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Improved CO_2 fixation with *Oscillatoria* sp. in response to various supply frequencies of CO_2 supply

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ABSTRACT

Sequestration of CO_2 by microalgae is one of the technically viable options to control the CO_2 emission. CO_2 sequestration by microalgae through photosynthesis also results in large quantity of biomass production with considerable fuel properties. Different types of micro algal species have got CO_2 tolerance to a different level. This work focuses on the tolerance capability of *Oscillatoria* sp. for 100% CO_2, bubbled into the ASN III media. CO_2 supply was given at a flow rate of 300 ml/min for 15 min. 12:12 h of light to dark cycle was followed.CO_2 supply was varied from one to three times a day during the light period. An increasing trend of biomass growth was observed with increased supply frequency of CO_2. An increase of biomass yield of 44.7%, CO_2 fixation of 64.2% and calorific value of 17.6% was observed corresponding to the three times supply frequency of CO_2, compared with the control. Ultimate analysis, stoichiometric analysis and molecular weight of microalgae grown in control and pure CO_2 (100%) gas were compared. Growth of microalgae obtained at various supply frequencies of CO_2 were fitted with logistic model with a correlation coefficient of 0.98. The present studies indicated that the *Oscillatoria* sp. was able to sustain in pure CO_2(100%) gas and may be considered as the species for converting the emissions from the distilleries to energy.

1. Introduction

Increase in CO_2 concentration is the prominent cause of global warming, having massive threats on human life and deleterious effect on environment and climate. [1–4]. CO_2 contributes for 52% of global greenhouse gas emission [5] and it has reached 402.56 ppm in January 2016 with an average rise of every 2 ppm for the past five years [6]. CO_2 levels exceeding beyond 450 ppm in the atmosphere will have adverse effects on the global climate conditions, sea levels and survival of many organisms [7]. However, the CO_2 mitigation techniques such as chemical adsorption, solid adsorbents, membrane technology and cryogenic fractionation have safety and environmental drawbacks in capturing CO_2 from the atmosphere and flue gas [8]. Hence, researches on utilising microalgae for CO_2 sequestration and biofuel generation have gained profound attention among the researchers, policy makers and in various sectors [9,10]. Compared to conventional technologies, microalgal cultivation is considered as an environmental friendly technology [11]. and having less impact on food supply and agricultural land [12–14]. In addition to carbon sequestration, microalgae also act as an efficient renewable sources towards fossil fuel replacement inspite of it's current limitations [13,15–17]. Microalgae possess certain advantages over terrestrial plants in terms of rapid growth rate, morphology, photosynthetic efficiency and higher CO_2 fixation efficiency [18–20].

CO_2 is majorly emitted from industrial sector (point source) or from transport sector (line source). Control and treatment of point sources of pollution is easier compared to line sources of pollution. The major point sources of CO_2 include the flue gas (15–20% CO_2 concentration) emitted from the thermal power plants and pure CO_2 (100%) emitted from the chemical industries such as distilleries. Micro algal technology could be used as a possible solution to control point source CO_2 emissions from chemical industries and thermal power plants. Number of research works have been carried out for controlling flue gas emission using microalgae. Table 1. gives the tolerance capability of various species for CO_2 concentration.

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Table 1
Tolerance capability of various microalgal species for varying CO_2 concentration.

