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Effect of light and temperature on growth and pH on biomass production of cyanobacterial species *Nostoc linckia* and *Synechocystis aquatilis* isolated from crop fields of Samastipur, Bihar, India

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ABSTRACT

Two strains of Cyanobacteria *i.e.*, *Nostoc linckia* and *Synechocystis aquatilis* collected from the crop fields of Samastipur, Bihar were studied under laboratory condition aiming to utilize their potential use in biotechnological products. Three different colours used in the present study were green, red, blue and florescent light and the temperature ranging from 15^{O}C to 40^{O}C and pH from 6.5 to 9.0. Growth of *N. linckia* and *S. aquatilis* was best at the temperature 30^{O}C and 35^{O}C respectively, at pH 8.5 and under florescent light.

Keywords: *Cyanobacteria, Growth, Biomass, Temperature, pH, Color, Biotechnology, Nostoc linckia, Synechocystis aquatilis.*

INTRODUCTION

Cyanobacteria are basically aerobic photoautotrophs. Their life processes require only oxygen, light and inorganic substances. But few species of cyanobacteria like, *Oscillatoria* that is found in mud at the bottom are able to live anaerobically. They can live in extremes of temperature -60°C to 85°C, and a few species are halophilic or salt tolerant (as high as 27%, for comparison, concentration of salt in seawater is 3%). Some species of cyanobacteria can grow in full sunlight and some in almost complete darkness. They are often the first plants to colonize bare areas of rock and soil, as an example subsequent to cataclysmic volcanic explosion. Unlike more advanced organisms, these need fewer substances for their metabolism. Biotechnological potential of cyanobacteria has been established in many fields (Bhadury *et al.*, 2004; Dahms *et al.*, 2006; Thajuddin and Subramanian, 2005). Biotechnological accounts of the cyanobatcteria native to Samastipur are not recorded as yet. Present paper is an attempt to find out maximum biomass production of local strains of cyanobacteria.

Considering the above bare facts about the cyanobacteria and to find out the most suitable environmental conditions for mass production, effect of different types of lights, temperature on the growth, and pH on biomass production was studied with reference to local strain of *Nostoc linckia* and *Synechocystis aquatilis*.

MATERIALS AND METHODS

Isolation of *Nostoc linckia* and *Synechocystis aquatilis* was done from the crop fields of Samastipur, Bihar. Identification of the taxa was based on Desikachary (1959). The collected soil samples were incubated under laboratory conditions after being irrigated by

Basal Bold inorganic nutrient solution (Nichols and Bold, 1965). Emergence of desired cyanobacteria from the incubated soil was carefully watched under microscope. Isolation and purification of the taxa emerged from the incubated soil samples were done following the standard methods described by Kaushik (1987). The growth of the test organisms was measured turbidimetrically by optical density determination of the homogenous culture suspensions at 663 nm. spectrophotometer.

Effect of different types of light on the growth of cyanobecteria was studied using wrapping coloured transparent papers over the florescent tube light. Similarly, effect of temperature on the growth of cyanobacteria was studied by placing the tungsten bulb of different watts. The temperature was maintained by adjusting the wattage of bulb. The Tungsten bulbs used for maintaining the temperature in the cultural well were hidden by a black cloth separating the exposure cabinet of tungsten bulb with florescent tube light. pH of the culture media was adjusted by adding dilute NaOH and HCl solution.

RESULTS AND DISCUSSION

In the present study, effect of different types of light and temperature was studied on the growth pattern of *N. linckia* and *S. aquatilis*. Biomass production of these two Cyanobacteria was also studied at different pH under the laboratory conditions.

The growth of *N. linckia* and *S. aquatilis* was studied upon the exposure to different types of light under laboratory condition. Three different colours used in the present study were green, red, blue and florescent light. The culture of *N. linckia* and *S. aquatilis* was exposed to different light under laboratory condition for continuous 24 days with proper 12 hour of dark phase. The growth of Cyanobacteria was measured turbidimetrically by Optical Density (OD) determination method as described in "Materials and Methods".

