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Application of artificial neural network and particle swarm optimization for modeling and optimization of CO_2 sequestration using microalgae

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In the present study, *Chlorococcum* sp., a green alga – collected from coke-oven effluent treatment plant, was applied for phyco-sequestration of carbon dioxide at pH (7–11), IS (inoculum sizes: 5–12.5%), and CO_2 concentrations (5–30%), respectively. The test strain remained viable upto highest tested concentration. However, the maximum CO_2 sequestration was found to be 79.08±0.89% at lowest tested CO_2, pH 9 and IS 12.5%. ANN (Artificial Neural Network) modeling technique was attempted to envisage the CO_2 phyco-sequestration at prevailing operating conditions. Various algorithm of ANN and lot of characteristics functions were tried to get best model for this experimental data. Developed ANN model performance was found very efficient. PSO (Particle Swarm Optimization) was done to assess the optimized input parameters to maximize phyco-sequestration of CO_2. With PSO algorithm the optimum input parameters are obtained to achieve 99% removal of CO_2. As far as known the studies on application of ANN and PSO for modeling and optimization of CO_2 phyco-sequestration using microalgae are very rare and the novelty of the present work lies here.

Keywords: CO_2 sequestration, green alga, macromolecules, FAME analysis, ANN.

Introduction

CO_2 is a considerable contributor to the global warming and climate change[1]. Fossil fuel fired power-plants are the major sources to atmospheric CO_2[2]. Flue gas from power plants has a temperature of 120°C and CO_2 in the range of 10–15%[3]. Several methods have been tried for the fixation of CO_2, among them phyco-sequestration using cyanobacteria/green algae has been seen as one of the effective steps[4]. Cultivation of algae has been exploited as a supplementary move for flue gas treatment, for the lowering of CO_2 concentration in the exhaust flue gas[5].

Microalgae in suspension culture are comparatively fast growing and higher biofixing agent than terrestrial plants[6]. The microalgal biomass associated with considerable concentrations of biomolecules such as carbohydrates, lipids, proteins and some costly bio components including pigments and vitamins, which can be utilized as an active constituent in pharmacy, supplements of feed and preservatives of food or in the production of biofuels[5]. Further, though a significant number of investigation was performed on phyco-sequestration of CO_2 using microalgae, an inclusive study including thorough experimentation, data driven AI (artificial-intelligence) based mathematical modeling and finding out optimized input parameters for highest phyco-sequestration of CO_2 is quite small.

Since application of ANN (artificial neural network) and PSO (particle swarm optimization) technique in biological system is very sparse, in the present article, both of these techniques have been used for modeling and optimizing the phyco-sequestration of CO_2 using a green alga of *Chlorococcum* sp. Parametric study has been done to see the effect of different working parameters such pH, concentration of CO_2, etc. The data have been used in ANN and finally, PSO is used to get optimum values of parameters for maximum biofixation of CO_2. The studies on application of ANN and PSO for modeling and optimization of CO_2 seques-

tration using microalgae are very few and uniqueness of the present work lies here.

Results and discussion

Growth and tolerance concentration assessment for the test strain

Biomass, carbohydrate, lipid, and protein were estimated during growth of the isolated green algal strain in BG-11 (Fig. 1a). The concentrations increased as 4.9 fold (biomass), 5.4 fold (lipid), 7.4 fold (protein) and 9 fold (carbohydrate) with respect to those concentrations on day 2 for *Chlorococcum* sp. One of the probable reasons for the proliferation is because of the presence of high amount of N-source and P-source in BG-11[7]. From literature it is seen that phosphorous plays the critical role in proliferation and metabolism of microalgae[8]. *Chlorococcum* sp. has shown growth upto 50% CO_2 concentration, however, the concentration of biomass is higher (0.315±0.0014 g/L) upto 10% CO_2 than that of the control (0.279±0.002 g/L) (Fig. 1b). The biomass concentration decreases (0.215±0.001 g/L) at and beyond 20% CO_2.