Microalgal species	% of CO_2 concentration	Inference	Reference
Anabaena. CH1	5,10 and 15	Maximum biomass concentration and CO_2 fixation rate was obtained at 10% concentration of CO_2 which is 1.16 g/l and 1.01 g/L/day.	[21]
Scenedesmus obliquus SA1 (KC733762)	0.03–35%	Maximum biomass concentration was reported at 35% concentration of CO_2 and is 1.39 ± 0.023 g/l. CO_2 fixation rate 97.65 ± 1.03 mg L^{-1} d^{-1} .	[22]
Dunaliella tertiolecta SAD-13.86, Chlorella vulgaris LEB-104, Spirulina platensis LEB-52 and Botryococcus braunii SAG-30.81	Air sparged with 5% CO_2	Among the four microalgal species, *Botryococcus braunii* SAG-30.81 produced CO_2 fixation rate of 497 mg/L/day along with biomass productivity of 3.11 g/L.	[23]
Scenedesmus obliquus SJTU-3 and Chlorella pyrenoidosa SJTU-2	0.03–50%	Maximum biomass productivity of 1.84 ± 0.01 g/L and CO_2 fixation rate of 50.58 ± 0.08 was obtained at 10% CO_2 concentration for *Scenedesmus obliquus SJTU-3* and it decreased further with increasing CO_2 concentration.	[24]
Chlorella sp.	0–70%	Maximum growth of 5.772 g/L was reported at 10% CO_2 concentration. It decreased further with increasing CO_2 concentration. Microalgae was able to sustain till 70% CO_2 of 0.766 g/L concentration	[25]
Nannochloropsis sp. and Phaeodactylum sp.	Actual flue gas 10–12% CO_2, SOx and NOx are 70–90 ppm. Simulated flue gas only with 12% CO_2 and desulfured gas	Microalgae is able to sustain in the flue gas as equal to that of 12% CO_2 concentration in simulated flue gas.	[26]
Spirulina platensis	12% Actual desulfured flue gas from a power plant smokestack	Dry biomass of 1220Kg/year was achieved and it was 2.35 g/L.	[27]
Anabaena sp.	Flue gas	Maximum CO_2 fixation rate of 3.0 gCO_2 L^{-1} day^{-1} was observed in outdoors	[28]
Chlorella sp. MTF-15	Coke oven gas CO_2- 23–27% Hot Stove CO_2- 24–28% Power plant CO_2- 22–26%	Maximum biomass concentration of 0.515, 0.314 and 0.342 g/L/day for coke oven gas, hot stove and power plant respectively.	[29]
Spirulina platensis	Simulated diesel generator exhaust CO_2 11%; CO 1.5%	Maximum biomass concentration of 1.85 g/L was obtained on the sixth day.	[30]
Nannochloropsis sp. *Chlorella* sp. *Pseudochlorococcum* sp.	0.04–2% (v/v)	Maximum biomass concentration is 0.87 ± 0.05 g/L/day and CO_2 fixation rate is 1.538 g/L/day for Chlorella sp. in nitrogen sufficient conditions for *Chlorella* sp.in nitrogen sufficient conditions.	[31]
Chlorella sp. *Isochrysis* sp. *Amphidinium carterae*	10, 15 and 20% of flue gas CO_2 concentration	Maximum biomass productivity of 0.268 g/L/day and maximum CO_2 fixation rate of 0.492 g/l/day for *Chlorella* sp.	[32]
Synechococcus elongatus	Ambient air, 5% and 10% CO_2 concentration	Maximum biomass concentration is 2.1 g/L at 10% CO_2 concentration for membrane contactor PBR and 2.45 g/L at 5% CO_2 concentration for membrane sparger PBR.	[33]

The quantum of research works available on the tolerance capability of microalgal strains for pure CO_2 in to the medium is very less [25,34–37]. Other than the CO_2 concentration, a number of process parameters and environmental conditions such as, initial inoculum concentration [38], nutrients [39–41], light intensity [38,40], light to dark cycle ratio [38], pH [2,42,43], temperature [44] have influence on the microalgal growth and subsequently on secondary metabolites produced.

Diffusion capablitity of CO_2 in water is 10,000 times slower than in air [45] and the solubility of CO_2 in water is also very less [46]. Hence when CO_2 is bubbled into the medium, there is a possibility of rapid release of CO_2 present in water to the atmosphere without significant biological utilization. Also during continuous supply of CO_2, pH is maintained at 5.5 which is highly unfavourable for microalgal growth. Due to above limitations continuous supply of CO2 into the system is not favourable for microalgal growth. Hence to improve the utilization of CO_2, it is purged in to the system intermittently having various supply frequency. This strategy may result in potential CO_2 mitigation through biological sequestration, by monitoring the availability of CO_2 in the medium. Also CO_2 is fixed by Ribulose −1,5-bisphosphate carboxylase oxygenase (Rubisco) through Calvin Cycle by the conversion of CO_2 to organic carbon. The maximum catalytic capacity of Rubisco can be achieved only when it is fully saturated with the substrate of CO_2 and ribulose-1,5-bisphosphate. Perhaps ambient CO_2 imparts only 25% catalytic capacity of Rubisco which may reduce the photosynthetic efficiency [22]. Hence to maintain the Rubsico in saturated form throughout the growth phase of the microbial system and to improve the CO_2 utilization in the medium, CO_2 could be sparged intermittently which might eventually increase the photosynthetic efficiency.

Also, addition of CO_2 into the medium is responsible for facilitating the exposure of cells to light and thereby minimizing the shading effect and avoids the dissolved oxygen accumulation in the system to reduce the toxic effects [47]. However, such addition may lead to the alteration of pH affecting the growth process. Hence, more studies are required to find out the growth rate and sustainability of species suitable for 100% CO_2 gas. To enhance the catalytic property of Rubisco enzyme, this study involves the growth and sustainability of *Oscillatoria* sp. supplied with pure CO_2 at various intervals and further to characterize the cultivated biomass under such conditions.

2. Materials and methods

2.1. Species and operating parameters

Oscillatoria sp. (Strain No: BDU 142191) was collected from the National Facility for Marine Cyanobacteria, Bharathidasan University, Tamil nadu. *Oscillatoria* sp. was cultivated in flat plate reactor