidosa* was cultivated in the SE (Bristol's solution) medium (Zhao et al., 2015b).

2.2. Experimental system

All the trials were carried out at 25 °C. The fluorescent lamps were fixed next to the photo-bioreactor in parallel to supply continuous illumination of 80 $\mu\text{mol m}^{-2} \text{s}^{-1}$ for microalgae growth, the distance of the lights to the axis of photo-bioreactor was ~100 mm. The photo-bioreactor (with a working volume of 400 mL) was a transparent polymethyl methacrylate (PMMA) cylinder with an inner diameter of 0.042 m and a height of 0.3 m. A purchased commercial uniform sand micro-bubbles aerator was fixed at the bottom of bioreactor for CO₂ gas supply. The designed uniform bubble device (gas distributor) was composed of a stomata plate, sand core flow plate and funnel as shown in Fig. 1. CO₂-enriched gas (balanced with air) was bubbled into the glass funnel, then flow sand core flow plate and arrived in the height is 5 mm buffer

chamber, finally through aeration hole plate flow in microalgae photo-bioreactors to form bubble flow. For the experiments of CO₂ concentrations effect on microalgae growth, the CO₂ concentrations was 3%, 5%, and 10% respectively. For the other experiments, the CO₂ concentration we used was 5%. The hole of stomata plate is in-line arrangement. Effects of different pore size, spacing, CO₂ concentration and rate on microalgae growth and CO₂ fixation characteristics were investigated. The rising process of the bubble was recorded by the data acquisition system which of a cool light, a high speed photography (Phantom V5.1 at the shooting speed of fps) and a data storage system. The recorded bubble images were processed and analyzed by the self-developed programme using Matlab software (Zhu et al., 2014). The images of were converted into grayscale firstly, and then the interference was removed using a background subtraction method. Finally, the bubble edge was extracted by the Canny operator. More than 200 pictures were used to state out the zigzag movement of bubble. The measurement repeated at least three times and data showed as the average value. All experiments in the manuscript were performed in duplicate, and all data showed were reported as mean values and standard deviations this study.

2.3. Biomass concentration and composition measurement

The concentration of the microalgae suspension was characterized by optical density at a wavelength of 680 nm (OD_{680nm}) measured by UV-VIS spectrophotometer (TU-1901, China). Dry weight of *C. pyrenoidosa* can be estimated by Eq. (1) (Ding et al., 2016).

$$D = 0.51OD_{680nm} - 0.029 \quad (R^2=0.99) \quad (1)$$

Where D (g L^{-1}) is the dry weight of microalgae and $OD_{680\text{nm}}$ is the optical density of microalgae suspension.

Biomass growth rate (P , $\text{g L}^{-1} \text{d}^{-1}$) was chosen to assess microalgae growth, which is calculated from the average variation in the dry microalgae biomass concentration D (g L^{-1}) within per day according to Eq. (2) (Ding et al., 2016).

$$P = \frac{\bar{D}_\tau - \bar{D}_0}{\tau} \quad (2)$$

Where $\bar{D}_\tau$ (g L^{-1}) is the average dry microalgae biomass concentration at day of τ , $\bar{D}_0$ (g L^{-1}) is the initial dry microalgae biomass concentration.

Chlorophyll content was determined according to the method described by Lorenzen (1967). At the end of cultivation, all the microalgal suspension was centrifuged at 8,000 rpm for 10 min to microalgae slurry and then dried at 105 °C for biomass composition measurement. The lipid content of dried biomass was measured by modified Bligh-Dyer method (Bligh & Dyer, 1959). Total carbohydrates were determined by phenol-sulfuric acid method (Michel Dubois, 1956), and total protein was determined by Coomassie brilliant blue method (Sedmak & Grossberg, 1977).

