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Role Of CO_2 On Algal Biomass Production In Diluted Wastewater

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Abstract: Presently wastewater treatment is the only economically viable way to produce algal biomass for conversion to energy such as biofuels with minimum environmental impact. Increasing industrialization and urbanization large amount of wastewater is generated which create a very serious problem for human and other animal health. The aim of this study was to produce algal biomass using waste water & its high level of nutrients and CO_2 through aeration. Waste water was collected and analyzed for pH, dissolved oxygen (DO), Biochemical oxygen demand (BOD), Chemical oxygen demand (COD), Nitrate (NO_3^-), Phosphate (PO_4^{--}), Ammonia (NH_3). The algal biomass was increased simultaneously with increasing CO_2 consumption rate. The maximum algal biomass was recorded in 8 times diluted waste water at one, two and three time aeration on 21st day is 14.1±0.71 g/l, 18.6±0.55g/l and 33.2±1.2 g/l respectively.Cultivating algae on nitrogen (N) and phosphorus (P) in animal manure effluents presents an alternative to the current practice of land application. Finally, cost effective algal biomass used as a source of energy.

Keywords: Wastewater, Algal biomass, Biofuels, Nutrients and CO_2.

Introduction

Water pollution is a major problem in the global context. Increasing industrialization and urbanization large amount of wastewater is generated which create a very serious problem for human and other animal health. High concentration ofnutrients such as phosphorous in sewage effluents are largely due to the increasing use of domestic synthetic detergents during the last decades, which have increased the phosphorus content of sewage to 3-4 times the level before the advent of detergents[1].Phosphorus (P) is a key nutrient that stimulates the growth of algae and other photosynthetic microorganisms such as toxic cyanobacteria (blue-green algae), and must be removed from wastewater to avoid eutrophication in aquatic water systems[2].The risk of adverse effects to the plant and animal communities in waterways declines as P concentrations approach background levels [3]. Eutrophication is one of the main water quality problems throughout the world[4].The role of nutrient in water is harmful but on land it is useful, that is used as a fertilizer. Algae growing in wastewater treatment assimilate nutrients and thus subsequent harvest of the algal biomass recovers the nutrients from the wastewater[5,6,7]. Algae growth in HRAPs has also been shown to achieve greater than 75% nutrient removal[8,9]. The actual magnitude of assimilation in algae for biomass production at any moment is determined by one or other of the maincontrolling conditions i.e. nutrient status, CO_2, light, or temperature, acting as a limiting factor. In aquatic systems, CO_2 is usually limiting, except perhaps in deep water where sediment decomposition provides CO_2 but light levels are low [10,11]. The removal of nutrients from waste water is called as waste water treatment and in this process different types of micro and macrophytes are used. If the provide suitable conditions in the pond, so a good amount of algal biomass is generated and used as different types of materials such as biofuel/ bioenergy, biofertilzers, alcohols and single cell protein .

Material and Methods

Collection of wastewater and its characteristics:

The nutrient rich wastewater sample was collected from Laxmi Tal pond in Jhansi Uttar Pradesh, India and they stored in refrigerator at 4°C temperature. The physicochemical characteristic of initial and after treatment of cultured wastewater was determined (Table-1&2) as per the standard procedure given in the Standard Method for the Examination of Water and Wastewater [12].

Selection of Experimental Site:

In order to provide identical environmental conditions open field in the research garden with 10 hours of light period was selected. All the culture containers were put inside the wire net chamber to protect culture experiment from any external injuries/disturbance. At the time of disturbance in local weather wire net chamber with culture containers was covered with jute sheets to avoid dust fall related contamination. To avoid frequent change in temperature of culture experiments, all the culture containers were put inside the ground in such a way that the level of the culture medium maintained at the same level as the ground of the experimental field. The ground soil was kept moist periodically to avoid excess heating by sun.

Selection of culture container and culture period:

According to the availability plastic bucket 10 litres (30cm diameter x 10 cm height)sizes were selected for the algal biomass production.After the pilot study, the experiment is setup for 21 days in 8 times dilution with addition of tap water (less nutrient) for maximum biomass production, hence all the culture container with the triplicates were setup in the selected experimental Site under observation for 21 days.

Algal Biomass estimation:

Waste water sample with algal biomass were taken for biomass estimation at different periods. Biomass estimated byusing dry weight method. First a required quantity of waste water with algal biomass was transferred into 50 ml centrifuge tube after thorough washing with distilled water (washing of clay particles). Then centrifuge tube with the sample was centrifuged for half an hour and the supernatant waters was removed followed by transferring dense biomass into preweighedaluminum plate and dried to a constant weight at 70^0C temperature in an oven and weighed for dry biomass.

