ABSTRACT

Bioaugmentation is one of the essential components of bioremediation, a biotechnological method of recent origin. Bioaugmentation is the process of introducing a known variety of bioremedial agent into contaminated environment to initiate and sustain a specific bioremedial process. The paper deals with the biomass production of Nostoc punctiforme under the influence of different concentrations of cadmium chloride in laboratory culture. The success of bioremedial process by N. punctiforme depends upon its biomass production, which in due course removes the heavy metal. In control culture the biomass of N. punctiforme as mg/50ml varied from 13.79 (0 day) to 22.88 (25th day). The corresponding values for 0.01 and 0.5 ppm cadmium chloride treatment after 25 days were 20.68, 14.98 and 10.01 mg/50ml respectively, indicating a decline in biomass of N. punctiforme with increase in concentration. A two way ANOVA revealed the variation of biomass with respect to concentration and duration of treatment statistically significant (F = 10.494, df5,3; p < 0.001 and F = 20.004, df 5,3; p < 0.001). The results indicate better performance of N. punctiforme at lower concentrations.

INTRODUCTION

The use of toxic chemicals in agriculture, industries and other sectors has grown phenomenally, as per estimations derived from correlation between hazardous waste and toxic chemicals (Sahu and Panigrahi, 2002). Every industry creates pollutants in the form of smoke, effluents, noise etc. A significant amount of these pollutants are released into the environment, affecting the flora and fauna. Many inorganic elements such as mercury (Hg), lead (Pb) Arsenic (As) and Cadmium (Cd) are biological poisons at concentrations even in parts per billion (ppb) range. Once these chemicals find their way into the environment, a major portion reaches the sand and sediments which in turn serve as sink (Haqne, 1975). Leaching of chemicals pose ground water contamination problems (Hamaker, 1972). Widespread contamination by these chemicals or compounds is a serious problem faced by the mankind. With increasing pollution and health hazard, a number of methods have been developed to mitigate the impact or remove the contaminants from the natural systems. Among the various technologies employed for the purpose, bioremediation is becoming an increasingly popular technology for remediation of the contamination. Bioremediation is a treatability technology that uses the biological activity to reduce the concentration or toxicity of a pollutant. It commonly uses the process by which microorganisms transform or degrade chemicals in the environment *(King et al., 1998). The process of bioremediation includes biostimulation of naturally occurring microbial communities or bioaugmentation with specific microbial strains known to biodegrade the oxygenate. (Smith et al., 2005).

Bioaugmentation is a process of introducing a known variety of bio-remanant to initiate and sustain a specific bioremedial process. The success of the bioremedial process depends upon biomass production of the strain used as bioremedial agent. The more biomass is produced the more contaminants are removed. With this idea the present project has been taken up to study the influence of Cadmium chloride on biomass production of Nostoc punctiforme at different durations and different concentrations.

MATERIALS AND METHODS

Pure strain of Nostoc punctiforme was cultured in BGII medium (Stanier et al., 197). Before the inoculation, the culture medium was subjected to autoclaving for sterilization. The cyanobacterial culture was maintained under continuous white light (2200 lux) at the temperature...
28 ±2º C. Besides the culture flasks were subjected to hand shaking intermittently.

**Experimental setup**

Conical flasks of size 250 ml were used for the experiment. Four sets of flasks each with four replicates containing 50 ml of basal nutritional media (BNM) were taken. To each flask 2ml of the *N. punctiforme* culture was introduced. One set of inoculated flask (four flasks) was kept as control. To the other three sets 0.01, 0.1 and 0.5 ppm of cadmium chloride (CdCl₂) was introduced. All the conical flasks were maintained at the temperature 28 ±2º C under continuous white light (2200 lux).

**Estimation of biomass**

For the estimation of biomass the total content of the cultured flask was filtered through a pre-weighed Whatman No. 41 filter paper. After filtration, the filter papers containing the algal mass were dried keeping in an air oven at 80ºC for 2 hrs and at 30ºC for 12 hrs. The final biomass of dried cyanobacteria was calculated from the difference between the final weight of filter paper containing the dried cyanobacterial biomass and the initial weight without cyanobacteria.