2.4. Calculation of CO₂ absorption and fixation rate

CO₂ concentration (C_{CO_2}) was measured using gas chromatograph (plus GC 2010, Shimadzu). CO₂ removal efficiency ε indicates of the ability of the photo-bioreactor absorb CO₂ and is estimated by Eq. (3).

$$\varepsilon = \frac{C_{\text{CO}_2, \text{in}} - C_{\text{CO}_2, \text{out}}}{C_{\text{CO}_2, \text{in}}} \times 100\% \quad (3)$$

Where, $C_{\text{CO}_2, \text{in}}$ is inlet CO₂ concentration to the bioreactor, $C_{\text{CO}_2, \text{out}}$ is outlet CO₂

concentration from the bioreactor.

Photosynthetic CO₂ fixation rate ξ ($\mu\text{mol m}^{-3} \text{s}^{-1}$) represents the intensity of CO₂ fixation via photosynthesis, defined as the CO₂ absorption amount by per unit volume of microalgal cells per unit time. ξ is calculated by using Eq. (4).

$$\xi = \frac{(C_{\text{CO}_2, \text{in}} - C_{\text{CO}_2, \text{out}}) \times Q_g}{1.47 \times V} \quad (4)$$

Where, Q_g (mL min^{-1}) is the gas flow rate, V (m^3) is the volume of the microalgal suspension.

3. Results and discussion

3.1. Effect of microalgae cells distribution on growth

Bubble carrying effect may cause uneven distribution of microalgal cells in the suspension, which thereby causes serious inhibition on microalgal growth. *C. pyrenoidosa* growth curves under the conditions of both uniform and non-uniform cells distribution were compared (Fig. 2). When cultivated with 5% CO₂ gas at a flow rate of 0.1 vvm under light intensity of $80 \mu\text{mol m}^{-2} \text{s}^{-1}$, *C. pyrenoidosa* only grew in the first 2 days and the biomass concentration was stable at 1.57 g L^{-1} without auxiliary mechanical mixing (non-uniform distribution). While if cells distributed evenly by mechanical stirring, *C. pyrenoidosa* kept thriving until the 8th day and the maximum biomass concentration was up to 4.73 g L^{-1} (2.97 times increased). This can be attributed to the following reasons. During microalgae growth, a large number of microalgal cells were carried by bubbles to the top of culture, where CO₂ and other nutrients that necessary for microalgae growth were lack without auxiliary mechanical mixing. Thereby it inhibited microalgae growth. Nevertheless, with the aid of

mechanical mixing, cells that were carried up by bubbles were carried down again (Yang et al., 2014; Yang et al., 2016). Cells would distribute evenly in microalgae culture. And the majority of homogenization of microalgal cells were exposed to CO₂ and nutrients in the culture and could effectively capture CO₂ and nutrients, which could promote cells' reproductive division and accumulation of organic matters. As a result, microalgae growth period extended and biomass productivity increased.

Although the uniformity of microalgae cells distribution in the bioreactor plays a very significant role in the microalgae growth, the mechanical mixing consumed lots of energy, it usually lose more than gain (in the form of generated biomass energy). While bubble itself could cause turbulence and mixing besides cells carrying effect. Therefore, a gas distributor can be designed based on the dynamics behavior of CO₂ bubbles, and the operation parameters can be optimized to make cells distribute evenly and reduce or eliminate the growth inhibition caused by uneven distribution of cells.

3.2. Dynamic behaviors of CO₂ bubbles in microalgae suspension

Mixed gas with 10% CO₂ (balanced with N₂) was aerated into microalgae suspension (OD_{680nm}=0.97) from a capillary with inner diameter of 0.5 mm. The rising trajectory (Fig. 3(a)) of bubble that detached from the capillary was determined by extracting the centroid position of bubble in different time. It can be observed that bubble rising trajectory in the microalgae suspension like Zigzag and could be mainly divided into three phases. At the beginning stage, the bubble rising straightly and the vertically rising velocity gradually increased under the driving of buoyancy force and drag force,

which is proportional to the average velocity of bubble (Zhao et al., 2015b). This stage can also be called phase 1. In phase 2, as bubble rising velocity increased with time, two vortices were evolved at the tail of bubbles due to microalgae culture flowing around the bubble. The vortices make bubbles under unbalanced horizontal forces. And bubble rising trajectory started to bend in the horizontal direction until the drag force increased to the equal value as the buoyancy force.

