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Efficacy of Micro Nutrients on *Banana fusarium* Wilt. (*Fusarium oxysporum* f. sp. *cubense*) and it's Synergistic Action with *Trichoderma viride*

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Abstract

In the present investigations, efficacy of micro nutrients viz., (calcium nitrate, ammonium sulphate, copper sulphate, potassium chloride, sodium chloride, borax, ferrous sulphate and zinc sulphate) against *Fusarium oxysporum* f. sp. *cubense* causing panama wilt of banana were studied. The results of experiment revealed that among the micro nutrients used, borax @ 500 and 750 ppm completely inhibited the mycelial growth of *Fusarium oxysporum* f. sp. *cubense* followed by zinc sulphate (47.3, 42.7, 31.0 and 14.3 mm at 100, 250, 500 and 750 ppm respectively). The incorporation of borax in solid medium at higher concentrations viz., 500 and 750 ppm., significantly decreased the mycelial growth of the *T.viride*, whereas the sporulation capacity of *T.viride* (207.48 x 108 spores/ sq.cm @ 500 ppm) was found to be maximised by the addition of borax. In general, the antagonist was compatible with borax at low concentrations. The maximum inhibition of mycelial growth of *F. oxysporum* f. sp *cubense* was found in borax and *T. viride* inoculated medium.

Keywords: micro nutrient, *Fusarium oxysporum* f. sp. *cubense*, *Trichoderma viride*

Introduction

Banana (*Musa* spp.) is one of the earliest crops cultivated by man which still remains to be one of the world's most important fruit crop. At present, it is grown in more than 120 countries throughout tropical and subtropical regions (Molina and Valmayor, 1999) and are the staple food for more than 400 million people. Since banana is being used as food, fiber and for medicinal, cultural and industrial purposes and also gives high returns to small holders it is referred to as "Kalpatharu"- a plant of virtues.

In order to cater to the needs of escalating population, banana production needs to be doubled and estimated production requirement by 2020 is around 25 million tones (Annon, 1996). Since increase in area of cultivation is impossible, the alternative approach is to increase the productivity is the threat posed by the pest. Among the diseases *Fusarium* wilt also known as panama wilt caused by *Fusarium oxysporum* f. sp. *cubense* is the major constraint to banana production and the disease has been ranked as No.1 fungal disease of banana in India. Disease reduction is the most often attributed to improved nutrition that boost the host defences or to direct inhibition of fungal growth and its activity. In this context, the present study has been undertaken with the aim of managing *Fusarium oxysporum* f. sp *cubense* with micronutrients and biocontrol agents.

Materials and methods

Effect of micronutrients on the mycelial growth of the Fusarium oxysporum f. sp. cubense

Potato Dextrose Agar (PDA) medium was amended with different micro nutrients viz., calcium nitrate, ammonium sulphate, copper sulphate, potassium chloride, sodium chloride, borax, ferrous sulphate and zinc sulphate at four different concentrations viz., 100, 250, 500 and 750 ppm (w/v) and they were tested on the mycelial growth of *Fusarium oxysporum* f. sp. *cubense* using by poisoned food technique following the procedure of Schmitz, (1930) the suitable control was also maintained without nutrients.

Effect of borax on the mycelial growth and sporulation by T. viride

The mycelial growth of *T. viride* was studied in PDA medium containing borax at different concentrations viz., 100, 250, 500 and 750 ppm. The mycelial disc of *T. viride* was placed in the middle of the Petri plate containing borax incorporated medium and incubated at room temperature. The mycelial growth was measured at regular intervals. The sporulation of *T. viride* was determined by counting the number of spores in one square cm area using haemocytometer from each plate, the number of conidia was counted randomly (Roja, 2006).

Table 1 Effect of certain micro nutrients on the mycelial growth of *Fusarium oxysporum* f. sp. *cubense*

Sl. No	Micro nutrients	Mycelial growth (mm)				Percent inhibition			
		100 ppm	250 ppm	500 ppm	750 ppm	100 ppm	250 ppm	500 ppm	750 ppm
1.	Calcium nitrate	86.0	80.3	82.4	78.3	4.44	10.78	8.44	13.00
2.	Ammonium sulphate	82.5	85.4	83.3	80.7	8.33	5.11	7.44	10.33
3.	Copper sulphate	81.2	75.7	74.3	71.2	9.78	15.88	17.44	20.89
4.	Potassium choride	73.5	70.3	69.2	69.0	18.33	21.89	23.11	23.33
5.	Sodium chloride	30.2	25.0	5.7	0.0	66.44	72.22	93.67	100.00
6.	Borax	18.5	10.3	0.00	0.0	79.44	88.56	100.00	100.00
7.	Ferrous sulphate	48.5	45.0	44.3	30.7	46.67	50.00	50.78	65.89
8.	Zinc sulphate	47.3	42.7	31.0	14.3	47.44	52.56	65.56	84.11
9.	Control	90.0	-	-	-	-	-	-	-
	CD (p = 0.05)	1.10	1.51	1.89	2.43	-	-	-	-

Effect of T. viride on the mycelial growth of Fusarium oxysporum f. sp. cubense on borax amended medium

$$I = \frac{C - T}{C} x100$$

Where,
I = Inhibition percentage;
C = Colony diameter in check (mm);
T = Colony diameter in treatment.