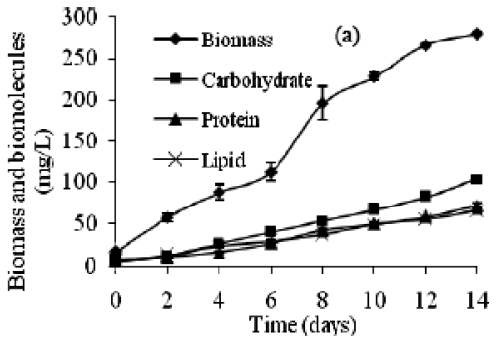

Fig. 1a. Growth of biomass and biomolecules of *Chlorococcum* sp. in BG-11 medium.

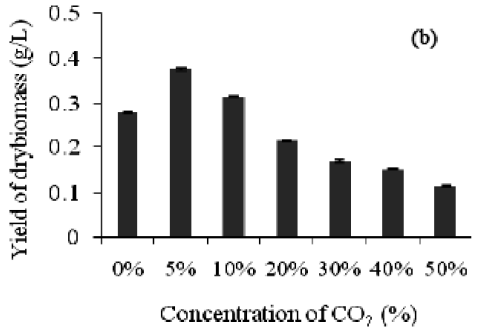

Fig. 1b. Lethal dose analysis of *Chlorococcum* sp. in BG-11 medium.

The biomass concentration at 5 and 10% CO_2 is 1.34 and 1.13 times higher than that of control. Anjos *et al.*[6] reported the biomass growth inhibition at higher CO_2 concentrations.

Optimization of input parameters during CO_2 sequestration

Effect of pH on sequestration of CO_2:

From the observed results, it is clear that pH variations from 7 to 9 has direct positive effect on the phyco-sequestration of CO_2. The phyco-sequestration of CO_2 using *Chlorococcum* sp. has been found to be 45.79±0.89%, 73.28±1.34% and 50.81±0.45% at 10% inoculum size, 5% CO_2, at pH 7, 9 and 11, respectively, after specified time of incubation (14 days) (Fig. 2a).

The maximum productivities (mg/L/day) of biomolecules and biomass such during optimization of input parameters were found as 25.07±0.25 (biomass), 6.99±0.12 (lipid), 7.5±0.15 (protein) and 9.16±0.12 (carbohydrate) for green algae at inoculum size of 10% and optimum pH (9) (Fig. 2b).

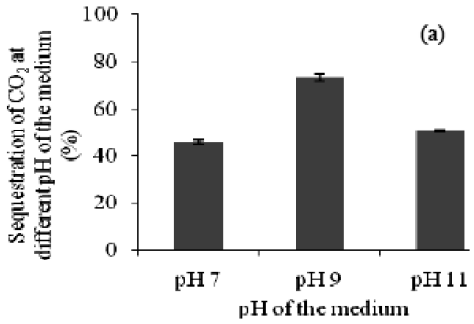

Fig. 2a. Sequestration of CO_2 using *Chlorococcum* sp. at different initial pH of the medium.

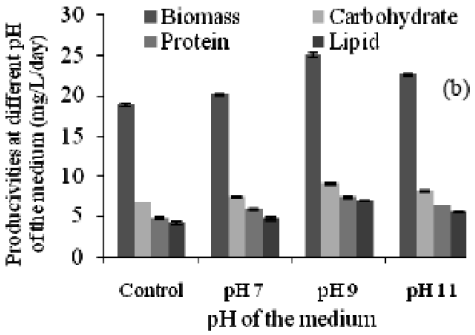

Fig. 2b. Productivities of biomass and biomolecules of *Chlorococcum* sp. at different initial pH of the medium.

Again pH beyond 9 the productivities decreased. It was also observed that the productivities obtained at dissimilar pH conditions supplemented with CO_2 are found to be maximum than that of control. The pH of the nutrient medium plays a significant role in growth of the microalgae species, as it has significant effect on the activity of diverse enzymes.

