

## Effect of Cobalt on the Primary Productivity of *Spirulina platensis*

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Cobalt, a micronutrient for biological organisms, is a metal of wide use (Jenkins 1980). It is used in the production of aircrafts, electromagnets, paints, ceramics, chemicals etc. Main sources of Co to the environment are combustion of fossil fuels, smelters, cobalt processing facilities, sewage and industrial wastes (Jenkins 1980). Atomic power plants and nuclear weapon detonations form an important source of radioisotopes of this metal to the environment (Lowman and Ting 1973; Young and Folsom 1973; Smedile and Queirazza 1975; Bastian and Jackson 1975). Wood (1974, 1975) has classified Co in the group of "very toxic and relatively accessible elements". Jenkins (1980) has included cobalt in the 14 toxic trace elements of critical importance from the point of view of environmental pollution and health hazards. Cobalt deficiency leads to diseases like stunted growth. At toxic level, Co inhibits heme biosynthesis (Tephly et al. 1978) and enzyme activities (Olson and Christensen 1982).

The present study reports the effect of cobalt on biomass productivity of blue-green alga *Spirulina platensis*.

### MATERIALS AND METHODS

Pure culture of *Spirulina platensis* obtained from Indian Agricultural Research Institute, New Delhi, was grown in artificial aqueous medium (Table 1). The experiment was conducted in 15 mL screw cap test tubes. The tubes were kept at a temperature of  $21.0 \pm 1.0^\circ\text{C}$  and light intensity of 2500 lux with light and dark cycle of 16 and 8 h respectively. The toxicant was added as  $\text{CoCl}_2 \cdot 6\text{H}_2\text{O}$ . The concentrations used were 0.1, 0.5, 1.0, 2.0, 4.0, 6.0, 8.0, 10.0, 12.0 and 14.0 mg/L. A metal-free control was also used. The experiment was run in triplicates. Optical density (OD) at 490 nm was measured at intervals of 24 h. The OD to dry weight biomass conversion was performed by the relation  $B = 7.57 + 800.77 \text{ OD}$ , where B = biomass dwt/L.

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Table 1. Chemical composition of the culture medium

Compound	Conc. g/L	A5 solution g/L	
NaHCO <sub>3</sub>	18.0	H <sub>3</sub> BO <sub>3</sub>	2.9
K <sub>2</sub> HPO <sub>4</sub>	0.5	MnCl <sub>2</sub>	1.8
NaNO <sub>3</sub>	2.5	ZnCl <sub>2</sub>	0.11
K <sub>2</sub> SO <sub>4</sub>	1.0	CuSO <sub>4</sub>	0.08
NaCl	1.0	(NH <sub>4</sub> ) <sub>2</sub> MoO <sub>4</sub>	0.18
MgSO <sub>4</sub>	0.2		
CaCl <sub>2</sub>	0.04		
FeSO <sub>4</sub>	0.01		
A5 solution	1.0 mL/L		

EC<sub>50</sub> was calculated by probit analysis (Finney 1971). TLM values (the time at which the biomass of the experimental culture reduced to 50% of the corresponding control biomass) were computed by plotting the survival ratio (survival ratio =  $x'/x$  where  $x'$  and  $x$  are the algal biomass in the experimental and control cultures respectively at time  $t$ ) against the time in hours. To find the variation in toxic action of element with time, the data up to 168 h were divided into two halves, 24–96 h and 96–168 h and the slopes of best fit curves were determined by linear regression analysis.

## RESULTS AND DISCUSSION

Within the time interval of 168 h, the metal-free control biomass increased to  $304.9 \pm 8.4\%$  of the initial biomass (B<sub>to</sub>) (Figure 1). The initial biomass was  $53.5 \pm 1.5$  mg dry wt/L. With different metal concentrations increasing from 0.1 mg/L onwards, a gradual decrease in biomass was seen. There was no significant difference ( $p > 0.1$ ) in final biomass in case of 0.1 and 0.5 mg/L of Co. The biomass with these concentrations were  $289 \pm 10.2$  and  $289.3 \pm 10.4\%$  respectively of B<sub>to</sub>. The initial biomass was  $56.1 \pm 3.3$  and  $58.0 \pm 2.0$  mg dwt/L in case of 0.1 and 0.5 mg/L Co respectively. A peculiarity observed in case of biomass at 168 h was higher reduction with 8.0 and 10.0 mg/L than higher concentrations used (Figure 2). The observed biomass at 168 h were  $118.9 \pm 9.9$ ,  $118.9 \pm 4.6$ ,  $123.3 \pm 3.4$  and  $145.2 \pm 6.2\%$  with 8.0, 10.0, 12.0 and 14.0 mg/L, respectively. Higher values in biomass were observed in case of concentrations 0.1 and 0.5 mg/L than the control at 72–144 h time interval (Figure 1). This phenomenon was not observed at 24, 48 and 168 h.