Aeration procedure for increasing Carbon dioxide concentration in wastewater:

Atmospheric air was injected into culture wastewater samples in three different rates. At each time aerations were continued for 30 minutes. First time aeration of a day was started at morning time. Second dose of aeration (2 times aeration) was performed with a gap of 3 hours then the first time aeration. Similarly, three time aeration were done with a gap of 3 hours between two aeration treatments.The carbon dioxide helps in the increase in the biomass production so provide CO_2 manually for half hours before sunrise, after three hours and after six hours.

Carbon Dioxide Estimation and Maintenance of Water Level:

Before and after aeration of each treatment of carbon dioxide sample was collected by means of a syringe (100ml) and used for analysis of carbon dioxide concentration as per the standard procedure. Water loss by evapotranspiration during the culture study was replaced by single distilled water. Each sampling for analysis was conducted just after maintenance of water level as previously marked in the culture.

Procedure for Measurement of Temperature:

One extra culturecontainer with a thermometer (dipped inside) was kept under observation for measurement temperature during period of culture experiment. The minimum temperature of day was recorded just before sunrise. Similarly, the maximum temperature of a day was recorded at 2.00pm regularly (Table 3).

Results and Discussion

Physicochemical Properties of Wastewater:

Table-1: Initial physicochemical properties of wastewater used for study of algal biomass production

Parameters	Wastewater alone (control)	Tap water	8 Times Diluted Wastewater#
Colour	**Brownish**	**Transparent**	**Slightly Brownish**
pH	9.8±0.5	7.9±0.3	8.6±0.6
BOD (mg/l)	85.5±1.5	1.8±0.5	12.1±2.1
COD (mg/l)	432.0±1.9	5.9±1.6	59.9±2.5
DO (mg/l)	2.9±1.2	5.4±0.9	3.9±1.4
Dissolved CO_2 (mg/l)	12.0±2.1	7.8±0.3	1.7±1.8
Total P (mg/l)	20.1±1.7	0.2±.01	2.5±0.6
NH3-N (mg/l)	43.2±1.8	0.1±0.06	5.5±2.9

#: Reported values (except pH) are calculated on the basis of dilution factor

Table-2: Properties of treated wastewater on 21st day of algal biomass production

Parameters	Control culture	8 Times diluted culture		
		One time aeration	**Two times aeration**	**Three times aeration**
Colour	**Brownish**	**Slightly Brownish**	**Slightly Transparent**	**Transparent**
pH	8.7±0.2	8.2±0.5	7.9±0.3	7.7±0.6
BOD (mg/l)	76.95±1.6	2.7±0.9	4.6±1.6	6.05±1.3
COD (mg/l)	388.8±2.8	43.9±1.8	34.9±2.4	27.0±21.
DO (mg/l)	3.2±0.8	4.8±1.7	5.4±1.2	6.8±1.3
Dissolved CO_2 (mg/l)	15.4±1.9	18.1±2.3	24.6±2.1	30.2±1.5
Total P (mg/l)	18.15±1.6	1.9±0.5	1.5±0.4	1.2±0.4
NH3-N (mg/l)	38.88±2.3	4.29±1.4	3.41±1.1	3.02±1.0

pH:

The study on pH value of wastewater, tap water and 8 times diluted wastewater was initially observed as 9.8±0.5, 7.9±0.3, 8.6±0.6 respectively. The pH value of the control (wastewater alone) was 8.7±0.2 at the end of the culture study. Similarly pH value of one time aeration, two times aeration and three times aeration was noticed as 8.2±0.5, 7.9±0.3, 7.7±0.6 respectively. After 21 days of algal production the pH value in control culture was reduced from 9.8 to 8.7 which may be possible due to volatilization loss and assimilation of ammonical nitrogen from the culture. In case of aeration the treated culture at the end of culture study revealed decreasing trend which indicates inhibition of pH due to mixing of carbon dioxide during aeration [13].

BOD, COD and DO:

This study revealed that BOD and COD value have been significantly reduced in treated culture in comparison to their initial value. The dissolve oxygen concentration in treated culture was found increased which may be due to algal photosynthesis in treated culture study. The reduction in pH value after treatment may be referred to microbial degradation of active organic matter in presence of higher concentration of oxygen evolved during process of photosynthesis.