Effect of CO_2 concentration:

The maximum sequestration of CO_2 has been found to be 73.27±1.34%, 53.32±0.89%, 36.87±0.60%, 25.71± 0.34% and 14.01±0.17% for *Chlorococcum* sp. for the initial concentrations of CO_2 as 5%, 10%, 15%, 20% and 30%, respectively, after specified incubation period (14 days) (Fig. 3a). All other parameters have been maintained constant at pH 9 and 10% inoculum size. It is obvious that maximum sequestrations are estimated at lesser CO_2 concentration for the test strain.

The productivities of biomass and macromolecules at various CO_2 concentrations have been found to be higher than control upto 10% CO_2 for the green algae (Fig. 3b). The maximum productivities of 25.07±0.25 mg/L/day (biomass), 6.99±0.12 mg/L/day (lipid), 7.5±0.15 mg/L/day (protein) and 9.16±0.12 mg/L/day (carbohydrate) were found at 5% CO_2, optimum pH (9) and IS of 10% and it is comparable with the green alga *Chlorococcum littorale*[9]. Also, the productivities of macromolecules at 15, 20 and 30% CO_2 are less than that of control. Though the phyco-sequestration of CO_2 is highest at 5% CO_2, the escalation in productivities are 2.06 fold (biomass), 2.01 (lipid), 1.47 (protein) and 1.49 fold (carbohydrate) at 10% CO_2. Lower concentration of CO_3^{2-} (1.3 g/L) at lowest tested CO_2 concentration may lead to lower productivities. From the study of Lam *et al.*[8], it can stated that *Chlorella* sp. growth rate was negatively affected above 2% (v/v) CO_2. Several scientists reported that above 5% (v/v) CO_2 concentration caused cell death and ultimately growth was inhibited for microalgae[10].

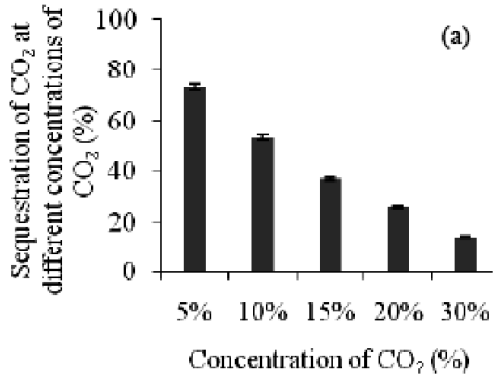

Fig. 3a. Sequestration of CO_2 using *Chlorococcum* sp. at different CO_2 concentrations.

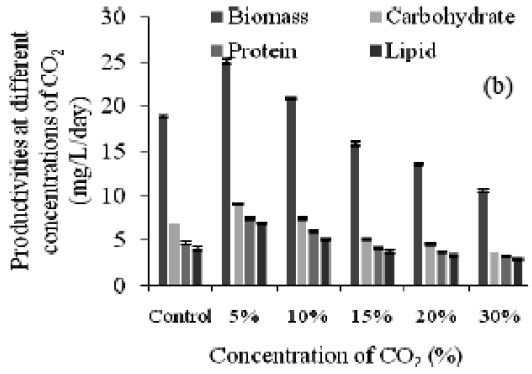

Fig. 3b. Productivities of biomass and biomolecules of *Chlorococcum* sp. at different CO_2 concentrations.

Effect of inoculum size:

The maximum CO_2 sequestration has been observed as 53.52±1.78%, 73.27±1.34% and 79.08±0.89% for the green alga when the inoculum sizes are at 5%, 10% and 12.5%, respectively (Fig. 4a). Other parameters have been set as pH 9, 5% CO_2. It is obvious that with raise in inoculum size, the number of microalgal cells also increases which eventually leads to the high sequestration of CO_2. Therefore, the productivities of biomass and macromolecules are maximum when the inoculum size is maximum (12.5%). The utmost productivities of biomass (30.54±0.20 mg/L/day), lipid (7.4±0.18 mg/L/day), protein (8.84±0.05 mg/L/day) and carbohydrate (11.95±0.10 mg/L/day) have been found at 12.5% inoculum size, pH 9 and 5% CO_2 for the test strain (Fig. 4b). These productivities are higher than the productivities of *Chlorococcum* sp. when grown in CHU-10 medium[11]. Researchers reported that the CO_2 phyco-sequestration, cell mass and biomolecule concentration changed for variation of IS[12]. Though the sequestration of CO_2 and productivities were found to be maximum at 12.5% inoculum size, effective inoculum size was selected as 10%, since the