It is evident from the Graph 1 that the growth of *N. linckia* varied considerably on the exposure to different colours of light. The maximum growth was found under the florescent light followed by Green and Red during the observation period of 24 days. Remarkably, negligible growth was observed upon exposure to blue light. It is also evident that maximum growth OD 1.10 was observed on 18^{th} day of inoculation in florescent light; OD 0.57 in green light on 18^{th}, 21^{st}, 24^{th} day; OD 0.43 in red light on 21^{st} and 24^{th} day and OD 0.08 in blue light on 9^{th} and 12^{th} day.

As shown in the Graph 2, the growth of *S. aquatilis* varied considerably on the exposure to different colours of light. The maximum growth was found under the florescent light followed by Green and Red throughout the observation period of 24 days. Remarkably, minimum growth was observed upon the exposure to blue light. It is also evident that maximum growth OD 0.95 was observed on 21^{st} day of inoculation in florescent light; OD 0.55 in green light on 18^{th}, 21^{st} & 24^{th} day; OD 0.45 in red light on 15^{th}, 18^{th} & 21^{st} day and OD .20 in blue light on 12^{th} day.

As shown in the Graph 3, the growth of *N. linckia* varied considerably on the exposure to different temperature. It is evident from the Graph 3 that the maximum growth was found at $30^{O}C$ temperature followed by $35^{O}C$, $25^{O}C$, $40^{O}C$ and $15^{O}C$ during the observation period of 24 days. It is also evident that maximum growth OD 0.41 was observed on 18^{th} day of inoculation at $15^{O}C$; OD 0.50 at $25^{O}C$ on 18th, 21^{st} & 24 day, OD 1.20 at $30^{O}C$ on 21^{st} day; OD 1.00 at $35^{O}C$ on 18^{th} & 19^{th} day and OD 0.41at $40^{O}C$ on 12^{th} day.

As shown in the Graph 4 the growth of *S. aquatilis* varied considerably on the exposure to different temperature. It is evident from the Graph 4 that the maximum growth was found at $35^{O}C$ temperature followed by $30^{O}C$, $25^{O}C$, $40^{O}C$ and $15^{O}C$ during the observation period of 24 days. It is also evident from the table 5.4 that maximum growth OD 0.40 was observed on 18th day of inoculation at $15^{O}C$; OD 0.90 at $25^{O}C$ on 18th; OD 1.10 at $30^{O}C$ on 18th day; OD 1.20 at $35^{O}C$ on 18th,21st and & 24th day and OD 0.52 at $40^{O}C$ on 18th and 24th day.

The Graph 5 shows the effect of pH on the biomass production of *Nostoc linckia* and *Synechocystis aquatilis* grown under laboratory condition. It is also evident from the Graph 5 that maximum biomass production was observed 140 mg/L of dry weight in case of *Nostoc linckia* and 145 mg/L of dry weight in case of *Synechocystis aquatilis* at the pH 8.5.

Effect of different types of light, temperature on the growth and pH on biomass production was studied with reference to local strain of *Nostoc linckia* and *Synechocystis aquatilis* to find out the most suitable environmental conditions for mass production. In spite of the publication of complete genome sequence of the cyanobacterium *Synechocystis* 6803 in the year 1996, our knowledge of cyanobacteria light-signal transduction remains fragmentary (Conrad, 2001). The present experimental results showed that the florescent light was ideal for the growth in *N. linckia* and *S. aquatilis* as compared to other monochromatic light - green, red and blue. Minimum growth of both the cyanobacteria was observed in blue light. Sinha *et al.* (2002) observed the effect of UV and visible light on the growth of *Anabaena* and *Scytonema* and found that the blue light was more effective than the red and green.

The experimental results further showed that the optimum growth of *N. linckia* was at $30^{O}C$ followed by $35^{O}C$, $25^{O}C$ and $15^{O}C$ temperature and that of *S. aquatilis* was at $35^{O}C$ followed by $30^{O}C$, $25^{O}C$, $40^{O}C$ and $15^{O}C$.

