

# Effects of Insecticide Malathion on growth, biochemical compounds and some enzymes of *Fischerella muscicola* NDUPC001

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## Abstract

Malathion effected the growth, biochemical compounds (Chl-a, Total protein and Carbohydrate) and enzymes (nitrate reductase and glutamine synthetase) of *Fischerella muscicola* NDUPC001. 100ppm was LC<sub>50</sub> concentration and 50ppm, 100ppm and 150ppm were treatment concentrations. All the treatment concentrations of insecticide inhibited the growth of cyanobacteria. 50 ppm treatment conc. induced the chl.-a and protein content 4.33% and 8.83% respectively whereas other treatments decreased the chl.-a and protein content with max. the decrease of 41.14% and 24.19% respectively in 150 ppm treatment. Carbohydrate content decreased in all treatments with a maximum decrease of 41.71% in 150ppm concentration. Nitrate reductase activity slightly induced (0.31%) in 50ppm treatment and decreased in the other two treatments with a maximum decrease of 27.83% in 150ppm treatment. All treatment concentrations of insecticide inhibited the activity of Glutamine synthetase with maximum inhibition of 34.98% in 150ppm treatment. Cyanobacterial strain showed high tolerance range and lesser inhibitory effects on growth, biomolecules and enzymes.

**Keywords:** Cyanobacteria, *Fischerella muscicola* NDUPC001 and Malathion

## 1. Introduction

Cyanobacteria (Blue-green algae) are Gram's negative, ancient, photosynthetic prokaryote. Cyanobacteria are capable of colonizing nearly all habitats on our planet (Whitton & Potts 2000). Role of cyanobacteria in soil genesis and conservation is well established. They constitute the initial successional state on nutrient-poor substrate soil. The algal growth consolidates at the surface leading to the formation of soil crusts, which improves infiltration, limits sheet erosion and provides a substratum suitable for seeds of higher plants for

germination. Photosynthetic nitrogen-fixing cyanobacteria are known to play an essential role in soil fertility especially in the waterlogged paddy fields (Singh, 1961). Now, it is a fact that cyanobacteria are one of the most important sources of biological nitrogen fixers in the paddy field. The paddy field ecosystem provides a favorable environment for the luxuriant growth of the cyanobacteria, concerning light, high water temperature, nutrient availability, etc. This may be reason for the higher abundance of cyanobacteria in rice-field soils rather than other cultivated soils (Watanabe et al., 1971) as reported under widely different climatic conditions in India (Mitra, 195).

The reduction of crop losses is the primary goal of agriculture. Use of synthetic chemicals in agriculture have played a fundamental role in suppressing pests and maintaining high crop yields. The world trade of pesticides in 1999 amounted to more than \$22 billion, of which 21% was for insecticides (Food and Agriculture Organization of United Nations, FAOSTAT-Agriculture\\Data.://apps.fao.org). Many experimental findings showed the adverse effects of pesticides on heterocystous cyanobacteria (Mahapatra, et al. 2003; Galhano et al. 2008). Thiodan and Phorate (insecticides) inhibited the growth, biochemical composition, nitrogenase and glutamine synthetase activity at the EC<sub>50</sub> concentration (Debnath et al. 2012). Insecticide Profenofos decreased the growth and biomolecules of *Anabaena* sp. (Chaurasia, 2014). Insecticide endosulfan inhibited the growth of *Anabaena fertilissima*, *Aulosirafertilissima*, *Westiellopsis prolific* (Kumar et al., 2012), *Nostoc calcicola* and *Nostoc muscorum* (Prasad et al., 2011). Insecticide malathion is frequently used in agricultural fields of Varanasi. *Fischerella muscicola* NDUPC001 is one of the most abundant cyanobacteria of agricultural fields of Varanasi and report about the effects of malathion on this cyanobacterial strain is lacking.

Hence, this experiment was designed to study the effects of malathion on growth, biochemical compounds and some enzymes of *Fischerella muscicola* NDUPC001.

## 2. Materials and Methods

### 2.1 Cultivation of cyanobacteria and Pesticide Treatment

*Fischerella muscicola* NDUPC001 was isolated from agricultural soils of Varanasi, India, following standard procedure and characterized by morphological and molecular means (16 rRNA gene, Accession No. JX912574). The strain was deposited at NAIMCC (NBAIM), Mau, India (Accession No. NAIMCC-C000121). cyanobacterium was grown in nitrogen-free, BG-11 liquid medium (Stanier, 1971) in a culture room maintained at a temperature of  $28 \pm 2^{\circ}$  C and illuminated with fluorescent light of  $12Wm^2$ .