The balance vertical height to the gas outlet was 11.4 mm. However this 11.4 mm of balance height was too small compared with the height of photo-bioreactor (300 mm). Which means the following stable rising stage (phase 3) was the most important for microalgae growth. In phase 3, the bubble rising trajectory wave was stable and like a Zigzag and the bubble rising velocity was only slightly fluctuated near the terminal velocity. The amplitude (A) and wavelength of bubble rising trajectory (λ) reflected the influenced microalgae suspension zone of bubble in the horizontal direction and the disturbance intensity on culture, respectively. They also indirectly reflect transfer zone of CO_2 molecule in microalgae suspension. In other words, the value of amplitude and wavelength of bubble rising effect directly microalgae cells distribution and growth.

While the bubbles rising in photo-bioreactor depends on structure of gas outlet and operation conditions. The effect of inner diameter (d_0) of gas outlet on the bubble dynamic behaviors (Fig. 3(b)) indicated the amplitude and wavelength of rising trajectory was increased with the increasing of inner diameter of gas outlet. With the increase of inner diameter of gas outlet, the gas-liquid interfacial specific surface area

decreased. Although the decreased gas-liquid interfacial specific surface area would weaken the disturbance in liquid phase and unfavorable for CO₂ transfer, it could increase the influence zone of the bubble rising (Zhao et al., 2015b). Moreover, the larger the inner diameter of gas outlet was, the more cells that would absorb on the gas-liquid interface and the bigger bubble rising velocity and acceleration became. That would strengthen the disturbance of microalgae suspension (Zhao et al., 2015a). Comprehensive considering influence area, turbulence intensity on liquid phase, and cells adsorption, the inner diameter was chosen from 0.1 mm to 1 mm in this study. Moreover, there are a lot of holes in a gas distributor. The distance of adjacent holes would also influence rising of bubble. According to a previous study (2015a), the optimal distance of adjacent holes was three times of d_0 .

3.3. Optimization of gas distributor based on CO₂ bubbles behaviors

The aperture of porous aeration plate have great influence in bubble formation and bubble detachment process and dynamics behavior of the bubble rising process, which affect the CO₂ mass transfer and bubble carrying, eventually impact on microalgae growth and carbon sequestration performance in the microalgae photo-bioreactor. Therefore, we first studied the effect of pore size (0.3 mm, 0.5 mm, 1 mm) on performance of microalgae growth and carbon sequestration under the inlet CO₂ concentration of 5%, the gas flow rate of 100 mL min⁻¹, the stomata plate spacing S of 2.5 mm.

A previous study showed that the smaller the diameter of gas outlet, the longer the bubbles residence time, which was favorable for CO₂ transfer (Zhao et al., 2015b).

Therefore, when cultivated with a gas pore size d_0 of 0.5 mm, microalgae showed the highest photosynthetic CO_2 fixation rate of $909.01 \mu\text{mol m}^{-3} \text{s}^{-1}$, maximum biomass yield of $0.63 \text{ g L}^{-1} \text{d}^{-1}$ (Fig 4(a)) and fastest CO_2 removal efficiency of 10.69% (Table 1). While cultivated in the photo-bioreactor with a gas pore size of 1.0 mm, microalgae showed a lower CO_2 removal efficiency of 8.31%. Besides, the shorter CO_2 bubbles residence time made CO_2 supply inadequate, microalgae suspension became more alkaline. As a result, the photosynthesis and growth of microalgae was restricted, microalgae had lower photosynthetic carbon fixation rate and biomass yield. However with a smaller gas pore size ($d_0=0.3$ mm), the detached bubble had a smaller diameter and longer residence time. This is beneficial for CO_2 molecules transfer. And pH of microalgae suspension was not too high. This is also beneficial for microalgae growth. However, due to the smaller bubble, the bubble carrying phenomenon more obvious, more microalgal cells were carried up to the top of microalgae suspension which inhibited microalgal growth. The negative effect of bubble carrying can offset its positive effect on CO_2 transfer. Therefore, microalgae that cultivated with a d_0 of 0.3 mm showed a lower carbon fixation rate and biomass yield.

