IOSR Journal Of Pharmacy And Biological Sciences (IOSR-JPBS)
e-ISSN:2278-3008, p-ISSN:2319-7676. Volume 14, Issue 5 Ser. III (Sep – Oct 2019), PP 85-88
www.Iosrjournals.Org

Growth and Study of *Spirulina platensis* in Different Conditions

Naveen Kumar[1], Sumit Kumar[2]
[1] *(Research Scholar, Faculty of Science and Technology, Department of Life Science, Rajasthan, India)*
[2] *(Associate Professor, Faculty of Science and Technology, Department of Life Science, Mewar University, Rajasthan, India)*
Corresponding Author: Dr.Sumit Kumar

Abstract: *In this research work we evaluate the growth and characteristics of Spirulina platensis that was grown in different conditions and mediums. Glass jar, poly bags and water tank were used in this work for the production of culture. Due to higher temperature, glass jar culture was found denser as compared to others. Glass jar culture produced more dry weight (1.0 g/L) than other (0.51 and 0.52 g/L). Percentage of protein level was found 56.4, 50.3 and 30.4 in water-tank, poly-bag and glass jar respectively. It was noticed that lesser volume culture increases its temperature faster than other, which is the reason for inhibiting the growth.*

Key Words: *Spirulina platensis, glass jar, poly bag, water tank, dry weight, protein level*

Date of Submission: 15-10-2019 Date of Acceptance: 31-10-2019

I. Introduction

Spirulina platensis is able of produce important metabolites, such as, proteins, vitamins and pigments for pharmaceutical and others [1-3]. They are very effective in the conversion of solar energy because of it they are good producer of biomass per unit area in the comparison of other terrestrial plants [4]. The *Spirulina platensis* have many significance and more demand for its high value phytonutrients and pigments, which play important role in health, animal feed, diagnostics and therapeutics. It represents the most important commercial blue green algae for the production of biomass as healthy food and animal feed [5-6].

Cyanobacteria have more than thirty thousand species which are used as unexploited resources and some species are used in different commercial production [7]. *Spirulina platensis* are very famous because it contains high value of protein [8] and is very common cyanobacteria which can be easily grown in water and can be harvested commercially. *Spirulina platensis* is commonly consumed as food by human in various countries, because it is a good source of vitamin, protein and aquaculture supplementary diets. These blue - green algae represent one of the mainly hopeful resources for many new products and purposes. *Spirulina platensis* is especially protein rich because it contains 65-73 percent protein with respect to its dry weight [9]. *Spirulina platensis* includes the constituents protein, vitamin B12, iron, essential amino acids etc., It has been considered as ‘‘Food of the future’’ and an ideal food for astronauts by NASA [10]. Various types of culture vessels like glass panels [11], photobioreactor [12] and open water tank [13] are used to grow culture in different mediums like Standard Zarrouk’s media, Modified Zarrouk’s media etc. Generally *Spirulina platensis* is produced commercially in open ponds for various reasons, such as low capital investment and free light energy from the sun [14].

II. Materials and Methods

The *Spirulina platensis* was arranged from Indian Agricultural Research Institute, Delhi, which was grown in Zarrouk’s medium [17], and the macronutrients were added into it. For the experiments three different types of culture strategy were used in this work. In first condition 30 L cylindrical glass jars (h-42cm, d-30cm), in second condition 300 L cylindrical transparent poly bag (h-100cm, d-62cm) and in third condition 3000 L cemented tank (h-32cm, d-345cm) were used. The glass jar and poly bag culture was covered by transparent plates to prevent the heat exchange. The depth of the tank culture was kept at 24 cm and the flow rate was 25 cm/s. The cells were exposed to outdoor conditions for 20 days without regulation in temperature. The circulation of the cultures was obtained by bubbling air in the glass jars and poly bag but paddle-wheels were used in water tank at the rate of 0.5 L min^{-1} L $culture^{-1}$.

Different required conditions as temperature, salinity, dissolved oxygen level and pH values were maintained. Filaments grown in water tank and culture were increased in volume for 11-12 days. After it when the density was enough in tank, culture shifted to the different vessels for growing under same nutrient conditions. UV Spectrophotometer was used for chlorophyll-a analyses at 670 nm. 25 ml of sample was used for measuring and calculating dry weight. Whatman’s filter papers were used for filtration and distilled water used for washing the samples and all the samples were dried in oven at 38-40℃ for 100- 120 minutes.