Increase in toxic action with increase in metal concentration is clear from the changes in the slope of best fitting curves of 24–96 h (For regression coefficients  $r$  of the survival ratios to  $h$  curves, see Table 2). The second half of the experiment also showed the same trend of increasing toxicity with increasing concentrations (Table 2). An increase in toxicity from

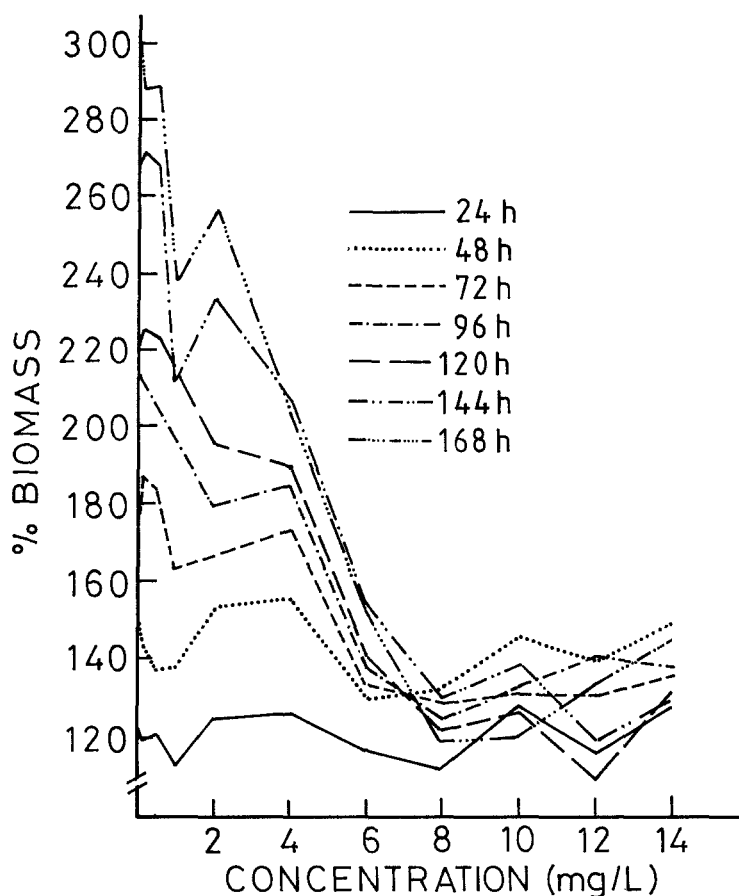


Figure 1. Change in biomass of *S. platensis* with different concentrations of cobalt.

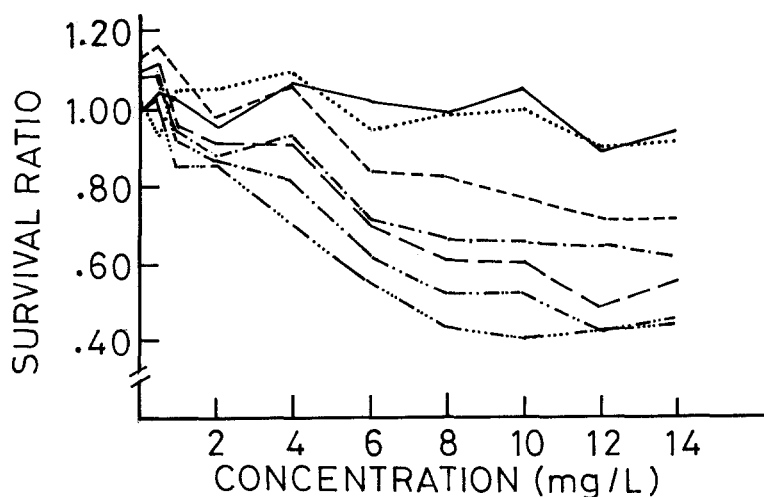


Figure 2. Survival ratio of *S. platensis* with different concentrations of cobalt.

the first to second half was seen for lower concentrations below 4.0 mg/L. All other concentrations showed the same level of toxic action in second half also (96-168 h). A trend of change from stimulatory to inhibitory action is seen with 0.1 and 0.5 mg/L of Co, the regression coefficients, which were positive during first half became negative during second half (Table 2).

Table 2. Regression coefficients (r) of survival ratios and hours with different concentrations of Cobalt

Cobalt concentrations (mg/L)	24-96* h	96-168* h	24-168** h	TLm h
0.1	0.73	-0.84	0.02	-
0.5	0.46	-0.65	0.24	-
1.0	-0.43	-0.90	-0.86	-
2.0	-0.49	-0.70	-0.78	-
4.0	-0.80	-0.97	-0.96	261
6.0	-1.00	-0.98	-0.99	179
8.0	-0.95	-0.99	-0.99	148
10.0	-0.98	-0.98	-0.98	143
12.0	-0.93	-0.89	-0.97	131
14.0	-0.96	-0.97	-0.98	137

\*r = 0.55, p < 0.05 (number of observations 12). \*\*r = 0.42 p < 0.05 (number of observations 21).

The observed TLm for different concentrations from 4.0 mg/L onwards were 261 and 131 h. The lowest value was observed at 12.0 mg/L. A slightly higher TLm was observed in case of 14.0 mg/L (137 h) due to slight increase in biomass observed during the later half of the experiment.

The estimated EC50's were  $8.13 \pm 0.93$ ,  $10.9 \pm 1.5$ ,  $14.4 \pm 2.1$  and  $23.8 \pm 6.4$  mg/L for 168, 144, 120 and 96 h, respectively.

Cobalt is absorbed and accumulated in very low or limited amounts by biological organisms (Jenkins 1980). Variations with species on toxicity and uptake of Co by algae has been reported by Coleman et al (1971). They reported that Co above 0.04 mg/mL reduces significantly the growth of the three species tested, viz., Pediastrum tetras, Euglena sp. and Chlorella vulgaris. The study reported here showed that even 0.1 mg/L Co during early hours of an experiment inhibited growth. But by 72 h onwards, 0.1 and 0.5 mg/L resulted in higher biomass. The phenomena of increase in biomass with sublethal concentrations of Cu, Ni and Cd was also observed by present workers on the same species of alga. This phenomenon can be ascribed to the hormetic effect of sublethal concentrations of metal on the species. Similar observations has been made on few other

species of algae (e.g., Deviprasad and Deviprasad 1982; Azeez and Banerjee 1986).

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