Creswell (2010) observed that most of the phytoplankton species grew best at temperature ranging from 16 to $27^{O}C$. Cyanobacteria typically grow better in hotter environment ($27^{O}C$-$35^{O}C$) in comparison to diatoms and dianoflagellates (Kudo *et al.*, 2000; Butterwick *et al*., 2005; Yamamoto and Nakahara, 2005; Boyd *et al*., 2013 and Lurling *et al*., 2013). Kumar *et al*. (2011) found that *Spirulina platensis* has a wide range of temperature tolerance from $20^{O}C$ to $40^{O}C$. Renaud *et al.* (2002) attributed the higher growth rate of *Chaetoceros* sp. to an increase in temperature from $25^{O}C$ to $30^{O}C$. In the present study two species of cyanobacteria *N. linckia* and *S. aquatilis* grew best at $30^{O}C$ and $35^{O}C$ respectively. The experimental results also showed that the maximum growth of both the cyancobacteria *N. linckia* and *S. aquatilis* was at pH 8.5. The growth of both cyanobacteria increased gradually in alkaline medium up to the value of pH 8.5.

Kleber *et al.* (2011) observed interesting fact on the cyanobacteria *Anabaenopsis elenkinni* that there was no significant difference in growth when grown at the pH 7.0 to 9.5. Sushanth and Madaiah (2014) reported that the cyanobacteria namely, *Chroococcus turgidus*, *Lyngbya confervoides* and *Nostoc commune* and the diatoms *Chaetoceros calcitrans* and *Skeletonema costatum* showed maximum growth at pH 7.5. The pH value lower or higher than these values was associated with their decreased growth. The limitation of phytoplankton growth and photosynthesis at elevated pH levels was observed by Chen and

Durbin (1994). At high or low pH, cells may have to spend energy for maintenance of an internal pH necessary for cell function (Raven and Lucas, 1985). Earlier studies related to pH effect on the growth of cyanobacteria has revealed that pH between 7.4 and 8.0 is favorable for the optimum growth of cyanobacterial species (Rippka *et al.*, 1979 and Bano and Siddiqui, 2004). The fact that all cyanobacteria were able to grow in acidic (pH 6.5) medium indicates that cyanobacteria can adapt to variable pH conditions as suggested by Buck and Smith (1995) and Burja *et al.* (2002).

Fig. 1. Effect of different types of light on the growth of *Nostoc linckia*

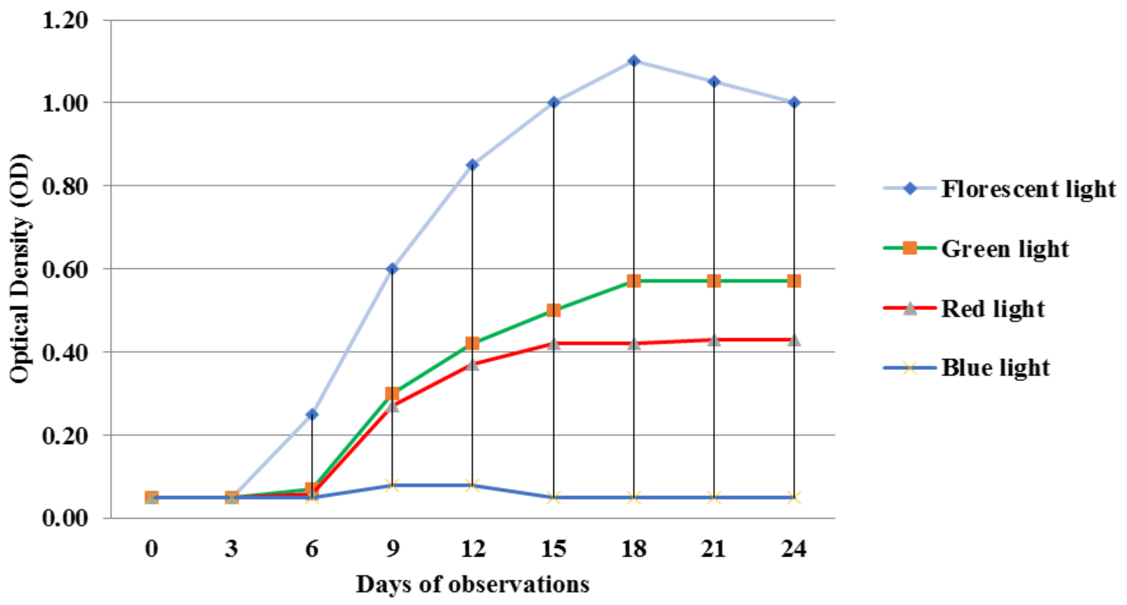


Fig. 2. Effect of different types of light on the growth of *Synechocystis aquatilis*