Filaments were counted by triplicate samples and specific growth rate (μ) and doubling time (d.t.) were calculated as:

$$\mu \text{ (cell /day)} = \frac{\ln A_2 - \ln A_1}{t_2 - t_1} \qquad \text{d.t (day)} = \ln 2/ \mu = 0.693/ \mu$$

where,
A_1 and A_2 represent the concentrations of biomass at different time intervals t_1 and t_2, respectively.
Kjeldahl method was used for protein determination. After filtration and drying the samples protein content was calculated.

III. Results

All experiments were carried out on clear day. Data for temperature (table-1) were recorded from early morning to evening in different experiments. The light variations) during day time was given in table-2.

Table 1: Temperature variations on a clear day in Water tank, Poly Bag and transparent glass jar

Time (hours)	**Temperature℃**		
	Water Tank	Poly Bag	Transparent glass Jar
6:30	16.5	15.5	17.0
8:30	18.5	17.6	19.0
10:30	21.6	20.8	22.0
12:30	27.4	26.8	28.2
14:30	32.8	31.5	33.4
16:30	27.8	27.0	28.5
18:30	24.5	24.0	24.8

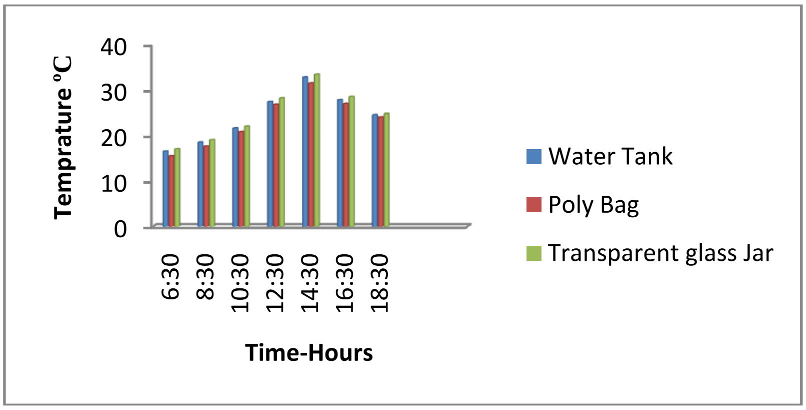

Figure 1: Variations in temperature on a clear day in Water Tank, Poly Bag and transparent glass jar

Table 2: Diurnal changes in light intensity on a clear day characterizing the period of the experiment in greenhouse (u) and outdoors (water tank)

Time (hours)	**Light intensity (μmol photon m^{-2} s^{-1})**	
	Water Tank (Outdoor)	Poly-Bag (Greenhouse)
6:30	315	110
8:30	350	130
10:30	730	315
12:30	1110	590
14:30	1330	825
16:30	715	410
18:30	08	00

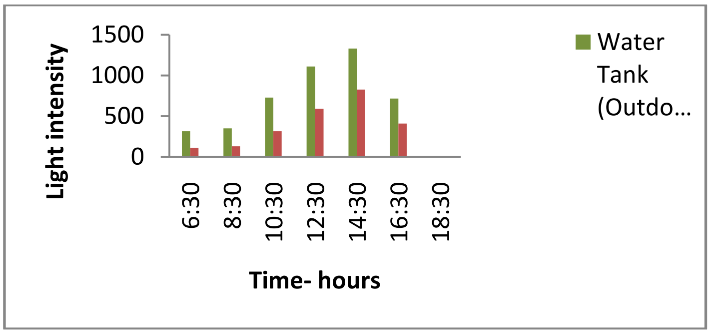

Figure 2: Light intensity on a clear day characterizing the period of the experiment in greenhouse and outdoors

In the present work, all experiments were initiated with 0.10 g/L in DW. It was noticed that the dry weight increase till the 9th day in transparent jar while in water tank and poly bag increased up to 7th day. Growth was mostly constant when limited light provided to the cell (fig-3). After the completion of experiment the dry weight for the transparent jar, water tank and poly bag cultures were 1.0, 0.51 and 0.52 g/L respectively. It was noticed that the volumetric productivities were 0.82 0.35 and 0.32 g /L in transparent jar, water tank and poly bag cultures respectively. According to data it was clear that no significant change occurs in terms of dry weight values in outdoor and greenhouse cultures.