Different concentrations of insecticide malathion were screened to determine  $EC_{50}$ . 100 ppm insecticide concentration was  $EC_{50}$  ( Table-1 ). 50ppm, 100ppm and 150ppm levels of insecticide were decided for treatment and untreated cyanobacterial culture was control (Table-1).

### 2.2 Growth and Biochemical Analysis

The growth of homogenous cultures was measured turbidometrically at 700nm in spectrophotometer-117(Systronic). Chlorophyll-a. total carbohydrate and total protein content were measured by Myers and Kratz ( 1955) method, Phenol-sulphuric method (Dubois et al., 1956) and Method of Lowry et al., (1951) respectively.

### 2.3 Enzymatic study

Nitrate reductase (NR) and Glutamine synthetase (GS) activity were determined by methods of Snell and Snell (1949) and Method of Shapiro and Stadtman (1970) respectively. The activity of nitrate reductase was expressed as  $\mu M NO_2$  formed  $mg chl^{-1} min^{-1}$  and the activity of Glutamine transferase was expressed as mMoles glutamyl hydroxamate produced  $mg chl^{-1} min^{-1}$ .

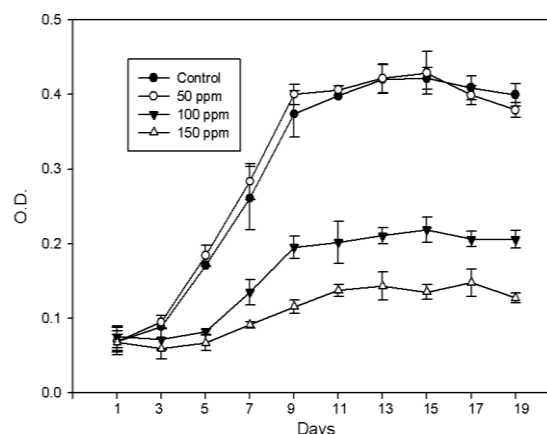
## 3. Results and Discussion

Malathion (an broad-spectrum, organophosphate insecticide) is used to control a variety of outdoor insects in both agricultural and residential settings

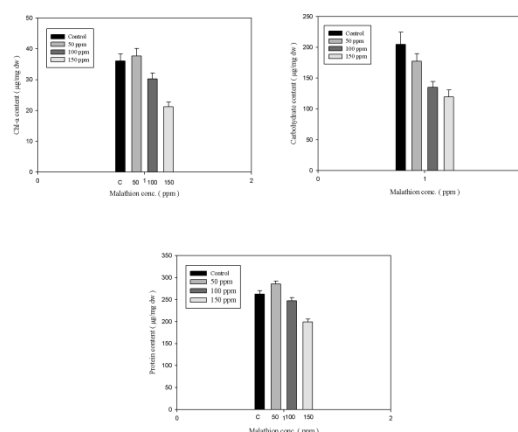
**Table-1:  $LC_{50}$  value of pesticide Malathion**

| Pesticide | Organism                              | $LC_{50}$ (ppm ) | Treatment concentrations ( ppm) |
|-----------|---------------------------------------|------------------|---------------------------------|
| Malathion | <i>Fischerella muscicola</i> NDUPC001 | 100              | 50                              |
|           |                                       |                  | 100                             |
|           |                                       |                  | 150                             |

Effects of insecticide malathion on growth, biochemical composition (Chl-a, Total protein, and Carbohydrate) and some enzymes (Nitrate reductase and Glutamine synthetase) of *Fischerella muscicola* NDUPC001 was studied.  $LC_{50}$  value of malathion was 100 ppm (Table-1). 50 ppm, 100 ppm and 150 ppm were treatment concentrations (Table-1). All concentrations of insecticide inhibited the growth of cyanobacterium (Fig.-1). Maximum inhibition was observed in 150 ppm treatment. Growth was slightly induced in exponential phase in 50 ppm but later on, decreased in stationary phase. Other insecticides also decreased the growth of cyanobacteria. The growth of cyanobacteria i.e. *Nostoc ellipsosporum*, *Scytonema simplex*, *Tolypothrix tenuis*, and *Westiellopsis prolifica* was reduced upto 50% by insecticides thiodan and phorate treatment in the concentration ranging from 0.025 to 0.05 ppm and from 0.40 to 0.80 ppm respectively (Debnath et al., 2012). The growth of *Anabaena fertilissima*, *Aulosira fertilissima*, *Westiellopsis prolifica* (Kumar et al., 2012), *Nostoc calcicola* and *Nostoc muscorum* (Prasad et al., 2011) was decreased by insecticide endosulfan treatment.