**Estimation of biomass production**

Biomass production was calculated from the summation of all positive increments in the biomass over a period of 25 days of culture.

**Estimation of biomass turnover**

Turnover of the biomass was calculated by using the formula:

\[
\text{Turnover} = \frac{\text{Biomass mass (observed)} - \text{Biomass mass (initial)}}{\text{Days}}
\]

**Estimation of Instantaneous Growth Rate (IG)**

Instantaneous growth rate was determined following Brafrild and Llewellyn (1982).

\[
\text{IG} (%) = \frac{\log_{10} YT - \log_{10} Yt}{T - t} \times 2.3026 \times 100
\]

where 
\( t \) = time at the beginning of the observation,  
\( T \) = time at the end of the observation,  
\( YT \) = weight at time T,  
\( Yt \) = weight at time t and  
2.3026 is conversion factor.

**Statistical Analysis**

The data obtained were subjected to the analysis of variance (ANOVA) following Shcedecor and Coachran (1967).

**Observations**

The biomass of *N. punctiforme* ranged from 1.79 mg/50ml (on 0 day) to a maximum of 22.88 mg/50ml (on 25th day) in the control condition. However the corresponding values for different concentrations of CdCl₂ i.e. 0.01, 0.1 and 0.5 ppm after 25 days (mg/50ml) were found to be 20.68, 14.98 and 10.01 respectively (Table 1), which shows a decline in biomass of *N. punctiforme* with the increase of CdCl₂ concentration. The data were subjected to two way analysis of variance. The analysis revealed the variation of biomass statistically significant with respect to different concentrations of CdCl₂ treatments and different days (Table 2).

The biomass production varied from 4.40 mg/50ml in control whereas in case of 0.01, 0.1 and 0.5 concentration it ranged from 4.28 to 18.89, 3.80 to 13.19 and 1.90 to 8.22 mg/50ml respectively. The variation implies the gradual decline in biomass production with increasing concentration of CdCl₂.

The biomass turnover value was 11.78 in control while in 0.01, 0.1 and 0.5 concentrations of CdCl₂ treatment, the values were found to be 10.55, 11.40 and 4.59 respectively (Table 3). Out of the 3 treatment conditions, maximum value was found with 0.01 ppm concentration and least with 0.5 ppm which shows that 0.5 ppm adversely affect the biomass.

The IG (% per day) of *N. punctiforme* in the control condition ranged from 4.28 (on 5th day) to 1.44 (on 20th day) while in dose of 0.01, 0.1 and 0.5 concentrations of CdCl₂ treatment showed the variation from 23.98 (on 5th day) to 3.2 (on 25th day), 22.76 (on 5th day) to 2.18 (on 20th day) and from 14.46 (on 5th day) to 2.32 (on 20th day) Table4.

**Discussion**

In the present study different concentrations of CdCl₂ (0.01, 0.1 and 0.5 ppm) showed a significant (F = 10.4946, df = 3, P < 0.001) decline in the biomass of *N.
The biomass turnover generally reflects the rate of replacement of individual organism and is considered, as an important ecological index, which can provide an insight into the functioning of the individual under certain conditions. In the present study, the turnover rate of *N. punctiforme* in the CdCl$_2$ treated condition was found maximum with 0.01 ppm and minimum with 0.5 ppm which reveals that 0.5 ppm of CdCl$_2$ treatment adversely affected the rate of replacement of *N. punctiforme*. A similar trend was observed with 0.1, 0.5 and 0.01 ppm concentrations of Butachlor treatment Patel (1997). The study revealed that lower concentration of Cadmium chloride provides better opportunity for biomass growth of the species.

**REFERENCES**


SPECIAL ISSUE OF THE BIOSCAN

The Editorial Board of The Bioscan is going to bring about special issue of the journal on

1. Physiology and Endocrinology
2. Ecological Productivity and Energetics

Interested Academicians, Researchers and Scientists are requested to contribute to the proposed issue. The pattern of preparation of manuscript will be the same as the instruction to the authors of this journal.