

# Effect of insecticide Malathion on *Nostoc humifusum* Carm. ex Born. et Flah., a rice field cyanobacterium

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**ABSTRACT:** Cyanobacteria are potential biofertilizer for rice cultivation. All paddy fields have a natural population of cyanobacteria that provides fixed nitrogen at no cost. Modern agriculture is heavily dependent on agrochemicals which are used immensely on rice crops to increase the yield. However, the use of these chemicals over the years severely affected non-target organisms especially cyanobacteria. The present study was focused on the effect of Malathion, a commonly used insecticide for paddy on *Nostoc humifusum* which is an abundantly occurring cyanobacterium in rice fields of Kerala. The cyanobacterium was isolated from paddy fields of Thrissur District, Kerala. Growth response, pigment content (chlorophyll-a and carotene), protein, carbohydrate content and ammonia excretion were estimated at different doses of pesticide treatments at 7 days interval upto 42 days under laboratory conditions. The growth was significantly reduced in all the treatments. The pigment was seen induced at a dose 20ppm but inhibited in all other treatments. There was significant reduction in carbohydrate and protein content in all the treatments. Ammonia excretion decreased as the concentration of Malathion increased. Hence it is very essential to monitor the effect of agrochemical residue present in the soil as they inhibit the biofertilizer potential of cyanobacteria.

**Key Words:** Cyanobacteria, Biofertilizer, *Nostoc*, Chlorophyll-a, Malathion

## INTRODUCTION

Cyanobacteria are a diverse group of gram negative photosynthetic prokaryotes. They are one of the potential organisms, which are useful to mankind in several ways. Cyanobacteria are capable of fixing atmospheric nitrogen and thus they contribute greatly to the nitrogen economy of various habitats (Chaurasia, 2014). Paddy fields are one of the most favourable niches for the growth and proliferation of cyanobacteria (Whitton, 2000). There is a natural population of cyanobacteria in most paddy fields which provides a potential source of nitrogen fixation at no cost (Mishra & Pabbi, 2004). The paddy field ecosystem provides a suitable environment for the growth of cyanobacteria in terms of light, water, temperature and nutrient availability. Modern paddy cultivation is heavily dependent on the use of various agrochemicals. These chemicals which are meant to protect the crop plants affect the survival of a large number of beneficial organisms in the paddy fields. Cyanobacteria comes first in the list of non target organisms that are affected by the agrochemicals applied in the paddy fields. It is estimated that out of the total amount of pesticides applied in a field, only 1% is taken up by the target pest while the remaining part percolates into the soil affecting the microflora of which cyanobacteria constitute a major part (Gupta & Gupta, 1980).

The affect of agrochemical residue present in the soil on cyanobacteria depends on the dose, period of exposure and individual characteristics of the organisms. The aim of this study was to determine the toxicity effects of an insecticide, Malathion on a moderately toxic non-systemic wide spectrum organophosphate insecticide widely used in paddy fields on the cyanobacterium *Nostoc humifusum* isolated from paddy fields of Thrissur district, Kerala.

## MATERIALS AND METHODS

### Test organism

*Nostoc humifusum* Carmichael ex Bornet & Flahault (CU 139243), a heterocystous cyanobacterium (fig. 1) was collected from paddy fields of Thrissur district, Kerala. Pure cultures were obtained by standard plating and streaking techniques. Pure colonies were transferred to BG11 (Rippka et al. 1979) liquid medium and incubated under controlled laboratory conditions.

### Insecticide treatment

Different Malathion treatments (20,40,60,80 & 100 ppm respectively) were used for the study. Cyanobacterial culture without any insecticide treatment was used as the control.

## Test methods

### 1) Growth Measurement

The growth was measured by reading the absorbance of the homogenized culture suspension at a wave length of 680 nm in a spectrophotometer.

### 2) Estimation of Chlorophyll-a

The chlorophyll content was estimated according to Mackinney (1941) by taking the absorbance at 663nm

### 3) Estimation of Carotenoid

The carotenoid content was measured by reading the absorbance at 450nm and was estimated following the method of Siegelman & Kycia (1979).

### 4) Estimation of total carbohydrates

Total carbohydrate was estimated according to Spiro (1996) by measuring the absorbance at 620 nm.

### 5) Estimation of protein

The protein content was estimated following the method of Herbert et al. (1971) by taking the absorbance of the sample at 650 nm.

### 6) Estimation of ammonia

Ammonia in the culture was estimated by Solorzano (1969) by measuring the absorbance at 640 nm.

### 7) Statistical evaluation

The experiments were set up in triplicates for each treatment and data were presented as means of triplicates with standard deviation.