Total Phosphorous and Ammonia:

The concentration of total phosphorous and ammonical nitrogen in wastewater were very high. The high concentration of these nutrients in wastewater body -Laxmi Tal is expectedly high due to accumulation of these nutrients since many decades. These nutrients in treated culture revealed a sharp reduction in both the value which may be referred to assimilation of nutrients by algal biomass production.

Mixing of Carbon dioxide Through Aeration:

Initial concentration of free CO_2in wastewater sample (control) was 3.8mg/l (Table 4). The concentration of free CO_2 in wastewater (control) showed a decreasing trend with increasing period of algal biomass production. After 21 days of wastewater culture through biomass production of algae indicated maximum reduction of carbon dioxide value as 1.8±0.25 mg/l. The maximum reduction of CO_2 in control culture was recorded on 14th day of culture treatment. The maximum reduction is supported by maximum production of algal biomass during this period. The 7th day biomass production in control was much less hence resulted less reduction of CO_2 by photosynthesis process. Lowest reduction of CO_2 value on 21st day in comparison to 14th day was recorded which may be referred to lowest fixation of CO_2 by algal biomass production by the process of photosynthesis. CO_2 concentration in aeration treatment cultures were consistently increased with increase of period of algal biomass production in treated culture. The maximum CO_2 increase was observed in three times aeration (33.2±1.2mg/l) of treated culture similarly the minimum increase(8.9±0.31) in the rate of CO_2 was recorded in one time aeration. Two times aeration showed medium rate of increase of CO_2 concentration with increase of period of culture for algal biomass production. The details of the CO_2 concentration in treated culture of aeration treatment were shown in Table 5. The increasing trend of CO_2 concentration in treated culture with increasing period of algal biomass production is explained by the fact that CO_2 concentration is constantly enhanced by the process of continuous and cumulative addition of CO_2 by aeration process and also due to increase microbial respiration in presence of high concentration of O_2 by photosynthesis.

Table 3: Variation of temperature during culture study:

Day	Initial	7th Day	14th Day	21th Day
Mean minimum Temp.(^{0}C)	17	18.3	18.92	22.00
Mean maximum Temp.(^{0}C)	27	27.14	27.07	29.80
Mean Temp.(^{0}C)	22	22.72	22.99	25.9

Biomass Production:

In our present investigation algal biomass production was studied in four different treatments. The details of the algal biomass production have been recorded in form of table (Table 4 and 5). In control culture biomass production revealed a continuous increased value with increase of period of culture. However in control the rate of algal biomass production was very slow in comparison to aeration treatment. At the end of culture treatment in control the biomass production was recorded as 14.3=0.52 g/l. The maximum biomass gain was recorded on 21st day than 7th and 14th day study which may be resulted due to cumulative growth. In comparison to aeration treatment control shown least biomass production which may be due to limiting factor i.e. CO_2 concentration [12]. In control culture the total biomass production was doubled on 14th day in compression to 7th day which may be due to the optimum environmental condition. However the biomass production rate was significantly decreased after 14th day which may be the adverse limiting factors. Algal biomass production in aeration treatment is already recorded in Table 5. The careful examination of algal biomass production in aeration treatment cultures revealed a consistent increase of the value with increase in CO_2 concentration. Similarly biomass production in treated culture also recorded a cumulative increase in value with increase of the period of culture time. One time aeration showed maximum production value 17.57+0.31g/l on 21st day. In two times treated culture the maximum production value 25.86+0.32 g/l recorded on 21st day. Similarly three times aeration also recorded maximum value 27.0+0.24 g/l algal biomass production on 21st day. The careful scrutiny of biomass production in the three aeration treatment revealed the rate of biomass growth was maximum in case of two aeration than other two times of aeration treatments. This trend may be sufficient to explain that two time aeration in our treatment with prevailing our environmental condition is best suited for maximum production.

