EFFECTS OF FUNGICIDE CM-75 ON GROWTH, BIOCHEMICAL COMPOUNDS AND SOME ENZYMES OF FISCHERELLA MUSCICOLA NDUPC001

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Abstract

Fischerella muscicola NDUPC001 was isolated from agricultural fields of Varanasi, India and characterized by morphological as well molecular means. The organism was deposited at NAIMCC (NBAIM), Mau, India (Accession No. NAIMCC-C-00121). Fungicide CM-75 inhibited growth, biochemical composition and some enzymes of Fischerella muscicola NDUPC00. LC₅₀ conc. of fungicide was 6ppm and 3ppm, 6ppm, and 9ppm were concentrations of treatments. All concentrations of CM-75 inhibited the growth of cyanobacteria, and maximum inhibition was observed in 9ppm treatment. Treatment concentrations of fungicide decreased the Chl.-a and carbohydrate content of cyanobacteria with maximum inhibition of 29.29% and 29.9% respectively in 9ppm treatment. Total protein content was slightly increased (2.52%) in 3ppm treatment and decreased in other treatments with maximum inhibition of 19.26% in 9 ppm treatment. Fungicide inhibited the activity of Nitrate reductase and Glutamine synthetase with maximum inhibition of 35.11% and 21.74% respectively in 9ppm. The findings of the experiment suggest that high tolerance level of Fischerella muscicola NDUPC001 against fungicide CM-75.

Key words: Cyanobacteria, Fischerella muscicola NDUPC001 and CM-75

Introduction

Cyanobacteria are capable of colonizing nearly all habitats on our planet (Whitton & Potts,s 2000) and are widely distributed from aquatic to the terrestrial environment. It has been established that cyanobacteria play a significant role in soil genesis and conservation. Cyanobacteria produce humus on the soil surface after their death and decay, and it dissolves certain soil minerals for maintaining a reservoir of the element in a semi available form for higher plants. The pertinent genera in soils include Anabaena, Cylindro spermum, Lyngbya, Microcoleus, Nodularia, Nostoc, Fischerella, Oscillatoria, Phormidium, Plectonema, scytonema, Synendra, and Tolypothrix, etc. Role of cyanobacteria in fertility of rice fields are well established. Paddy fields favor the luxuriant growth of cyanobacteria, and most of the biological nitrogen fixation of this ecosystem is done by N₂-fixing cyanobacteria (Irisarri et al., 2001). Many nitrogen-fixing strains of cyanobacteria have been isolated and used in biofertilizer consortia in Southeast Asian countries. Pesticides are regularly used in agricultural fields to decrease the loss due to pests. A score of fungi causes disease on plants throughout the world. Damage due to fungal diseases are one of the major problems of agriculture. Crop area of paddy is about 24% with pesticide use of 17.2% (Mamthanayagam and Sharinila, 2004). Use of fungicides increased from 10% ( 1996 ) to 21% (2000) amounting 10910 tons (Agnihotri, 2000). Fungicides besides controlling target fungi also have shown an adverse impact on beneficial microbes of soil including cyanobacteria. Fungicide zineb inhibited the growth of Cylindro spermum sp. and Nostoc muscorum at 1ppm and 5ppm respectively (Venkataraman and Rajyalakshmi, 1971). Soil fungicides nabam and vapan also showed varying degree of toxicity towards rice field cyanobacteria (Venkataraman and Rajyalakshmi, 1972). Captan decreased the growth of Westiellopsis prolifica, Aulosira fertilissima, Nostoc sp., Tolypothrix tenues, and Calothrix sp. (Gangawane and Saler, 1979a). The growth of Westiellopsis sp., Aulosira sp., Calothrix sp., Nostoc

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sp., and *Tolypothrix* sp. was decreased by treatment of Roval, a broad spectrum fungicide (Gangawane and Kulkami, 1979b). CM-75 is a broad spectrum systemic fungicide. It is used to controls leaf spot, rust diseases of groundnut, and blast disease of paddy crop. This fungicide is regularly applied in paddy fields of Varanasi. *Fischerella muscicola* NDUPC001 is one of most abundant cyanobacteria of agricultural fields of Varanasi and report on the effects of CM-75 on this cyanobacteria is lacking. Hence, This experiment was designed to study the effects of fungicide on growth, biochemical composition and some enzymes of *Fischerella muscicola* NDUPC001.

**Materials and Methods**

**Cultivation of cyanobacteria and Pesticide Treatment**

The cyanobacterium *Fischerella muscicola* NDUPC001 was isolated from agricultural soils of Varanasi, India, characterized by morphological method and confirmed by molecular means (16 rRNA gene, Accession No. JX912574). The strain was deposited at NAIMCC (NBAIM), Mau, India (Accession No. NAIMCC-C-000121). It was grown in nitrogen-free, BG-11 liquid medium (Stanier, 1971) in a culture room maintained at a temperature of 28 ± 2°C and illuminated with fluorescent light of 12 Wm².

