

Evaluation of entomopathogenic fungi against tomato thrips, *Thrips tabaci* Lindeman

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ABSTRACT

Eleven fungal isolates belonging to *Beauveria bassiana* (Balsamo-Crivelli) Vuillemin, *Metarhizium anisopliae* (Metschnikoff) Sorokin, *Lecanicillium lecanii* (Zimmerman) Zare and Gams and *Metarhizium flavoviride* Gams and Rozsypal var *minus* with different host origins were assayed for their pathogenicity against the tomato thrips, *Thrips tabaci* Lindeman. The tomato thrips were found susceptible to all the examined isolates of entomopathogenic fungi. The fungal isolate Bb111 of *B. bassiana* was found highly virulent to *T. tabaci* with an LC₅₀ of 1.6×10^5 spores/mL as evidenced by its non overlapping fiducial limits to other isolates. Mycosis on *T. tabaci* by Bb111 isolate had fast lethal effect after treatment with conidial suspensions at the concentration of 10^8 conidia/mL. The values of the median lethal time required for 50% mortality (LT₅₀) of the respective isolate was 104.91 h. The rest of the tested isolates showed higher LC₅₀ and LT₅₀ values, indicating the intermediate or low virulence to *T. tabaci*. The results of the present investigation revealed that the Bb111 could be further exploited on a field scale against *T. tabaci*.

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Key words: *Thrips tabaci*, *Beauveria bassiana*, *Metarhizium anisopliae*, *Lecanicillium lecanii*.

INTRODUCTION

Tomato, *Lycopersicon esculentum* (Miller) is a staple fruit vegetable. Fresh fruits are very important source of vitamins and minerals which are essential for human health. That is one of the most important fruit consumed as vegetables in the world. It is considered as an important cash and industrial crop in many parts of the world (Babalola *et al.* 2010). The tomato thrips, *Thrips tabaci* Lindeman (Thysanoptera: Thripidae) is an important pest of field and greenhouse crops around the world. It causes damage directly through feeding and indirectly through the transmission of lethal plant viruses. It is difficult to control this pest with insecticides because of its small size and cryptic habits (Lewis, 1997). Entomopathogenic fungi are currently being investigated for the control of many important insect pests on various crops around the world, and some are commercially available. There are many studies on the efficacy of several entomopathogenic fungi on

thrips. Vestergaard *et al.* (1995) and Brownbridge (1995) showed that *Beauveria bassiana*, *Metarhizium anisopliae* and *Verticillium lecanii* were more active against the western flower thrips, *Frankliniella occidentalis* (Pergande) than *Paecilomyces fumosoroseus*. Hall *et al.* (1994) and Saito (1991) suggested that *Hirsutella* sp., *P. fumosoroseus* and *B. bassiana* may be useful in the management of the melon thrips, *Thrips palmi* Karny. Ekesi *et al.* (1998) stated that *B. bassiana* and *M. anisopliae* are highly pathogenic to the legume flower thrips, *Megalurothrips sjostedti* (Tryborn). In the glasshouse, *V. lecanii* has been used successfully to control *T. tabaci* on cucumber (Gillespie 1986). The studies of Maniania *et al.* (2003) indicated that *M. anisopliae* had a potential to control *T. tabaci* in the field. With this view the present study was conducted to investigate the pathogenicity of different entomopathogenic fungi against *T. tabaci* larvae under laboratory condition.

MATERIALS AND METHODS

Test insect collection and rearing

The *Thrips tabaci* Lindeman were collected from the field, mass reared and maintained in the tomato plants (Variety, PKM) at Insectary, unit of the Department of Agricultural Entomology, Tamil Nadu Agricultural University, Coimbatore.

Source of fungal isolates

Pure cultures of the eleven entomopathogenic fungal strains obtained from field survey, Department of Agricultural Entomology and Plant Pathology, Tamil Nadu Agricultural University (TNAU), National Bureau of Agriculturally Important Insects (NBAIL), Bangalore and Sugarcane Breeding Institute (SBI), Coimbatore (Table 1) were used for this study.

Preparation of spore concentrations

All the fungal isolates were cultured in 100mL SMA+Y liquid medium in 250mL conical flask and incubated at room temperature for 10 days. After sporulation of the fungal isolates, it was ground in ordinary mixer and made into liquid spore suspension. This was filtered through double layered muslin cloth to remove the mycelial mat. The suspension was shaken thoroughly with a drop of 0.05%

aqueous Tween 80 solution in order to disperse the spores in the solution. The conidial suspension was vortexed for 5 min to produce a homogenous conidial suspension. The spore count in the suspension was assessed by using a haemocytometer and was estimated using the formula suggested by Lomer and Lomer, (1996). Based on the number of spores, all the cultures were adjusted to 1×10^8 spores mL⁻¹ from which the lower concentrations were prepared by serial dilution technique for bioassay studies.

Bioassays

The pathogenicity of eleven fungal isolates against tomato thrips was determined by using the detached leaf method (Yokomi, 1988). The tomato leaves were rinsed in tap water for 15 min, washed three times with distilled water, and air dried in a sterile laminar flow hood. Working in the sterile hood, leaves of the respective crops were placed on 1.5% agar in 90×20 mm² plastic Petri dishes. The 1.5% agar contained no nutrients but supplied water to the leaves and helped to maintain 100% RH during the test. At least 30 adult thrips were placed on each leaf, settled for 1 day before conidial treatment.

