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MASS MULTIPLICATION OF BIOCONTROL AGENTS IN BRINJAL SHOOT AND FRUIT BORER, *L. ORBONALIS* UNDER *IN-VITRO* (LAB) AND *IN-VIVO* MICROPLOT CONDITION

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Abstract

The bio-formulation of boric acid based *Beauveria bassiana*-Bb@1×10^8cfu exhibited maximum survival percentage of 91.70 in freshly development. The order of merit the medium containing clay soil based *Trichoderma viride* (Tv)@1×10^8cfu, exhibited the second highest survival while least survival of spores was determined in bentonite fine powder based *Metarhizium anisopliae* Ma@1×10^8cfu.

Key words : *L. orbonalis*, microplot condition, bio-formulation, isolate of antagonist.

Introduction

The vegetable production in our country has touched a new height in recent years, occupying an area of 6.07 million hectares and production of 91.3 million tones. Due to advent of hybrid varieties and increasing awareness about nutritional security, vegetable production is getting continuous momentum in our country. Brinjal is a hardy crop and can be grown practically on all sorts of soils ranging from sandy to heavy clay. However, various factors have been recognized that are responsible for its low yield including poor quality of seed, incidence of disease and pests etc.

In the world, India happens to be the second largest producer of vegetables next to the China. The estimated area under vegetable cultivati0.n during 2007-08 was 78, 03,000 ha with an annual production of 12.5887 million tons. The per capita consumption of vegetables in our country is still very low, which in view of the existing population worked out to be 175g/head/day recommended by National Institute of Nutrition Hyderabad.

Brinjal is infested by many insect pests and some are of major importance affecting the yield and quality such as, jassid (*Amrasea biguttula biguttula* Ishida), whitefly (*Bemisia tabaei* Gennadius), stem borer (*Euzophera perticella* Reg), hadda beetle (*Epilachna* spp.), lace wing bug (*Urentius centis* Distant) and the shoot and fruit borer (*Leucinodes orbonalis* Guenee). On the basis of extent of damage brinjal shoot and fruit borer alone is considered as a serious menace and has been causing heavy losses as high as 31.61 and 73.59 per cent damage to winter and summer brinjal, respectively.

The developed bio-control based bio-pesticide along with some other botanical based products, which would be effective for management of shoot and fruit borer with eco-friendly and least or no adverse effect on plants. Therefore, the bio efficacy of developed bio-formulations was evaluated against shoot and fruit borer of brinjal. *Trichoderma viride, T. harzianum* and *T. virens* are most extensively used fungal antagonists. They are mass-produced using fermentation technology. They are used for the management of soil borne pathogens by seed treatment and soil application.

Materials and Methods

The present investigations were conducted at the Farm and lab of Institute of Agricultural Sciences, Bundelkhand University, Jhansi (U.P.), India. It is situated at 77.30^0 E Longitude and 27.15^0 N Latitude and is about 178.37 m above mean sea level. The climate is subtropical and semi-arid. The metrological data recorded during the experimentation periods (2012-13) based on observation made at the metrological observatory of the IGFRI, Jhansi.

Isolate of antagonist *viz. T. viride*, *B. bassiana*, *M. anisopliae* were selected for present *in-vitro* and *in-vivo* tests in the present investigations. The fungal antagonists were maintained on PDA slants at 4°C after growing for seven days at 25±2°C. Above fungal bio-control agents were inoculated separately on fresh prepared potato dextrose broth medium in conical flask and incubated at 25±2°C for 10 days for mass multiplication. The suspension of profusely developed mycelium and conidia on Potato Dextrose broth were transferred in polypropylene bags containing pre-soaked and sterilized sorghum grains under aseptic conditions and kept it an ambient temperature for 7 days. Well colonized and profusely developed conidia and spores were transferred on sterile bloating paper for air drying under laminar flow. The dried colonized sorghum grains were powdered and sieved with 80-mesh sieve under aseptic conditions separetly. The concentration of conidia and chlamydospores were further determined using a haemocytometer prior to preparations of bio-formulations.

Results and Discussion

The investigation carried out on the brinjal shoot and fruit borer, *L. orbonalis* under *in-vitro* (lab) and *in-vivo* microplot condition with reference to mass production of biocontrol agents, development of biopesticide through formulation, determination of shelf life and their testing against brinjal shoot and fruit borer *L. orbonalis* along with neem oil and insecticides. Present investigation is mainly focused on comparative evaluation of eco-friendly management components against shoot and fruit borer, causing pest *L. orbonalis* on brinjal.

The management of brinjal shoot and fruit borer, *L. orbonalis* through very potent biopesticide of *T. viride* (@1x10^8cfu) formulated in Clay soil+CMC@5%w/w, *B. bassiana*-Bb_1 (@1x10^8cfu) in boric acid powder+CMC, Bentonite powder + CMC + *M. anisopliae* Ma, @1x10^8cfu, Neem oil @1% (Nimulson -P-1500PP Aza), Quinolphos @1ml/L, out of which 3, antagonistic bio-control agents *Trichoderma viride Beauveria bassiana* and *Metarhizium anisopliae* were mass multiplied on liquid and solid medium. Profusely developed biocontrol agents were formulated in different filler/career to maintain the prolonged shelf life of spores and mycelium during storage in laboratory.