Table-4: Algal biomass production in control (without aeration)

Parameters	Initial	7th day	14th day	21st day
Biomass(g/l)	ND	5.6±0.18	11.87±0.22	14.3±0.52
Free CO_2 (mg/l)	3.8±0.2	3.2±0.15	2.2±0.3	1.8±0.25

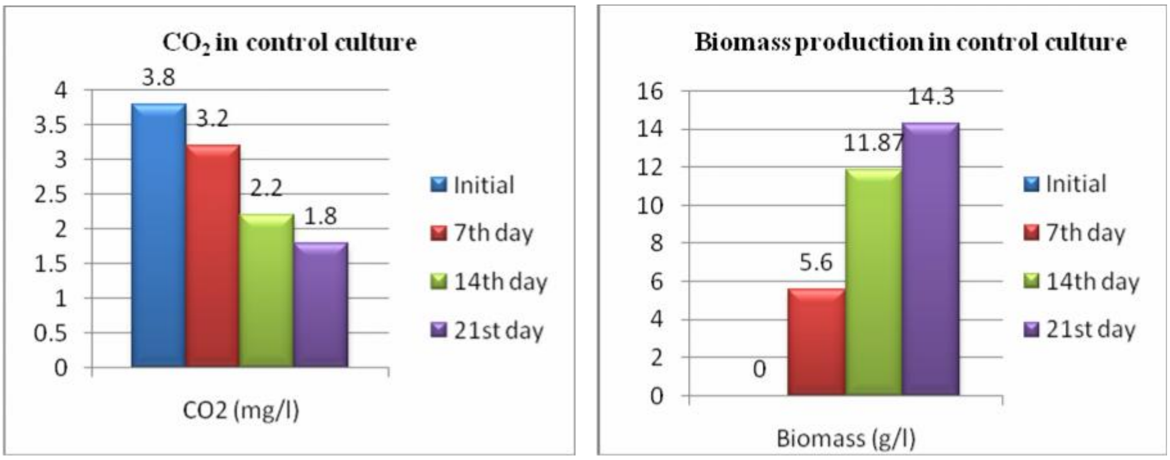

Fig 1: Algal biomass production in control (without aeration)

Table-5: Algal biomass production in 8 times diluted wastewater with different rate of aeration

Parameters	Initial	One time aeration			Two times aeration			Three times aeration		
Day		7^{th}	14^{th}	21^{st}	7^{th}	14^{th}	21^{st}	7^{th}	14^{th}	21^{st}
Biomass production (g/l)	Nil	6.96± 0.12	14.75± 0.32	17.57± 0.21	11.72 ± 0.22	19.65± 0.31	25.86± 0.32	13.34± 0.12	22.69± 0.12	27.0± 0.24
Free CO_2 (mg/l)	4.1± 0.26	8.9± 0.31	12.8± 0.42	14.1± 0.71	10.2± 0.46	15.5± 0.52	18.6± 0.55	28.5±0 .41	24.8± 2.0	33.2± 1.2