## RESULTS

There was a considerable reduction in growth of *Nostoc humifusum* at different concentrations of Malathion (fig. 2). The growth of all the treated cultures were lower than the control. However all the cultures showed an increasing trend in growth on all days of incubation.

Eventhough the chlorophyll-a content in all the Malathion treated cultures was lower than the control culture, there was an increasing trend in chlorophyll-a content in all the cultures on all days of incubation. There was a sudden increase in chlorophyll-a content in control and 20 ppm treated cultures after 14 days of incubation (fig. 3). In 40 ppm treated culture the sudden increase in chlorophyll-a content was found after 28 days of incubation. In 80 ppm and 100 ppm treated cultures, chlorophyll-a content showed a sudden increase after 28 days followed by a decrease after 35 days of incubation.

Above 40 ppm Malathion concentration, the carotenoid content was very much reduced. On the 35<sup>th</sup> day the carotenoid content was shown a sudden increase in the 20ppm and 40 ppm treated cultures and reached a value higher than the control and then reduced (fig. 4).

Up to 60 ppm concentration of Malathion carbohydrate content was enhanced in all days of incubation and beyond 60 ppm the carbohydrate content was reduced after 21 days of incubation (fig. 5).

Protein content was seen enhanced only at 20 ppm treated culture (fig. 6). At 40 ppm concentration protein content was reduced after 21 days of incubation. In all the treated cultures above 40 ppm concentration of Malathion, the protein content slightly increased after 14 days of incubation and decreased after 21 days of incubation.

Up to 35 days of incubation, ammonia excretion at 20 ppm concentration of Malathion was greater than the control, later it decreased and reached below the control culture. In all the treated culture maximum ammonia excretion was marked on the 28<sup>th</sup> day of incubation (fig. 7). After 28<sup>th</sup> day of incubation, the ammonia excretion was seen decreasing in all the treated cultures.