The effect of spacing of adjacent holes (S) in gas outlet indicated that when cultivated with a spacing of 1.5 mm ($S=3d_0$), microalgae had the largest CO_2 removal efficiency of 11.31%, the biggest photosynthetic CO_2 fixation rate of $961.73 \mu\text{mol m}^{-3} \text{s}^{-1}$, and the maximum biomass growth rate of $0.64 \text{ g L}^{-1} \text{d}^{-1}$ (Table 1). And the largest biomass concentration was 2.88 g L^{-1} increased by 83.4% to that of 1.57 g L^{-1} cultivated with a ordinary sand core aeration (Fig 4). When the spacing increased to

2.5 mm ($S=5d_0$), there was just a little decrease in microalgae growth rate and biomass concentration. While when the spacing decreased to 1mm ($S=2d_0$), microalgae grow slowly and biomass concentration decreased to 2.39 g L^{-1} (Fig 4(b)). This mainly due to bubbles dynamic behaviors. The adjacent bubbles would not merge that emitted out from gas outlet with a spacing of 2.5 mm, the bubbles had a smaller size and rising rate. When the spacing was from 1mm to 2 mm, the two adjacent bubbles would merge together. Especially the detached bubbles diameter was smallest with the spacing of 1.5 mm, even smaller than that emitted from the spacing of 2.5 mm. Thereby, when emitted out from the gas outlet with the spacing of 1.5 mm ($S=3d_0$), the bubbles was smallest and fluctuation of gas-liquid interface was very strong. CO_2 transmission rate was largest (Ding et al., 2016; Zhao et al., 2015b). That would supply more effective area and chances for microalgae cells to contact and fix CO_2 molecules. And microalgae showed a maximum CO_2 removal efficiency of 11.31%.

The bubbles that from the gas outlet with spacing of 2.5 mm was slightly larger than the bubbles that from the gas outlet with spacing of 1.5 mm. The residence time of CO_2 bubbles in culture was a little shorter and fluctuation of gas-liquid interface a little weaker. These eventually lead to a lower CO_2 transmission rate. But the difference was not obvious. While with the spacing of 1 mm, the growth characteristics of the merged bubbles was similar with the single bubble. Thereby the gas-liquid interface fluctuation of the merged bubble would be much smaller but with a largest detached diameter, fastest rising rate, and shortest CO_2 bubbles residence

time in microalgae culture. Thus, CO₂ transmission rate was lowest. And microalgae showed a minimum CO₂ removal efficiency of 8.78% and lowest photosynthetic CO₂ fixation rate of 746.60 $\mu\text{mol m}^{-3} \text{s}^{-1}$ (Table 1).

To sum up, microalgae showed a largest growth rate and carbon sequestration ability that cultivated in the bioreactor equipped with the plate gas outlet in the pore size of 0.5 mm, spacing of 1.5 mm. These results consistent with the prediction results according to bubble dynamics, which also verified the feasibility of gas distributor design based on bubble dynamics behaviors.