Table 3: Changes in dry weight amounts of *Spirulina platensis* cultivated in glass jar, polythene bag (Greenhouse) and water tank (Outdoor)

Time (Days)	Dry Weight (g/L)		
	Glass Jars	Water Tank (Outdoor)	Poly Bag (Greenhouse)
1	0.10	0.10	0.10
5	0.66	0.38	0.40
10	0.92	0.44	0.45
15	1.3	0.46	0.48
20	1.0	0.51	0.52

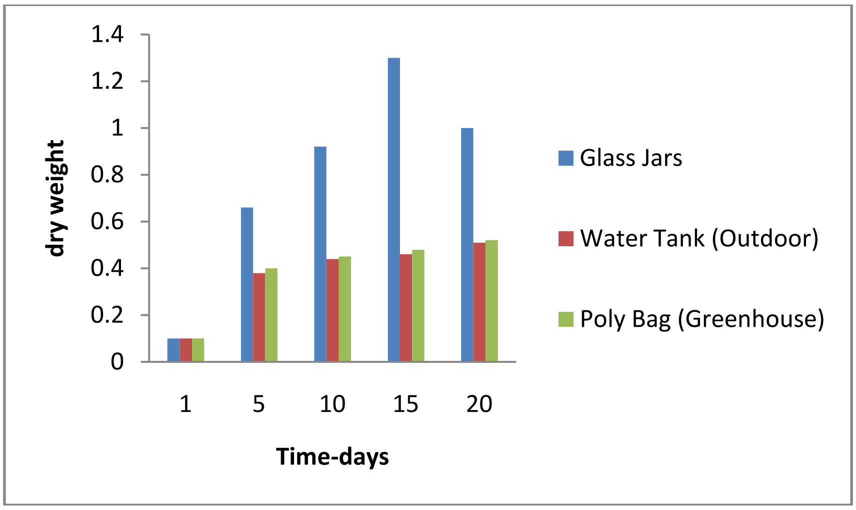

Figure 3: Changes in dry weight amounts of *Spirulina platensis* in transparent glass jars, Poly Bag (Greenhouse) and Water Tank (Outdoor)

It was found that the poly-bag (green house) has maximum protein content. The mean value of dissolved oxygen in different cultures vessels increased every day with the increase in the biomass. The amount of oxygen production was maximum in glass jar culture as compare to others until the 10th day. The dissolved oxygen levels in the cultures were found to be 20.1, 26.3 and 15.7 mg/L at the end of the experiment for glass jar, poly-bag (Green house) and water tank cultures, respectively. The mean pH values of the cultures were found to be 10.8, 9.8 and 9.6 at the end of the experiment for glass jar, poly-bag and water tank cultures, respectively (table-4). The color of glass jar culture had been changed to green which is main characteristic of *Spirulina platensis*.

Table 4: Changes in specific growth rates, doubling time, protein, temperature, pH and dissolved oxygen in different cultures

Experiments	Protein (%DW)	Doubling time (d.t.)	Growth rate (μ)	T (˚C)		pH		DO (mg/L)	
				Min	Max	Min	Max	Min	Max
Glass Jar	30.4	2.12	0.32	17.0	34.0	9.0	10.8	10.4	20.1
Poly-Bag (Greenhouse)	50.3	3.15	0.20	17.2	33.2	8.9	9.8	10.2	26.3
Water tank (Outdoor)	56.4	3.40	0.19	15.8	30.5	9.0	9.6	10.0	15.7

IV. Discussion

During this research work the intensity of the sun light was low, when the experiments were started. While the light intensity during afternoon (2:30 pm) in outdoors is as high as 2000 µmol photon m^{-2} s^{-1} in the summer period when sunlight enter the earth directly, it was just 1330 µmol photon m^{-2} s^{-1} in the starting of March. The major factor for low production was decrease in temperature. The biomass increase strongly correlated with the temperature of the culture which can be noticed by the comparative study of Figure 1 and Figure 3.