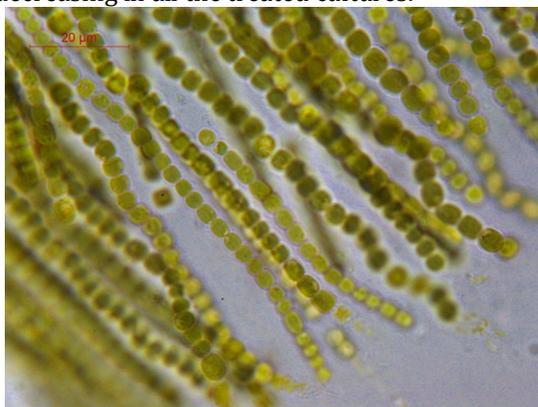


Fig.1 *Nostoc humifusum*

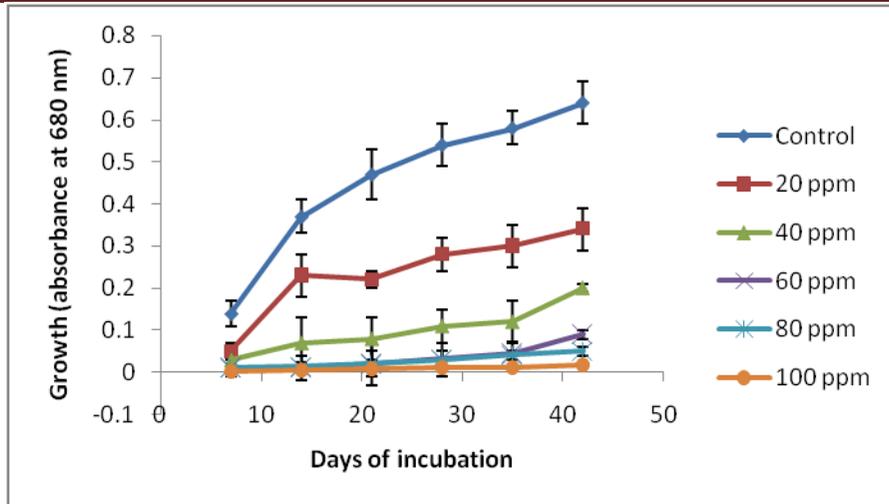


Fig 2. Effect of Malathion on growth of *Nostoc humifusum*

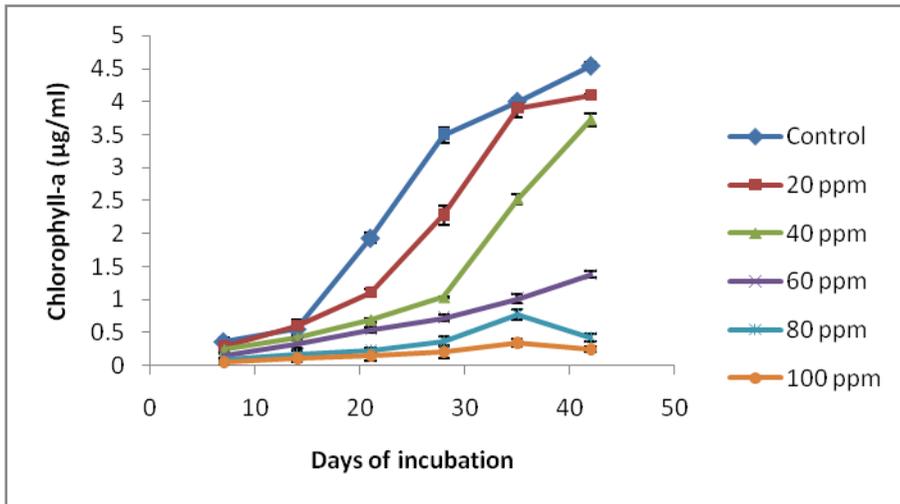


Fig 3. Effect of Malathion on chlorophyll-a content of *Nostoc humifusum*

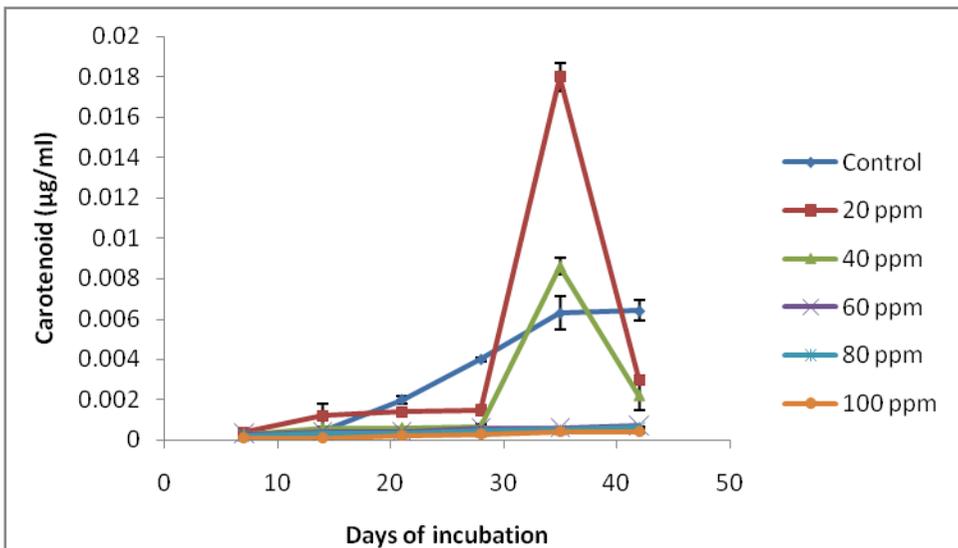


Fig 4. Effect of Malathion on carotenoid content of *Nostoc humifusum*

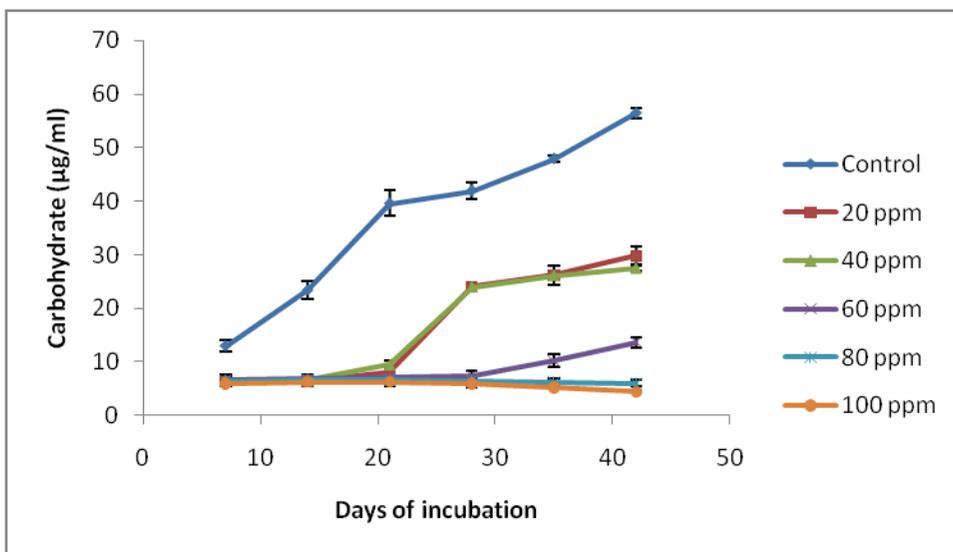


Fig 5. Effect of Malathion on carbohydrate content of *Nostoc humifusum*

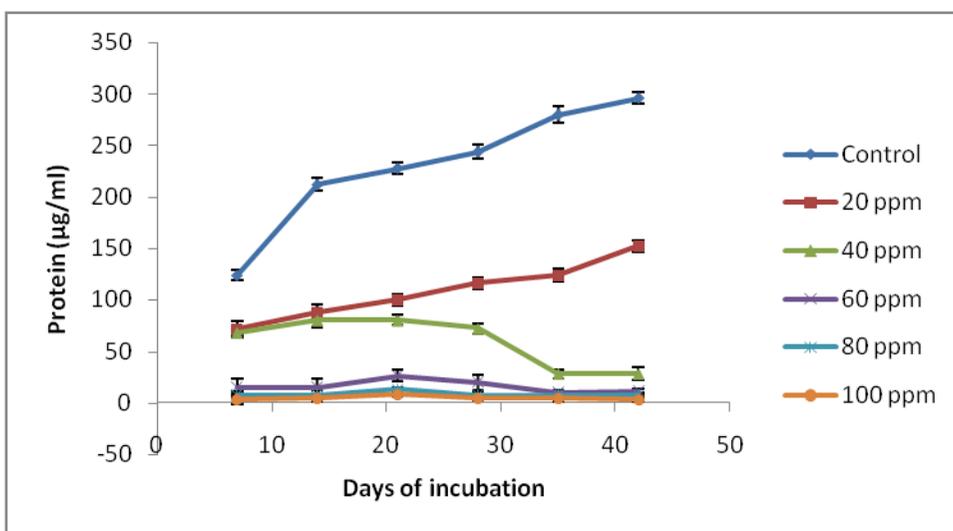


Fig.6. Effect of Malathion on protein content of *Nostoc humifusum*

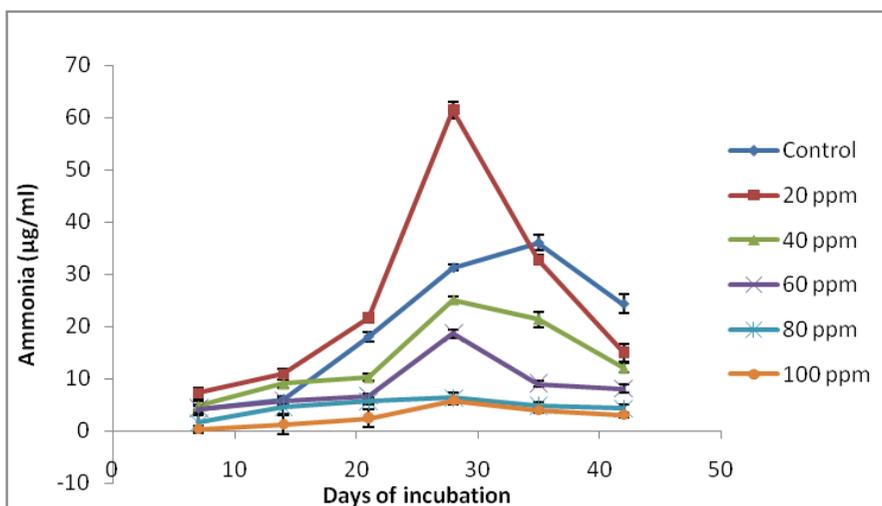


Fig.7. Effect of Malathion on ammonia excretion of *Nostoc humifusum*

## DISCUSSION

Malathion had a partial inhibitory effect on the growth of cyanobacteria (Lal & Lal, 1988). The results obtained from the present study reveals that higher and continuous use of Malathion causes detrimental effect on *Nostoc humifusum*. There is an inverse correlation between Malathion concentration and the cyanobacterial growth (Mohapatra & Mohanty, 1992). Growth, chlorophyll-a, carbohydrate and protein content was higher in control culture as compared to the insecticide treated cultures. The inhibitory effect of Malathion may be due to the adsorption of this compound on the plasma membrane of the cyanobacterial cells, thus altering the membrane permeability (Rioboo et al. 2002) and diminishing photosynthetic activity. The continuous and higher doses of Malathion had affected the total carotenoid content and ammonia excretion. From the present investigation it is clear that *Nostoc humifusum* cannot tolerate a higher concentration of Malathion. The indiscriminate use of insecticide on cyanobacterial population has been considered to be inhibitory at high doses (Panigrahi et al. 2003).

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