3.4. Performance of gas distributor in a cylindrical bioreactor

Besides the size of the gas distributor itself, ventilation conditions (such as gas flow rate and CO₂ concentration) may also affect the dynamic behavior of bubbles and microalgae growth (Zhao et al., 2015b). CO₂-enriched gas was bubbled into microalgae culture as a carbon source for *C. pyrenoidosa* growth. The effect of CO₂ gas (5%, v/v) flow rate on microalgal growth is as shown in Table 2 and Fig. 5. When cultivated with a gas flow rate of 0.250 vvm, the maximum microalgae biomass concentration was up to 2.88 g L⁻¹ (with a growth rate of 0.64 g L⁻¹ d⁻¹), which was 1.22 and 1.07 times higher than that cultivated with gas flow rate of 0.125 vvm and 0.375 vvm, respectively (Fig. 5(a)). The results indicated that lower gas flow rate cannot provide sufficient CO₂ for microalgal growth. By contrast, higher CO₂ gas flow rate (above 0.250 vvm) could supply sufficient CO₂ but it seem to be harmful for microalgae cells and inhibit their growth. This can be due to more acidic of culture (Chiswell et al., 1999; Vogel et al., 2010) and higher shear stress caused by higher

flow rate (Barbosa et al., 2003; Barbosa & Wijffels, 2004).

When aerated with different flow rates of CO₂ gas, *C. pyrenoidosa* culture was saturated with CO₂ after ca. 3 days. Fig. 5(b) showed that the pH of the cultures decreased from 7.50 to 7.00 with increases of CO₂ gas flow rate from 0.125 to 0.375 vvm. At 0.250 vvm of gas flow rate, the pH of culture was stable at 7.19 ± 0.06 , which is suitable for chlorophyll and protein synthesis. The maximum chlorophyll content (60.52 mg L⁻¹) of the *C. pyrenoidosa* was obtained (Fig. 5(c)), which would benefit the microalgal growth rate and biomass production promotion. In addition, a suitable CO₂ flow rate might enhance gas–liquid mass transfer (Sánchez Mirón et al., 2000) and replenish sufficient CO₂ for the microalgal growth. And a proper mixing of the culture would prevent the microalgae cells from sinking to the bottom of the bioreactor to make sure that all the microalgal cells could contact completely with nutrients, CO₂ and light. Ultimately, it improved microalgae biomass production. However, at a gas flow rate of 0.125 vvm, it could not supply enough CO₂ for microalgal growth. The content of energy storage component of microalgae biomass, such as lipids and carbohydrates was lower (Fig. 5(d)). However, the bubble coalescence phenomena was not serious, and the residence time of CO₂ bubbles was longer with a lower gas flow rate of 0.125 vvm. Therefore, microalgae showed CO₂ removal efficiency suspension of 17.21% (Table 2). By contrast, a higher gas flow rate would supply too much CO₂ and make culture a little acidification with a pH of ca. 6.90. Besides, the bubble carrying effect would also be enhanced at a higher gas flow rate. It would inhibit microalgal growth. It had reported such a stressful

condition would improve synthesis of energy storage component (Lin and Lin, 2011). Also in the present study, the acidulated condition improved the content of lipid and carbohydrate to 26.22% and 20.42%, respectively (Fig. 5(d)).

The effect of CO₂ concentrations on microalgae growth cultivated with the optimized gas distributor were also investigated (Fig. 6). Similarly to the effect of flow gas rate, lower concentration could not supply enough CO₂ for cells, and higher concentration of CO₂ would inhibit microalgal growth. The optimal CO₂ concentration was 5% for *C. pyrenoidosa* cultivated in the bioreactor with optimized gas distributor (Fig. 6(a)). It indicates that the microalgal growth was completely inhibited with 10% CO₂ (Chiu et al., 2008; Yoo et al., 2010). This is because high concentration of CO₂ could result in a lower pH of the culture (Fig. 6(b)), which would cause an activity decrease of ribulose 1,5-bisphosphate carboxylase-oxygenase, which is a key enzyme of photosynthesis (Miziorko & Lorimer, 1983), and decrease in chlorophyll synthesis (Fig. 6(c)). In addition, bubble carrying effect was also increased largely. Therefore, microalgae showed a lower of CO₂ removal efficiency (4.26%), photosynthetic carbon fixation rate ($724.49 \mu\text{mol m}^{-3} \text{s}^{-1}$) and biomass growth rate ($0.52 \text{ g L}^{-1} \text{d}^{-1}$) (Table 2) when cultivated with 10% CO₂. Besides, the lipid and carbohydrate content of microalgae showed an increasing trends with the CO₂ concentration increasing (Fig. 6(c)). This suggested that higher CO₂ concentration could improve the lipid content. Similar results were obtained by Abd El Baky et al. (2012) and Tang et al. (2011).