The bio-formulation of boric acid based *Beauveria bassiana*-Bb@1x10^8cfu exhibited maximum survival percentage of 91.70 in freshly developed at 0 day which showed gradual reduction with time *i.e.* 89.50, 86.90, 84.30, 81.90, 79.45, 76.55 after the intervals of 30, 60, 90, 120, 150 and 180 days, respectively. From the order of merit of number of conidia taking into account at same time intervals of observations as above the performance of the medium containing clay soil based *Trichoderma viride* (Tv)@1x10^8cfu, exhibited second highest survival (89.50, 82.40, 76.20, 72.80, 70.80, 63.33 and 62.0%) while least survival of spores was determined in bentonite fine powder based *Metarhizium anisopliae* Ma@1x10^8cfu (90.67, 82.33, 79.67, 77.67, 76.67, 61.0 and 55.0%).

In the present investigations (table 1) promising results were obtained with all 3 selected fillar/career based formulations of *B. bassiana*, *T. viride* and *M. anisopliae* to attain maximum survival of spores during storage at ambient temperature. A comparison of 3 different formulations of above said biocontrol agents were made for testing viability of conidia of each, out of which both were found compatible between the ingredients as confirmed by their viable spores. However, the

Table 1 : Survival of *T. viride*, *B. bassiana* and *M. anisopliae* in different formulations during storage at room temperature (20-35°C).

Treatments	Percent viability days after						
	0	30	60	90	120	150	180
T-1 Clay soil fine powder + CMC + (*T. viride*-Tv2) @1x10^8cfu)	89.50 (71.09)	82.40 (65.20)	76.20 (60.80)	72.80 (58.56)	70.80 (57.29)	69.33 (56.37)	62.00 (51.94)
T-2 Boric acid powder + CMC (*B. bassiana* -Bb_1 @1x10^8cfu)	91.70 (73.26)	89.50 (71.09)	86.90 (68.78)	84.30 (66.66)	81.90 (64.82)	79.45 (63.04)	76.55 (61.04)
T-3 Bentonite powder+CMC+*M. anisopliae* Ma	90.67 (72.22)	82.33 (65.15)	79.67 (63.21)	77.67 (62.37)	76.67 (61.12)	61.00 (51.35)	55.00 (47.87)
S. Em±	0.45	0.31	0.38	0.34	0.53	0.37	0.29
LSD (P= 0.05)	1.33	1.13	1.13	0.99	1.55	1.09	0.86

incompatible have not found after 180 days in the treatment of bentonite fine powder which showed significantly least viable count in comparison to other two combinations. Sarode *et al*. (1998) reported evaluated talc along with other substrates as supporting carrier for *T. harzianum* and observed Trichoderma can be stored safely at room temperature in talc powder up to the period of 8 months. For field application of a potential fungal bioagent, an inert immobilizing substrate is essentially required which could carry maximum number of propagules of the biocontrol agent. Prasad and Rangeshwaran (2000) also tested three carrier materials *viz.,* talc, kaolin and bentonite for their effect on shelf life of *Trichoderma harzianum* and these formulations assessed against *Rhizoctonia solani*. They observed Talc and kaolin retained more than 10^6 viable propagules up to 90 days and by 120 days propagules declined below the optimum level while Bentonite was least suitable as a carrier since a drastic decline in the Trichoderma population at 120 days. Seed treatment with kaolin and talc formulation of the bioagent recorded more than 50% plant stand even at 120 days storage. Kaolin treatment recorded more plant stand than talc and bentonite at different storage periods. Seed treatment with bentonite formulation stored for 120 days recorded only 34.8% plant stand. It was concluded that kaolin and talc are better carriers of *T. harzianum*.

Various formulations of bioagents have been developed and tried against several diseases of crop plants (Lumsden *et al*., 1996; Lewis and Papavizas, 1987; Lewis, *et al*., 1995; Lewis *et al*., 1998; Harman, 2000). Their efficacies varied according to the crop plants against bioagents applied and their mode of application for example Root Shield a formulation developed from *Trichoderma harzianum* T-22 more effective in furrow drench in comparison to seed treatment (Harman, 2000).

References

Harman, G. E. (2000). Myths and dogmas of biocontrol: changes in perceptions derived from research on *Trichoderma harzianum* T22. *Plant Dis.,* **84** : 377-393.

Lewis, J. A., R. P. Larkin and D. L. Rogers (1998). A formulation of *Trichoderma* and *Gliocladium* reduce damping off caused by *Rhizoctonia solani* and saprophytic growth of the pathogen in the soil less mix. *Plant Dis.,* **82** : 501-506.

Lewis, J. A., R. D. Lumsden, D. R. Fravel and B. S. Shasha (1995). Application of pregelatinized starch flour granules formulation of biocontrol fungi to control damping off disease caused by *Rhizoctonia solani*. *Biol. Control*, **5** : 397-404.

Lewis, J. A. and G. C. Papavizas (1987). Application of *Trichoderma* and *Gliocladium* in alginate pellets for control of *Rhizoctonia* damping off. *Plant Pathol*., **36** : 397-404.

Lumsden, R. D., J. F. Walter and C. P. Baker (1996). Development of *Gliocladium virens* for damping off disease control. *Can. J. Plant. Pathol*., **18** : 463-468.

Prasad, R. D. and R. Rangeshwaran (2000). Effect of soil application of granular formulation of *Trichoderma* on *Rhizoctonia solani* incited seed rot and damping off of chickpea. *J. Mycol. Pl. Pathol.,* **30** : 216-220.

Sarode, S. V., V. R. Gupta and M. N. Asalmol (1998). Suitability of carriers and shelf life of *Trichoderma harzianum*. *Indian Journal of Plant Protection*, **26** : 188-189.