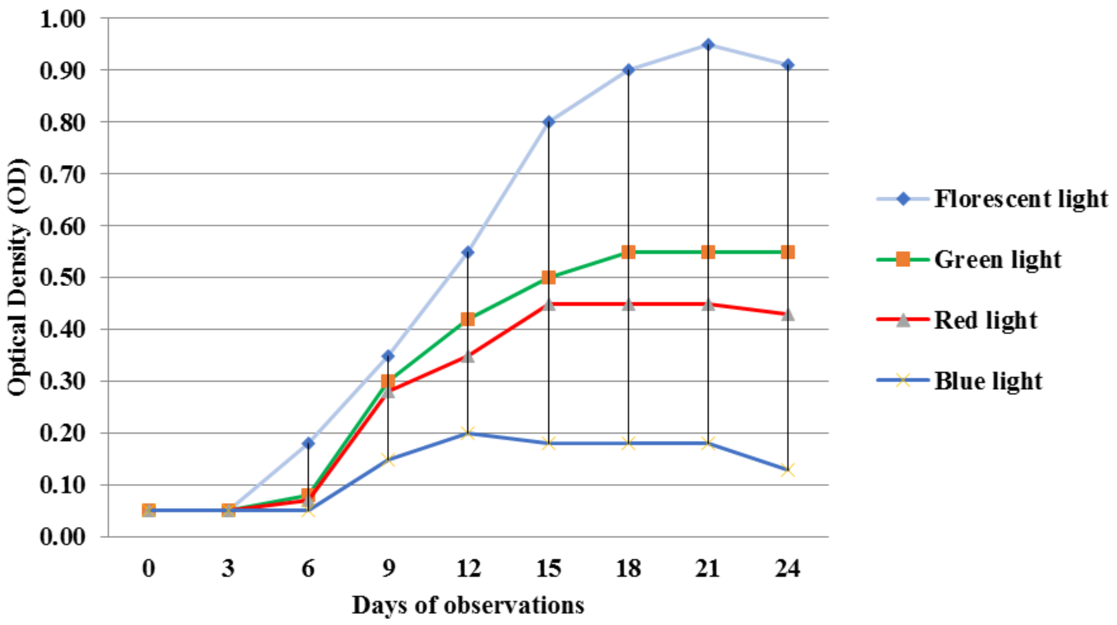


Fig. 3. Effect of temperature on the growth of *Nostoc linckia*

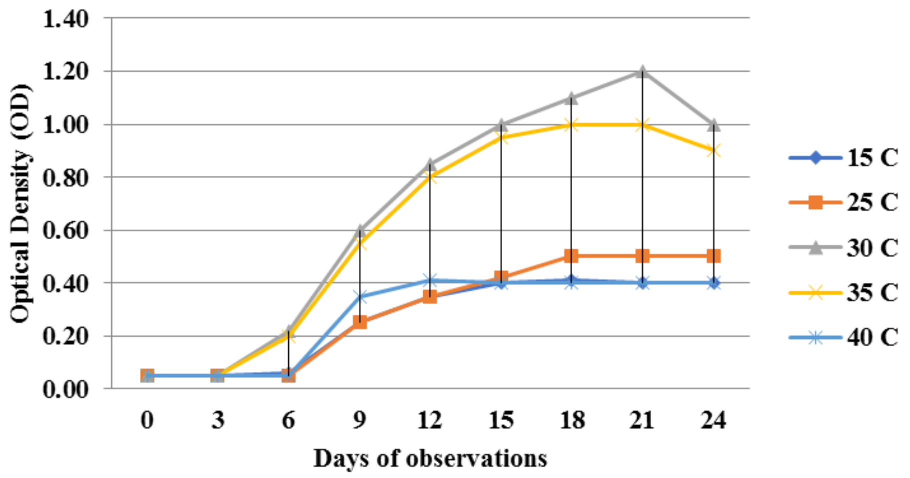


Fig. 4. Effect of temperature on the growth of *Synechocystis aquatilis*

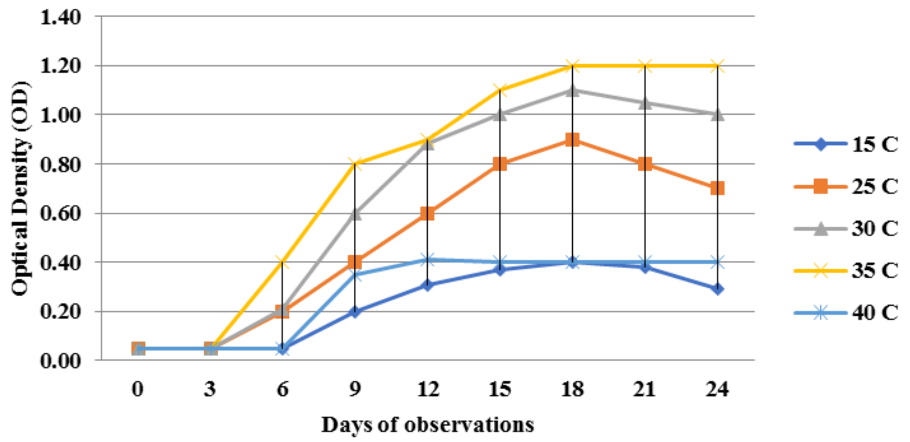
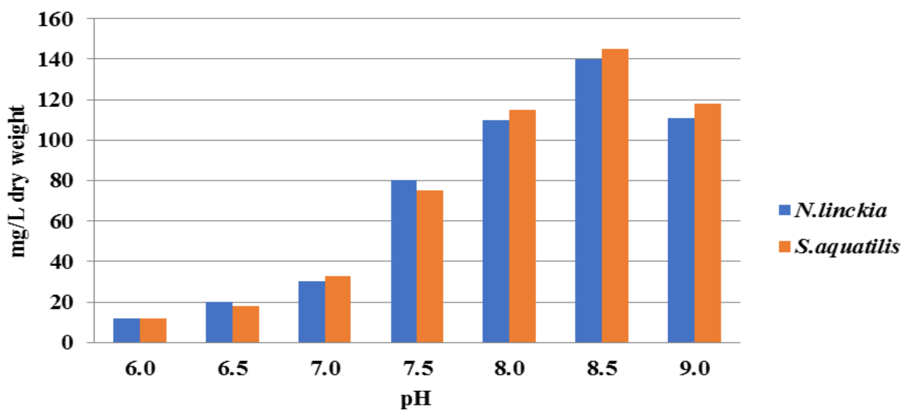


Fig. 5. Effect of pH on the biomass production of *Nostoc linckia* and *Synechocystis aquatilis*



CONCLUSION

Two strains of Cyanobacteria *i.e.*, *Nostoc linckia* and *Synechocystis aquatilis* collected from the crop fields of Samastipur showed best growth under laboratory condition at temperature of $30^{O}C$ and $35^{O}C$ respectively, at pH 8.0 and under florescent light. It can be utilized for their potential use in biotechnological products.