The growth inhibition of *T. viride* against *Fusarium oxysporum* f. sp. *cubense* was studied in borax amended medium by dual culture technique (Utkhade and Rake, 1983). The PDA medium was amended with borax at four different concentrations *viz.,* 100, 250, 500 and 750 ppm. The amended medium was poured in to sterilized Petri plate @ 20 ml / plate. The mycelial disc (9 mm) of the test pathogen *Fusarium oxysporum* f. sp. *cubense* was placed 25 mm away from periphery of the Petri plate. The mycelial disc (9 mm) of *T. viride* was placed in the opposite direction at a distance of 25 mm away from periphery of the Petri plate.

The plates were incubated and mean colony diameter of the pathogen as well as antagonist was measured. The per cent inhibition growth was calculated using below mentioned formula.

Results and discussion

Effect of certain micro nutrients on the mycelial growth of Fusarium oxysporum f. sp. cubense

The experiment was conducted to find out the effect of different conc. on the mycelial growth of *Fusarium oxysporum* f. sp. *cubense* causing panama wilt of banana (Table 1).

Among the eight micro nutrients tested, (calcium nitrate, ammonium sulphate, copper sulphate, potassium chloride, sodium chloride, borax, ferrous sulphate and zinc sulphate), borax @ 500 ppm completely inhibited the growth of *Fusarium oxysporum* f. sp. *cubense* followed by zinc sulphate (47.3, 42.7, 31.0 and 14.3 mm in 100, 250, 500 and 750 ppm, respectively). The maximum mycelial growth was recorded in ammonium sulphate. Lakpale et al. (1997) reported that micronutrients at 750 ppm conc. retarded the mycelial growth of *Rhizoctonia solani* causing sheath blight of paddy.

Efficacy of borax on mycelial growth and sporulation of Trichoderma viride

The synergistic action of borax with fungal antagonist *T. viride* has been experimented under in vitro condition (Table 2).

The mean colony diameter of *T. viride* in the borax amended solid medium at different concentrations viz., 100, 250, 500 and 750 ppm was recorded in 70.0, 67.16, 69.66 and 50.16 mm respectively. As the micro nutrient concentration increased the colony diameter decreased. The colony diameter recorded after 96 h of inoculation

Table 2 Efficacy of borax on mycelial growth and sporulation of *T. viride*

Sl. No.	Conc. of borax (ppm)	Mycelial growth (mm)				Per cent inhibition (%)			Sporulation of *T. viride* (1x10 ^{8} spores/sq. cm)
		48 h	72 h	96 h	mean	48 h	72 h	96 h	
1.	100	41.25	80.75	88.00	70.00	2.36	8.76	2.36	170.12 (10.14)
2.	250	40.30	72.50	88.25	67.16	4.62	18.08	1.94	175.25 (10.53)
3.	500	44.75	75.25	89.00	69.66	4.73	14.97	1.94	207.48 (10.32)
4.	750	30.75	51.00	68.75	50.16	27.22	42.37	23.61	94.06 (9.97)
5.	Control	42.25	88.50	90.00	73.58	-	-	-	136.21 (10.13)
	CD (p=0.05)	1.05	2.00	0.71	0.50	-	-	-	

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Table 3 Effect of *T. viride* and borax on the mycelial growth of *Fusarium oxysporum* f. sp. *cubense*

Sl. No.	Conc. of borax (ppm)	Pathogen colony diameter (mm)	Growth inhibition (%)
1.	100	20.0	77.78
2.	250	20.0	77.78
3.	500	16.5	81.67
4.	750	10.2	88.67
5.	Control	35.0	-
	CD (P=0.05)	0.50	-

was 88.0, 89.00 and 88.25 mm at 100, 250 and 500 ppm, respectively. Hence, it can be concluded that the antagonist namely *T. viride* was compatible with borax at low concentrations. In addition to the increase in the mycelial growth of *T. viride* the sporulation of the *T. viride* was also found to be increased in borax amended medium. The maximum sporulation was recorded in conc. of 500 ppm (207.48 x 10^8 spores/ sq.cm).

Effect of T. viride and borax on the mycelial growth of Fusarium oxysporum f. sp. cubense

Dual inoculation of *Fusarium oxysporum* f. sp. *cubense* and *T. viride* in borax amended medium (solid) was conducted and the linear growth of *Fusarium oxysporum* f. sp. *cubense* was calculated and recorded in table 3.

The growth of *Fusarium oxysporum* f. sp. *cubense* in different concentrations of borax viz., 100, 250, 500 and 750 ppm was recorded as 20.0, 20.0, 16.5 and 10.2 mm, respectively. The growth of *Fusarium oxysporum* f. sp. *cubense* in the control was 35.0 mm.

Sundaravadana and Alice, (2006) reported that the micronutrient amended medium generally improved the bio control potential of *T. viride* against *M. phaseolina*. The above findings lend support to the present investigation and the addition of micronutrients particularly borax increased the biocontrol activity of *T. viride.* Hence, this investigation suggests that the borax could be effectively used as viable control strategy for agriculturally important soil borne diseases.

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