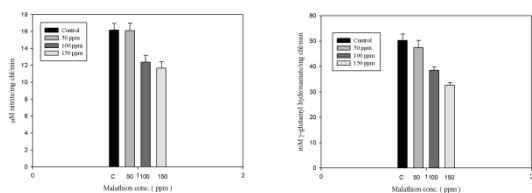


**Fig.1** Growth behavior of *Fischerella muscicola* NDUPC001 in response to different concentrations of Malathion. Values are mean of triplicate  $\pm$  S.D., bars indicate standard deviation.



**Fig.2** Effect of Malathion on biochemical composition of *Fischerella muscicola* NDUPC001. Values are mean of triplicate $\pm$ S.D. bars indicate standard deviation.

Effect of all treatments on biochemical composition (Chl-a, Total protein, and Carbohydrate) of *Fischerella muscicola* NDUPC001 was studied. 50 ppm treatment conc. induced the chl.-a and protein content 4.33% and 8.83% respectively whereas other treatments decreased the chl.-a and protein content with max. Decrease of 41.14% and 24.19% respectively in 150 ppm treatment ( Fig.2). Carbohydrate content decreased in all treatments with a maximum decrease of 41.71% in 150 ppm concentration (Fig.2). A similar pattern of the result was observed with other insecticide treatments on cyanobacteria. Chl-a content of *Anabaena fertilissima*, *Aulosira fertilissima* and *Westiellopsis prolifica* was decreased on endosulfan treatment (Kumar et al., 2012). Carbohydrate content of *Anabaena fertilissima*, *Aulosira fertilissima* *Westiellopsis prolifica* was reduced up to 97 % on endosulfan treatment (Kumar et al., 2012). Lower concentrations of endosulfan increased the total protein content of *Aulosira Fertilissima*, *Anabaena variabilis* and *Nostoc muscorum* whereas higher concentrations decreased the protein content (Kumar, et al., 2008). Increase in total protein content may be due to the formation of stress proteins.



**Fig. 3** Effects of Malathion on some enzymes of nitrogen metabolism of *Fischerella muscicola* NDUPC001. Values are mean of triplicate $\pm$ S.D. bars indicate standard deviation.

Effects of treatment on the activity of nitrate reductase and glutamine synthetase were studied. Nitrate reductase activity slightly induced (0.31%) in 50 ppm treatment and decreased in the other two treatments with a maximum decrease of 27.83% (Fig.3) in 150 ppm treatment. All treatment concentrations of insecticide inhibited the activity of Glutamine synthetase with maximum inhibition of 34.98% in 150 ppm treatment (Fig.3). NR activity of *Anabaena fertilissima*, *Aulosira fertilissima* and *Westiellopsis prolifica* was decreased by 77%, 90% and 95% respectively on endosulfan treatment (Kumar et al., 2012). GS activity of *Anabaena fertilissima*, *Aulosira fertilissima* and *Westiellopsis prolifica* was inhibited by endosulfan treatment (Kumar et al., 2012). GS activity of *Nostoc hatie* was significantly decreased by endosulfan and chloropyriphos treatment (Jha and Mishra, 2005). GS

activity of *Tolypothrix tenuis* was inhibited by 62% on phorate treatment (Debnath et al., 2012). Overall findings of the experiment suggest the high tolerance range, lesser inhibitory effects on growth, biomolecules (Chl-a, Total protein, and Carbohydrate) and enzymes (Nitrate reductase and Glutamine synthetase) of *Fischerella muscicola* NDUPC001 by insecticide malathion treatments. Hence, *Fischerella muscicola* NDUPC001 may prove a suitable cyanobacterial inoculum for algalization of rice fields of Varanasi, India.

## 4. Conclusion

A score of pesticides are regularly poured in agricultural fields to minimize damage from pests and insecticides are one of them. Insecticides produce adverse effects on beneficial microbes of soil including cyanobacteria. Insecticide malathion is used frequently in rice fields of Varanasi. *Fischerella muscicola* NDUPC001 showed the high tolerance range, lesser inhibitory effects on growth, biomolecules (Chl-a, Total protein, and Carbohydrate) and enzymes (Nitrate reductase and Glutamine synthetase) to insecticide malathion treatments. Hence, cyanobacterial strain may be considered as one of suitable cyanobacterial inoculum for algalization of rice fields of Varanasi, India.

## Acknowledgement

University Grants Commission (UGC), New Delhi is acknowledged for providing financial assistance. Principal, U.P.College and Head, Dept. of Botany is acknowledged for providing laboratory facilities.

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