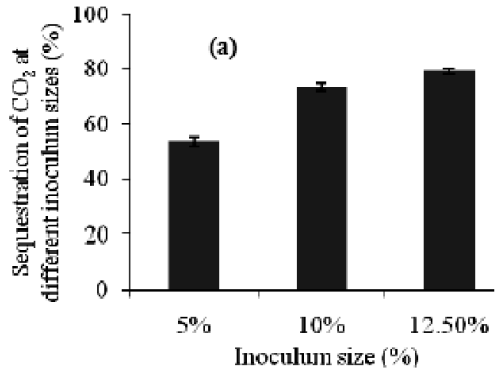

Fig. 4a. Sequestration of CO_2 using *Chlorococcum* sp. at different inoculum sizes.

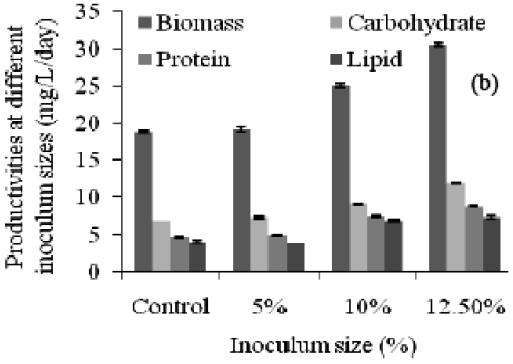

Fig. 4b. Productivities of biomass and biomolecules of *Chlorococcum* sp. at different inoculum sizes.

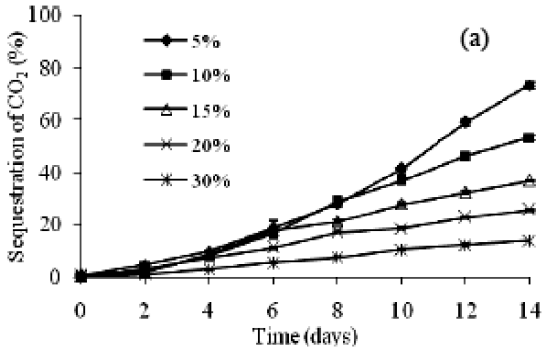

Fig. 5a. Sequestration of CO_2 using *Chlorococcum* sp. at different CO_2 concentrations.

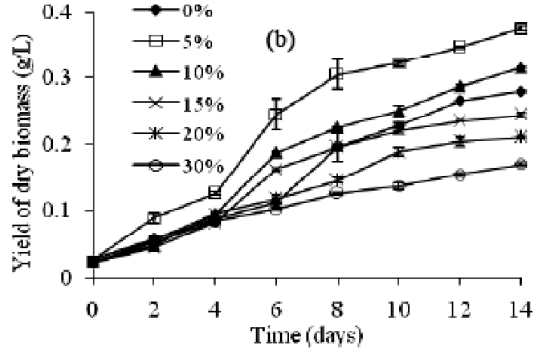

Fig. 5b. Yield of dry biomass of *Chlorococcum* sp. at different CO_2 concentrations.

productivities were comparatively same at 10% and 12.5% inoculum size.

Time variation of CO_2 sequestration using microalgae

Time variation study of CO_2 sequestration has been done at different CO_2 concentration (5–30%) at pH 9 and IS (10%) for 14 days. The CO_2 sequestration decreases from 73.27±1.34 to 14.01±0.17% for change of CO_2 concentration in above mentioned range (Fig. 5a). Lower CO_3^{2-} content (1.3 g/L) at 5% CO_2 may cause maximum sequestration.

