Bull. Environ. Contam. Toxicol. (1990) 45:232–234


Bulletin of Environmental Contamination and Toxicology

# Effect of Benthiocarb and Butachlor on Growth and Nitrogen Fixation by Cyanobacteria

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The vast source of biological nitrogen in cyanobacterial form offers a unique opportunity to harness them for maximum agricultural production. The use of vast array of pesticides to overcome the economic losses exerts varied environmental stress on non-target organisms present in the soil. Various workers have studied the effects of some pesticides on the growth, nitrogen fixation etc. in cyanobacteria (El-Sawy et al 1984; Goyal 1986; Dar and Zargar 1988). Most of the studies have been conducted with a single culture, however, the present study highlights the effects of two extensively used herbicides, benthiocarb and butachlor on the growth, nitrogen fixation chlorophyll 'a' content and heterocyst formation in a mixed culture of Anabaena,Nostoc and Oscillatoria species which are recommended for paddy field inoculation.

## MATERIALS AND METHODS

Anabaena, Nostoc and Oscillatoria species were isolated from local paddy field and maintained on Fogg's nitrogen free medium. Equal proportions of the above mentioned organisms were pooled to form a mixed culture. The experiment was carried in triplicate in 100 ml erlenmeyer flasks containing 50 ml of Fogg's nitrogen free medium ( Fogg 1949). Benthiocarb [8-(4-chlorobenzyl) N, N-diethyl thiocarbamate] @ 35, 45 and 55 ppm and butachlor (2-chloro-2', 6' diethyl-N-butoxymethy lacetanilide) @ 70, 90 and 110 ppm were added to these flasks; 2 ml of 7 days old mixed culture was inoculated in each flask and the flasks were kept under fluorescent tube light at a light intensity of 1500 lux at 25±2°C for 8 hrs daily for three weeks. Dry weight of the culture was determined after filtering and oven drying at 60°C for 24 hrs. Nitrogen estimation in the culture fluid was done by Microkjeldahl's method and chlorophyll 'a' content was measured spectrophotometrically at 665 nm after acetone extraction (Witham et al 1971). Effect on heterocyst differentiation was determined by microscopic examination.

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## RESULTS AND DISCUSSION

Inhibitory effect of the two herbicides was quite marked at higher concentrations. 55 ppm of benthiocarb and 110 ppm of butachlor had a drastic reduction in the dry biomass yield (Table 1). At 35 ppm of benthiocarb and 70 ppm of butachlor nitrogen content, dry biomass yield, chlorophyll 'a' content and heterocyst differentiation registered a negligible variation. However, further increase in the herbicide concentration reduced nitrogen (20-38%), chlorophyll 'a' content (35-46%), dry biomass (23-37%) and heterocyst formation (14-47%).

Table 1. Effect of benthiocarb and butachlor on cyanobacteria.

| Treatment | Conc. used ppm | Dry biomass yield mg/100 ml | Nitrogen mg/100 ml | Chlorophyll 'a' (mg/100 ml) ±0.02 | Heterocyst Ana- baena | Nostoc |
|---|---|---|---|---|---|---|
| Control | 0 | 60 | 3.20 | 0.68 | 15 | 12 |
| Benthiocarb | 35 | 64 | 3.20 | 0.69 | 15 | 12 |
| | 45 | 50 | 2.56 | 0.44 | 13 | 12 |
| | 55 | 38 | 2.00 | 0.38 | 9 | 7 |
| Butachlor | 70 | 66 | 3.17 | 0.68 | 14 | 12 |
| | 90 | 54 | 2.37 | 0.42 | 12 | 11 |
| | 110 | 40 | 2.11 | 0.37 | 8 | 7 |

Microscopic examinations showed a reduction in the populations of Anabaena and little effects on Oscillatoria in the samples treated with benthiocarb 45 and 55 ppm. Butachlor 70 ppm caused a slight decline in Oscillatoria population whereas at 90 and 110 ppm Oscillatoria and Anabaena registered considerable reduction but Nostoc remained unaltered. Our results corroborate with the results from the effects of insecticides and weedicides like Zineb, BHC, Stam F-34, Trifluralin, 2,4-D on the growth, heterocyst differentiation and nitrogen fixation in cyanobacteria (Tyagi 1973; Das and Singh 1977; Zeitseva 1979; Padhy 1985).

**Acknowledgments.** The authors are thankful to late Dr. Ghulam-uddin for extending the necessary facilities for the present study.

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**Received February 17, 1989; accepted March 10, 1990.**