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REFERENCES

1. Bano, A. and P. J. A. Siddiqui. 2004. Characterization of five marine cyanobacterial species with respect to their pH and salinity requirements. *Pak. J. Bot*. 36:133 - 143.
2. Bhadury, P. and Wright, P. C. 2004. Exploitation of marine algae: biogenic compounds for potential antifouling appli-cations. *Planta* 219:561–578.
3. Boyd, P. W., T. A. Rynearson, E. A., Armstrong, F. Fu., Hayashi, K., Hutchins, Z. Hu., Kudela, D. A. E. Litchman, E., Mulholland, M.R., Passow, U., Strzepek, R. F., K. A. Whittaker, K. A., and M. K. Thomas, M. K. 2013. Marine phytoplankton temperature versus growth responses from polar to tropical waters – outcome of a scientific community-wide study. *PLoS ONE* 8(5).
4. Buck, D. P. and Smith, G. D.1995. Evidence for a Na+/ H+ electrogenic antiporter in an alkaliphilic cyanobacterium *Synechocystis*. *FEMS Microbiol. Lett*. 128: 315 - 320.
5. Burja, A. M., E., Abu-Mansour, B., Banaigs, C., Pyari, J., Burgess, G. and Wright, P. C. 2002. Culture of marine cyanobacterium, *Lyngbya majuscula* (Oscillatoriaceae), for bioprocess intensified production of cyclic and linear lipopeptides. *J. Microbiol. Methods* 48: 207 - 219.
6. Butterwick, C., Heaney, S. I. and Talling, J. F. 2005. Diversity in the influence of temperature on the growth rates of freshwater algae, and its ecological relevance. *Freshwater Biology* 50: 291-300.
7. Chen, C. Y. and Durbin, E. G. 1994. Effects of pH on the growth and carbon uptake of marine phytoplankton. *Mar. Ecol. Prog. Ser*. 109: 83 - 94.
8. Conrad, W., Mullineaux, T. 2001. How to cyanobacteria sense and respond to light? *Molecular microbiology*, 41(5) :955-977.
9. Dahms, H.-U., Xu, Y. and Pfeiffer, C. 2006. Antifouling potential of cyanobacteria: a mini-review. *Biofouling* 22:317–327.
10. Desikachary, T. V. 1959. Cyanophyta. Pub. Indian council of Agr. Res., New Delhi, pp. 686.
11. Kaushik, B. D.1987. Laboratory Methods for blue-green algae. Associated Publishing Company, New Delhi, *p.* 171.
12. Kleber, Renam de Souza Santos, Fernanda, Rios Jacinavicious and Celia, Leite Sant' Anna, 2011. Effects of the pH on Growth and morphology of *Anabaenopsis elenkinii* Miller (Cyanobacteria) isolated from the alkaline shallow lake of Brazillian Pantanal, *Fottea*, 11: 119-126.
13. Kudo, I., Miyamoto, M., Noiri, Y. and Maita, Y. 2000. Combined effects of temperature and iron on the growth and physiology of the marine diatom *Phaeodactylum tricornutum* (Bacillariophyceae). *Journal of Phycology* 36:1096-1102.
14. Kumar, M., Kulshreshtha, J. and Singh, G. P. 2011. Growth and biopigment accumulation of cyanobacterium *Spirulina platensis* at different light intensities and temperature. *Braz. J. Microbiol*. 42: 1128 - 1135.

15. Lurling, M., Eshetu, F., Faassen, E. J., Kosten,S. and Huszar, V. M. 2013. Comparison of cyanobacterial and green algal growth rates at different temperatures. *Freshwater Biology* 58:552-559.

16. Nichols, H. W. and Bold, H. C. 1965. *Trichosarcina polymorphgen*. et. sp. nov. *J. Phycol.* 1: 34-38

17. Raven, J. A. and W. J. Lucas, W. J. 1985. Energy costs of carbon acquisition. In: Lucas, W. J. and J. A. Berry (Eds), Inorganic Carbon Uptake by Aquatic Phytosynthetic Organisms. *American Society of Plant Physiologists,* Rockville. *p.* 305 - 324.

18. Renaud, S. M., Thinh, L. V., Lambridis, G. and Parry, D. L. 2002. Effect of temperature on growth, chemical composition and fatty acid composition of tropical Australian microalgae grown in batch cultures. *Aquaculture* 211: 195 - 214.

19. Rippka, R., Deruelles,J., Waterbury, J. B. Herdman, M. and Stainer, R. Y. 1979. Generic assignments, strain histories and properties of pure cultures of cyanbacteria. *J. Gen. Microbiol*. 111: 1 - 61.

20. Sinha Rajeshwar P., Peter Richter.,Jaoudat Faddoul., Markus Braun. and Donat-P- Hader. 2002. Effects of UV and visible light on cyanobacteria at the cellular level. *Photochemial and Photobiological Science*, 1(8): 553-559.

21. Sushanth, V. R. and Madaiah, R. 2014. Effect of pH, salinity and temperature on the growth of six species of marine phytoplankton. *J. Algal Biomass Utln.* 5(4): 55-59.

22. Yamamoto Y. and Nakahara, H. 2005. The formation and degradation of cyanobacterium Aphanizomenon flos-aquae blooms: the importance of pH, water temperature, and day length. *Limnology* 6:1-6.