The concentration of biomass was proliferated from 0.28±0.002 to 0.315±0.001 g/L for change in CO_2 concentration from 0 to 10% (Fig. 5b). The biomass concentration has been decreased to 0.17±0.003 g/L with additional increase in CO_2 concentration to 30%. However, the maximum concentration of biomass (0.376±0.0014 g/L) has been observed at 5% CO_2. The reduction in biomass growth has been observed for the test strain supplemented at elevated CO_2 concentrations (15%). Researchers reported that the CO_2 sequestration and microalgal growth was negatively affected at high values of CO_2[5]. When microalgae proliferated at high values of CO_2 (15% or more), several microalgae showed lower uptake of CO_2, higher photosynthetic warmth to O_2 and lower functionality of the enzyme CA, responsible for photo-synthetic exploitation of inorganic carbon source[13]. As per previous reports the highest biomass production of *Chlorella* sp. was found at 15% CO_2 under high inoculum[14]. However, in the current study, the test strain showed biomass concentration of 0.25±0.003 g/L of at 15% CO_2. The CO_2 phyco-fixation and cell mass production depend on the uniqueness of microalgal species and culture conditions[15].

Evaluation of macromolecules content:

For the present strain, the lipid concentration increases

(65.71±3.3–75.7±1.7 mg/L) when the CO_2 changes (0–10%) and decreases to 45.72±1.6 mg/L with additional increase in CO_2 concentration to highest value. However, the maximum concentration of lipid has been found as 104.28± mg/L at 5% CO_2, pH 9 and 10% inoculum size (Fig. 5c).

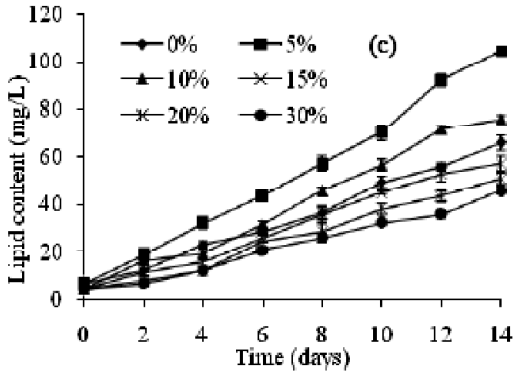

Fig. 5c. Lipid content of *Chlorococcum* sp. at different CO_2 concentrations.

The carbohydrate and protein concentrations follow the same trend as that of biomass and lipid content under identical condition. The protein content increases from 71.87±2.2 to 113.75±2.2 mg/L with increase in CO_2 concentration from 0 to 5% for green alga (Fig. 5d). The concentration of protein decreases to 50.63±0.72 with further increase in CO_2 to 30% for the test strain. The maximum content of carbohydrate has been measured as 137±1.7 mg/L at 5% CO_2 for green alga (Fig. 5e) after specified incubation period (14 days). Again, increase in concentration of CO_2 from 15 to 30%, the carbohydrate value decreases.

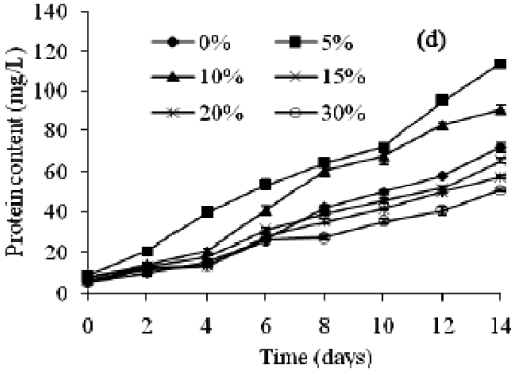

Fig. 5d. Protein content of *Chlorococcum* sp. at different CO_2 concentrations.