Different concentrations of fungicide CM-75 was screened to determine EC₅₀ which is the treatment concentration of pesticide that reduces the growth of the sample population by 50% in comparison to control in a specified period of exposure. EC₅₀ value of fungicide was 6ppm (Table 1). 3ppm, 6ppm and 9ppm concentrations (Table 1) of fungicide were treatment concentrations. Effect of 3ppm, 6ppm and 9 ppm conc. of CM-75 on *Fischerella muscicola* NDUPC001 growth behavior was studied. All treatment concentrations of fungicide inhibited the growth of cyanobacteria (Fig. 1). Maximum inhibition was observed in 9ppm treatment. Growth was slightly induced in exponential phase in 3ppm but later on, decreased in stationary phase. A similar trend

**Table-1:** LC₅₀ value of pesticide CM-75

<table>
<thead>
<tr>
<th>Pesticide</th>
<th>Organism</th>
<th>LC₅₀ (ppm)</th>
<th>Treatment concentrations (ppm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>CM-75</td>
<td><em>Fischerella muscicola</em> NDUPC001</td>
<td>6</td>
<td>3</td>
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<td></td>
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**Results and Discussion**

CM-75 is a broad spectrum systemic fungicide and composed of Carbendazim 12 + Mancozeb 63%. It is used to control leaf spot, rust diseases of groundnut, and blast disease of paddy crop. This fungicide is applied regularly in paddy fields of Varanasi. Effects of fungicide on growth, biochemical composition and some enzymes of *Fischerella muscicola* NDUPC001 was studied. Different concentrations of fungicide CM-75 was screened for determination of EC₅₀ which is the treatment concentration of pesticide that reduces the growth of the sample population by 50% in comparison to control in a specified period of exposure. LC₅₀ value of fungicide was 6ppm (Table 1). 3ppm, 6ppm and 9ppm concentrations (Table 1) of fungicide were treatment concentrations. Effect of 3ppm, 6ppm and 9 ppm conc. of CM-75 on *Fischerella muscicola* NDUPC001 growth behavior was studied. All treatment concentrations of fungicide inhibited the growth of cyanobacteria (Fig. 1). Maximum inhibition was observed in 9ppm treatment. Growth was slightly induced in exponential phase in 3ppm but later on, decreased in stationary phase. A similar trend

**Fig. 1:** Growth behavior of *Fischerella muscicola* NDUPC001 in response to different concentrations of CM-75. Values are mean of triplicate ± S.D., bars indicate standard deviation.
Effects of fungicide CM-75 on growth, biochemical compounds and some enzymes


Most of the cyanobacteria showed tolerance limit of 5-6 ppm to fungicides but *Fischerella muscicola NDUPC001* have tolerance limit up to 9ppm (Fig. 1).

Effect of all treatments on biochemical composition (Chl-a, Total protein, and Carbohydrate) of *Fischerella muscicola NDUPC001* was studied. All treatment concentrations of fungicide decreased the Chl.-a, carbohydrate and protein content of cyanobacteria with maximum inhibition of 29.29%, 29.9%, and 19.26% respectively in 9ppm treatment (Fig. 2). However, the decrease in biomolecules content was lesser than other reported cyanobacteria. Bavistin decreased the chl-a of

![Fig. 2: Effect of CM-75 on biochemical composition (Chl-a, Carbohydrate, and Total protein) of *Fischerella muscicola NDUPC001*. Values are mean of triplicate ± S.D. bars indicate standard deviation.](image)

![Fig. 3: Effects of CM-75 on Nitrate reductase (NR) and Glutamine synthetase (GS) activity of *Fischerella muscicola NDUPC001*. Values are mean of triplicate ± S.D. bars indicate standard deviation.](image)
Tolypothrix scytonemoides (Rajendran et al., 2006). chlorophyll-a content of Anabaena fertilissima, Aulosira fertilissima and Westiellopsis prolific was decreased by fungicide Tebuconazole (Kumar et al., 2012). Carbohydrate content of cyanobacteria Anabaena fertilissima, Aulosira fertilissima Westiellopsis prolific decreased up to 94 %, 96% and 97% respectively by tebuconazole treatment (Kumar et al., 2012). Bavistin decreased the carbohydrate content of Tolypothrix scytonemoides (Rajendran et al., 2006). Total protein content was slightly induced in conc. below LC₅₀ treatment (Fig. 2) and decreased in the other two treatments. Induction of total protein content in lower conc. of treatment may be due to the formation of stress proteins. Some fungicide like Tebuconazole decreased the total protein content of Tolypothrix scytonemoides (Rajendran et al., 2006) and Anabaena fertilissima, Aulosira fertilissima, and Westiellopsis prolific up to 90 %, 95% and 93% respectively (Kumar et al., 2012).

All the treatment concentrations of CM-75 inhibited the Nitrate reductase (NR) and Glutamine synthetase (GS) activity of Fischerella musicicola NDUPC001 (Fig. 3). NR and GS activity of cyanobacteria was also decreased by fungicides treatment. NR activity of Anabaena fertilissima, Aulosira fertilissima, Westiellopsis prolific was decreased by 90%, 93% and 95% respectively on fungicide Tebuconazole treatment (Kumar et al., 2012). GS activity of N. ellipsosporum was decreased up to 70% on Bagalol treatment and up to 46% in W. prolific on Mancozeb treatment (Debnath et al., 2012). Bavistin treatment decreased the GS activity of Tolypothrix scytonemoides (Rajendran et al., 2006).

Fischerella musicicola NDUPC001 showed high tolerance range to fungicide CM-75 and amount of decrease in growth, biomolecules and enzymes (NR&GS) was lesser than other fungicides as per available reports. Hence, Fischerella musicicola NDUPC001 may be considered as suitable cyanobacterial inocula for agricultural fields of Varanasi.

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References