4. Conclusions

The gas distributor was optimized based on the effect of vent hole's diameter and arrangement on CO₂ bubble dynamic behaviors. When microalgae cultivated using gas distributor with a holes' diameter of 0.5 mm, spacing of 1.5 mm, microalgae showed largest growth rate and carbon sequestration ability. When aerating 5% CO₂ at 0.250 vvm with the optimized gas distributor into microalgal suspension, the maximum biomass concentration was up to 2.88 g L⁻¹ and maximum photosynthetic CO₂ fixation rate was 961.73 μmol m⁻³ s⁻¹. The results indicates bubbles dynamic behaviors indeed is an important theoretical guidance for bioreactor design and operating conditions optimization.

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Table 1. Effect of gas distributor size on the microalgae growth and CO₂ fixation cultivated with 5% CO₂ at gas flow rate of 0.250 vvm.

Diameter (mm)	Spacing (mm)	CO₂ removal efficiency (%)	Photosynthetic CO₂ fixation rate ($\mu\text{mol m}^{-3} \text{s}^{-1}$)	Biomass productivity ($\text{g L}^{-1} \text{d}^{-1}$)
0.3	2.5	7.63 $\pm$ 0.785	648.81 $\pm$ 66.752	0.46 $\pm$ 0.041
0.5		10.69 $\pm$ 0.926	909.01 $\pm$ 78.741	0.63 $\pm$ 0.023
1.0		8.31 $\pm$ 0.459	706.63 $\pm$ 39.031	0.51 $\pm$ 0.033
0.5	1.0	8.78 $\pm$ 0.678	746.60 $\pm$ 57.653	0.53 $\pm$ 0.027
	1.5	11.31 $\pm$ 1.327	961.73 $\pm$ 112.840	0.64 $\pm$ 0.024
	2.0	9.29 $\pm$ 0.399	789.97 $\pm$ 33.929	0.58 $\pm$ 0.023

Table 2. Effect of inlet CO₂ concentration and gas flow rate on the microalgae growth cultivated with a gas distributor (gas outlet diameter of 0.5 mm, spacing of 1.5 mm).

Gas flow rate (vvm)	CO ₂ conc. (%)	CO ₂ removal efficiency (%)	Photosynthetic CO ₂ fixation rate ($\mu\text{mol m}^{-3} \text{s}^{-1}$)	Biomass productivity ($\text{g L}^{-1} \text{d}^{-1}$)
0.125	5	17.21 $\pm$ 0.978	731.72 $\pm$ 41.581	0.53 $\pm$ 0.033
0.250		11.31 $\pm$ 1.327	961.73 $\pm$ 112.840	0.64 $\pm$ 0.024
0.375		6.56 $\pm$ 0.871	836.73 $\pm$ 111.095	0.6 $\pm$ 0.023
0.250	3	17.18 $\pm$ 1.815	876.53 $\pm$ 92.602	0.61 $\pm$ 0.026
	10	4.26 $\pm$ 0.716	724.49 $\pm$ 121.768	0.52 $\pm$ 0.040

vvm: unit of gas flow rate, which means 1 L min⁻¹ of gas to 1 L of culture.

Graphic Abstract