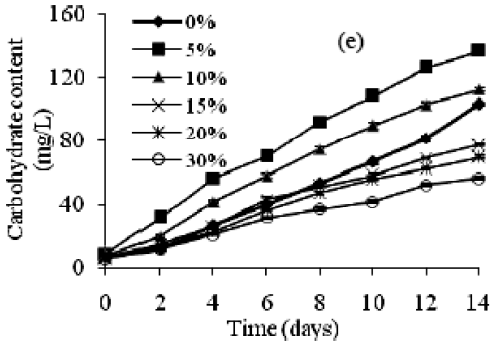

Fig. 5e. Carbohydrate content of *Chlorococcum* sp. at different CO_2 concentrations.

FAME analysis

The fatty acid contents suitable for the production of biodiesel (C16:0) have been found as 43.26% and 46.64% for *Chlorococcum* sp. before and after sequestration of CO_2, respectively (Figures not shown). These amounts have been found to be higher than that of *Scenedesmus* sp. at 15% CO_2[16]. The fatty acid contents (C16:0) for *C. vulgaris*, *B. terribilis* and *B. braunii* were found to be 34.82%, 35.46% and 30.39%, respectively, which were lower than that of the present test strain under diverse CO_2 concentrations[17].

Development of a suitable ANN model and optimization using PSO model

ANN optimization:

Experimental data has been employed in training of artificial neural network for CO_2 phyco-sequestration (Data not shown). The model is as follows; total data: 133, optimum node: 15, activation function: Sigmoidal (Hidden layer) and linear (output), training algorithm: Levenberg-Marquardt and prediction accuracy of R^2 (0.999) and very low MSE (1.18). These values suggest the good fitting of ANN model predicted data to the experimental ones. Thus, the suitability of proposed ANN is established.

PSO optimization:

Maximum and minimum input factors are chosen as: medium pH [11, 7], concentration of CO_2 [30%, 5%], IS [12.5%, 5%] and time [14 days, 2 days], respectively. The values of four input parameters will be assumed in these ranges and PSO passes these values to ANN-trained model to get optimum values.

Particle swarm optimization keeps on changing until CO_2 removal (%) is maximized[18]. Five sets of optimum input parameters are obtained using PSO algorithm to achieve 99% removal (Table 1). The advantage of PSO algorithm lies in the fact that one can get optimum values by performing minimum number of experimentations.

Table 1. Multiple optimum responses and corresponding operating parameter obtained by Particle Swarm Optimization (PSO) for the removal of CO_2 using *Chlorococcum* sp.

Sl. No.	Time (days)	Initial concentration (%)	Inoculum size (%)	pH	Sequestration of CO_2 (%)
1.	13.99	5.04	12.50	11.00	100.00
2.	13.99	5.00	12.50	11.00	100.00
3.	14	5.00	12.50	10.99	100.00
4.	13.99	5.00	12.50	10.99	100.00
5.	14	5.004	12.50	11.00	100.00

Experimental

Growth and tolerance concentration investigation for the isolated sample

The growth of the test strain was studied in BG-11 medium ($NaNO_3$: 1.5 g/L, K_2HPO_4: 0.04 g/L, $MgSO_4.7H_2O$: 0.075 g/L, $CaCl_2.2H_2O$: 0.036 g/L, citric acid: 0.006 g/L, ferric ammonium citrate: 0.006 g/L, Na_2CO_3: 0.02 g/L, EDTA: 0.001 g/L, trace metal: 1 mL/L)[19]. The test strain was cultivated in different conical (Erlenmeyer) flasks. Finally, the flasks were placed in microalgal incubator at specified conditions. The lipid, protein and carbohydrate were extracted from cell mass using standard methods[19]. The tolerance concentration analysis was performed to find out the tolerance level of the test strain under high CO_2 level. The specific concentrations of CO_2 were prepared by dissolving the sodium carbonate salt in BG-11 equivalently using stoichiometric relations[12].