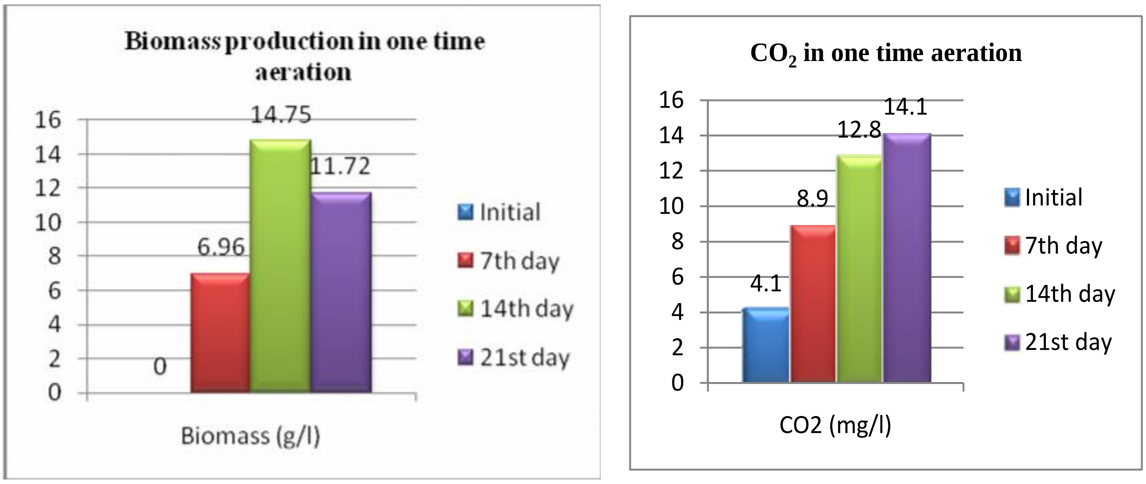

Fig 2: Biomass production in one time aeration

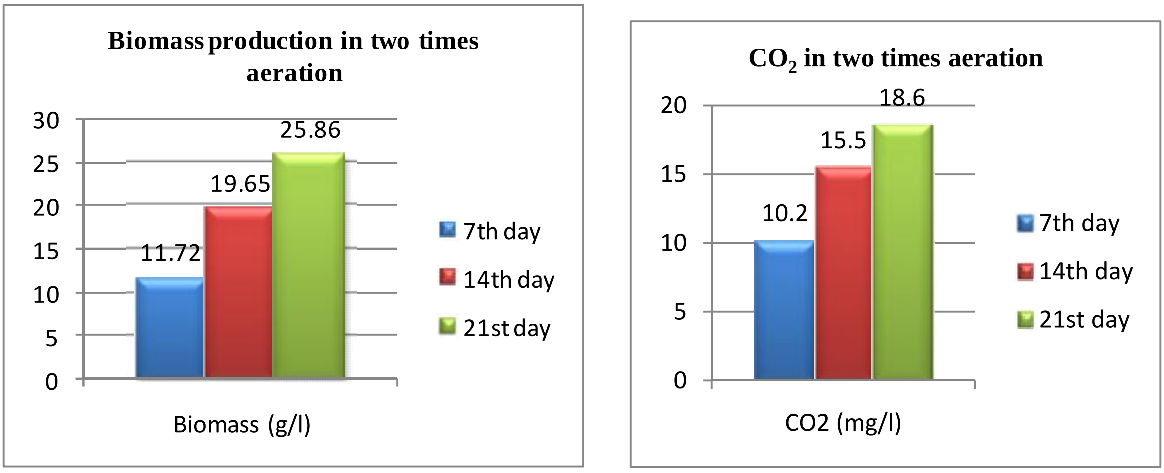

Fig 3: Biomass production in two time aeration

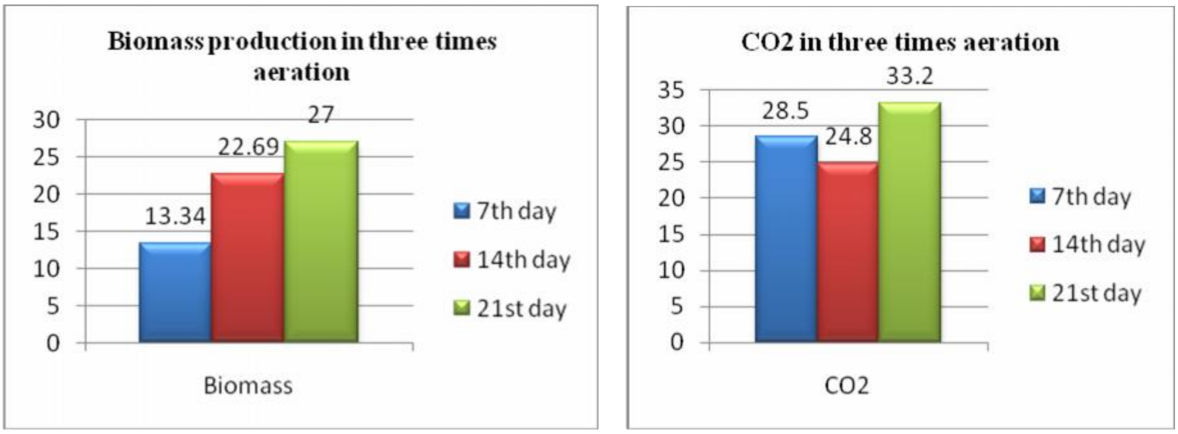

Fig 4: Biomass production in three times aeration

Conclusion

The very high alkaline pH (9.8) of the wastewater was observed in our study which is caused by high concentration of ammonical nitrogen. The ammonical nitrogen value is very high in (43.2mg/l) wastewater sample indicates unfavorable aquatic environment for growth of aquatic organisms.High BOD and COD value in our study of the wastewater sample also revealed a high rate of active organic pollution in Laxmi Tal whereas Very low concentration of DO indicates anaerobic aquatic environment which may cause absence of aquatic organisms i.e. fish.High nutrient status (NH_3 and P) of wastewater promise for a high potential for algal biomass production.Diluted wastewater culture with two times aeration seems to have commanding influence on algal biomass production in prevailing condition at Jhansi. This study finally explored the possibility for using wastewater and its nutrients for algal biomass productions hence enhance biodiesel production from algal biomass. The biodiesel so produce may be possibly replace traditional fuel, hence save our present day fuel crisis.

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