Factors responsible for protein deficiency in the glass jar culture are as the higher biomass concentration, fast growth of the cells and the addition of $NaNO_3$ (M = 1.25 g/ L) in the medium. pH also play an important role for growth of cells so it should be maintained properly. The maximum pH value attained during this research work for glass jar, poly bag and water tank were10.8, 9.8 and 9.6 respectively. It was noticed during the experiment that pH more than 10.8 is harmful for the growth of cells. The accumulation of CO_2 into the extremely fruitful small volume cultures, in which maximum biomass concentrations are reached, would be helpful in keeping the pH in most favorable range. According to our work, the cells in the water tank culture were limited by the light due to the greater diameter compared to the others and the growth was reduced.

V. Conclusion

The temperature and light were two main factors which affect the growth of culture. In this research work growth is affected more due to optimum temperature as compared to light. In small volume culture temperature increases faster in greenhouse conditions so growth of culture was hindered. Optimum pH was maintained by the addition of CO_2 into the culture medium for the growth of biomass. Short light-path lengths also support *Spirulina plantesis* to the small culture volumes for higher productivity. The use of the macronutrients in the experiment at half strength also gave a positive result for the growth of *Spirulina plantesis*. Some of the commercial microalgae production plants use the Zarrouk medium in the water tank for reducing the costs of nutrient.

Acknowledgement

The author’s thanks IARI New Delhi and Dr. J. P Singh for his kind help during this research work. I am also grateful to Mewar University, Rajasthan for giving me an opportunity to pursue my Ph.D.

References

[1]. M. Cartens, E. Molina, A. Robles et al., Eicosapentaenoic acid (20:4n-3) from the marine microalga Phaeodactylum tricornutum, *Journal of the American Oil Chemists Society*, 73: 1996, 1025–1031.

[2]. M. Guerin, M.E Huntley, M. Olaizola, Haematococcus astaxanthin: applications for human health and nutrition, *Trends Biotech*, 21: 2003, 210–216.

[3]. Q. Hu, Industrial production of microalgal cell mass and secondary products – major industrial species: Arthrospira (Spirulina) platensis. In: Richmond A. ed. Handbook of Microalgal Culture: Biotechnology and Applied Phycology. Blackwell Science Ltd.; Oxford, 2004, 264-272.

[4]. T. Schulz, The economics of micro-algae production and processing into biofuel, Australia, 2006.

[5]. A. Richmond, Efficient utilization of high irradiance for production of photoautotrophic cell mass: a survey, *Journal of Applied Phycology*, 8, 1992, 381–386.

[6]. Raoof, B. D Kaushikand, R. Prasanna, Formulation of a low-cost medium for mass production of Spirulina, *Biomass and Bioenergy*. 30(6), 2006, 537-542.

[7]. Berg-Nilsen, *J. Production of micro algea-based products*, Norway. 2006,

[8]. Salunke K. J., Magar S. A., Joshi R. R and Wadikar M. S., , Comparative study on growth of Spirulina platensis on different culture media, *Bioscience Discovery*, 7(1), 2016, 90-92.

[9]. Shams M., Haji- Aghababa A.,. Kardani S. M., Esfahani and Amini N. G., Industrial Production of Microalgae Arthrospira (Spirulina) platensis in the Central Iran, *Int. J. Pure App. Biosci.* 5 (4): 31-36, 2017.

[10]. J.P Pandey and A. Tiwari, Optimization of Biomass Production of Spirulina maxima, *J Algal Biomass Utill*., 1(2), 2010, 20- 32.

[11]. Q. Hu, H. Guterman, A. Richmond, A flat inclined modular photobioreactor (FIMP) for outdoor mass cultivation of photoautotrophs. *Biotechnol. Bioeng*, 51: 1996, 51-60.

[12]. G. Torzillo, B. Pushparaj, F. Bocci et al., Production of Spirulina biomass in closed photobioreactors, *Biomass*, 11: 1986, 61-74.

[13]. Y.K Lee, Commercial production of microalgae in the Asia-Pacific rim, *J. Appl. Phycol*. 9: 1997, 403-411.

[14]. M. Tredici, G. Z Chini, S. Biagiolini et al., Novel photobioreactors for the mass cultivation of Spirulina spp, Bull. Inst. Oceanogr. *Monaco* 12: 1993, 89–96.

Naveen Kumar " S Growth and Study of *Spirulina platensis* in Different Conditions" IOSR Journal of Pharmacy and Biological Sciences (IOSR-JPBS) 14.5 (2019): 85-88.