$$\log [HCO_3^-] = pH - \log [CO_2] - 6.30 \quad (1)$$

$$\log [CO_3^{2-}] = pH - \log [HCO_3^-] - 10.09 \quad (2)$$

At pH 8, the values of $NaCO_3$ equivalent to 5%, 10%, 20%, 30%, 40% and 50% CO_2 in the BG-11 medium are 10.9 g/L, 21.8 g/L, 43.6 g/L, 65.5 g/L, 87.3 g/L and 109.2 g/L, respectively. The isolated strain was cultivated at different CO_2 concentrations separately for the growth of strain. The production of biomass was investigated for specified incubation time of period (14 days), the preferred CO_2 concentration were selected on the basis of produced biomass for isolated strain.

Determination of optimum values of input factors during CO_2 phyco-sequestration

Phyco-sequestration of CO_2 was done by one factor at a time analysis (OFAT) as described by Upendar *et al.*[20]. Initially, medium pH (7-11) was varied and all other factors such as concentration of CO_2 and IS were set invariable at 5% and 10%, respectively. Next, the effect of initial concentration of CO_2 (5–30%) was checked. Finally, inoculum size was varied from 5–12.5% keeping other variables at their adequate values. For the analysis of biomass and residual concentrations of carbonate, samples were separated from the grown culture. All the adequate values of initial CO_2 concentration, inoculum size and pH of the medium were found on the basis of biofixation of CO_2 and biomass growth. The harvested biomass after CO_2 biofixation was utilised for macromolecules assessments. The biomass productivities were calculated using eq. (3)[21]:

$$P = \frac{(x_1 - x_0)}{(\theta_1 - \theta_0)} \quad (3)$$

where, $\theta_0 = 0^{th}$ day (days), θ_1 = beginning of stationary phase (days), x_0 = biomass and biomolecules concentration on 0^{th} day (mg/L), x_1 = biomass and biomolecules concentration after log phase (mg/L).

Time variation of CO_2 biofixation using test strain

Time variation of phyco-sequestration of CO_2 was investigated at several CO_2 values (5–30%). All other factors were fixed at their most suitable values i.e. the conditions at which the sequestration of CO_2 and concentration of biomass and biomolecules were found to be maximum. The solutions were cultivated for specific time period (14 days) using the isolated strain. Harvested biomass was used for estimation of macromolecules as described earlier section.

FAME analysis

Fatty acid methyl esters (FAMEs) are the molecules that are principally present in biodiesel. FAME analysis reveals that the fraction of lipid content of the test strain can directly

be used as biodiesel production. Total lipid content was extracted from dry biomass before and after biofixation of CO_2. The total lipid extracted and its fractions such as phospholipids (PL), glycolipids (GL) and neutral lipids (NL) were transesterified into FAMEs as described by Mistry *et al.*[11].

Development of a suitable ANN model and optimization using PSO model

Development of ANN model:

Experiments were carried out to validate CO_2 sequestration using test strain. The four factors were chosen which affect phyco-sequestration of CO_2: (i) time of contact, (ii) initial CO_2 concentration, (iii) pH, (iv) inoculum size. Percentage of removal of CO_2 is selected as response. For development of ANN model, the algorithm as discussed by Upendar *et al.*[18] was followed. The topology consists of four input nodes and one output node. Best ANN algorithm was selected during building time.

The aptness of ANN model is examined by MSE and coefficient of determination (R^2). Less than one hour was required for calculation of this large number of ANN models using intel i-5 processor.

Development of adequate particle swarm optimization model:

Once a dependable ANN model was built up, the optimization algorithm is attempted for optimizing the input factors to obtain maximum phyco-sequestration of CO_2. ANN algorithms are complex and consists of a number of exponential terms and weights, conventional optimization strategy is not compatible to ANN model. For development of PSO algorithm the steps of Upendar *et al.*[18] were followed to maximize CO_2 sequestration.

Conclusion

CO_2 mitigation using *Chlorococcum* sp. has been observed to be an effective alternative for CO_2 fixation with higher biomass yield. The maximum biofixation of CO_2 has been shown as 79.08±0.89% at 5% CO_2, IS of 12.5% and at optimum pH (9). The algal strain could remain alive upto 30% CO_2. Besides the bio-fixation of CO_2, macromolecules have also been produced from the spent cell-mass. Several ANN model with different algorithms were tested to predict phyco-sequestration of CO_2. High R^2 value and very low RMSE of artificial neural network model signifies the efficacy of proposed model.

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References

1. N. M. Langely, S. T. L. Harrison and R. P. van Hille, *Biochem. Eng. J*., 2012, **68**, 70.
2. D. A. Roberts, N. A. Paul, M. I. Bird and R. de Nys, *J. Environ. Manage*., 2015, **153**, 25.
3. G. Yadav, A. Karemore, S. K. Dash and R. Sen, *Bioresour. Technol*., 2015, **191**, 399.
4. B. Wang, Y. Li, N. Wu and C. Q. Lan, *Appl. Microbiol. Biotechnol*., 2008, **79(5)**, 707.
5. M. Anjos, B. D. Fernandes, A. A. Vicente, J. A. Teixeira and G. Dragone, *Bioresour. Technol*., 2013, **139**, 149.
6. P. Chelf, L. M. Brown and C. E. Wyman, *Biomass & Bioenergy*, 1993, **4(3)**, 175.
7. A. Goli, A. Shamiri, A. Talaiekhozani, N. Eshtiaghi, N. Aghamohammadi and M. K. Aroua, *J. Environ. Manage*., 2016, **183**, 41.
8. M. K. Lam, K. T. Lee and A. R. Mohamed, *Int. J. Greenh. Gas Cont*., 2012, **10**, 456.
9. S. P. Singh and P. Singh, *Renew. Sustain. Energ. Rev*., 2014, **38**, 172.
10. R. Ramanan, K. Kannan, A. Deshkar, R. Yadav and T. Chakrabarti, *Bioresour. Technol*., 2010, **101(8)**, 2616.
11. A. N. Mistry, G. Upendar, S. Singh, J. Chakrabarty, G. Bandyopadhyay, K. C. Ghanta and S. Dutta, *Sep. Sci. Technol*., 2020, **55(2)**, 332.
12. G. Upendar, A. N. Mistry, R. Das, S. G. Thakurata, J. Chakrabarty, K. C. Ghanta and S. Dutta, *Environ. Prog. Sustain. Energy*, 2018, **37(5)**, 1594.
13. J.-R. Xia and K.-S. Gao, *J. Integr. Plant. Biol*., 2005, **47**, 668.
14. S. Y. Chiu, C. Y. Kao, C. H. Chen, T. C. Kuan, S. C. Ong and C. S. Lin, *Bioresour. Technol*., 2008, **99(9)**, 3389.
15. U. B. Singh and A. Ahluwalia, *Mitig. Adapt. Strat. GL*, 2013, **18(1)**, 73.
16. R. Tripathi, J. Singh and I. S. Thakur, *Renew. Energ*., 2015, **74**, 774.
17. I. A. Nascimento, I. T. D. Cabanelas, J. N. dos Santos, M. A. Nascimento, L. Sousa and G. Sansone, *Algal. Res*.,

2015, **8**, 53.

18. G. Upendar, S. Singh, J. Chakrabarty, K. C. Ghanta, Md. Shahnawaz, S. K. Lahiri and S. Dutta, *Water Environ. J*., 2020, DOI:10.1111/wej.12646.
19. K. Anjana, A. Kaushik, B. Kiran and R. Nisha, *J. Hazard. Mater*., 2007, **148(1-2)**, 383.
20. G. Upendar, S. Singh, J. Chakrabarty, K. C. Ghanta, S. Dutta and A. Dutta, *J. Environ. Manage*., 2018, **218**, 234.
21. D. Tang, W. Han, P. Li, X. Miao and J. Zhong, *Bioresour. Technol*., 2011, **102(3)**, 3071.