Table 1. Origin of fungal isolates assayed against *S. dorsalis*

Fungal pathogens	Isolate	Origin	Source of isolates
<i>Beauveria bassiana</i>	Bb111	Coimbatore	Dipteran fly
	Bb101	Department of Agricultural Entomology, TNAU*, Coimbatore	<i>Tetranychus urticae</i>
	BbSBI	SBI, Coimbatore**	Shoot borer
	BbNBAIL	NBAIL, Bangalore***	Not Known
	B2	Department of Plant Pathology, TNAU*, Coimbatore	Soil
<i>Metarhizium anisopliae</i>	MaSBI	SBI, Coimbatore**	Shoot borer
	MaNBAIL	NBAIL, Bangalore***	Not Known
	M2	Department of Plant Pathology, TNAU*, Coimbatore	Not Known
<i>Metarhizium flavoviride</i> var <i>minus</i>	Mf	Coimbatore	Brown Plant Hopper (BPH)
<i>Lecanicillium lecanii</i>	L1NBAIL	NBAIL, Bangalore***	Not Known
	L2	Department of Plant Pathology, TNAU*, Coimbatore	Not Known

*Obtained from Tamil Nadu Agricultural University (TNAU); ** Obtained from Sugarcane Breeding Institute (SBI); *** Obtained from National Bureau of Agriculturally Important Insects (NBAIL) and 111 number were assigned to indicate the isolate number of the pathogen

Five different spore concentrations (1×10^8 to 1×10^4 spores mL^{-1}) were prepared and each concentration was replicated three times. Ten mL of respective concentrations were sprayed on thrips using atomizer. Thrips sprayed with 0.05 per cent Tween 80 solution served as control.

Mortality of thrips

Percentage of thrips infected was recorded up to seven days of treatment. Cadavers with fungal growth only were considered as a successful infection. The control value was determined by using Abbott's formula.

Statistical analysis

The corrected mortality data were analyzed by Probit analysis (Finney, 1971) and the median lethal concentration (LC_{50}) and the median lethal time (LT_{50}) values were computed by using the statistical computer programme, SPSS ver.16.00 (SPSS Inc., USA).

RESULTS AND DISCUSSIONS

Fungal Infection in Adults

Thrips infected by fungi were mummified and hard to touch. Mycelial growth developed

after 24 to 48 hrs of death. Initially, growth of the fungus was inconspicuous through the intersegmental membrane of abdomen, legs and finally the entire cadaver was fully covered with fungal mycelium.

Median Lethal Concentration (LC_{50})

Bioassay results showed that *T. tabaci* were susceptible to all the fungal isolates examined. The test for the goodness of fit indicated no significant heterogeneity in the linear relationships for all fungal isolates tested ($P > 0.05$). Based on the estimates of the LC_{50} and associated 95% confidence limits (Table 2), Bb111 and Bb101 isolates of *Beauveria bassiana* had higher virulence to *T. tabaci* with the lowest LC_{50} values of 1.6×10^5 and 1.7×10^5 spores/mL, respectively. The isolate, MaSBI was less virulent with an LC_{50} value of 5.2×10^5 spores/mL. In remaining isolates, viz. B2, BbSBI, BbNBAIL, MaNBAIL, M2, Mf, L1NBAIL and L2, the LC_{50} values ranged from 2.3×10^5 to 3.8×10^5 spores/mL and were not significantly different in their virulence as evidenced by the overlapping fiducial limits.

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Table 2. Dose mortality response of fungal isolates against thrips in tomato.

Fungal isolates	Heterogeneity (χ^2)*	Regression equation	LC_{50} ($\times 10^5$ spores mL^{-1})	95% Fiducial Limits (spores mL^{-1})
<i>Beauveria bassiana</i> (Bb 101)	2.867	$Y=0.433x+2.742$	1.7	$4.5 \times 10^4 - 6.4 \times 10^5$
<i>Beauveria bassiana</i> (Bb 111)	1.329	$Y=0.464x+2.593$	1.6	$4.6 \times 10^4 - 5.2 \times 10^5$
<i>Beauveria bassiana</i> (BbSBI)	1.079	$Y=0.330x+3.169$	3.6	$7.6 \times 10^4 - 1.7 \times 10^6$
<i>Beauveria bassiana</i> (BbNBAIL)	3.155	$Y=0.466x+2.492$	2.5	$8.0 \times 10^4 - 8.0 \times 10^5$
<i>Beauveria bassiana</i> (B2)	2.509	$Y=0.278x+3.506$	2.3	$3.8 \times 10^4 - 1.5 \times 10^6$
<i>Metarhizium anisopliae</i> (MaSBI)	2.885	$Y=0.333x+3.100$	5.2	$1.2 \times 10^5 - 2.3 \times 10^6$
<i>Metarhizium anisopliae</i> (MaNBAIL)	1.133	$Y=0.389x+2.837$	3.6	$9.5 \times 10^4 - 1.4 \times 10^6$
<i>Metarhizium anisopliae</i> (M2)	1.121	$Y=0.300x+3.368$	2.7	$5.0 \times 10^4 - 1.5 \times 10^6$
<i>Metarhizium flavoviridae</i> var <i>minus</i> (BPH)	0.115	$Y=0.340x+3.157$	2.6	$5.9 \times 10^4 - 1.2 \times 10^6$
<i>Verticillium lecanii</i> (VINBAIL)	3.905	$Y=0.451x+2.506$	3.8	$1.1 \times 10^5 - 1.3 \times 10^6$
<i>Verticillium lecanii</i> (L2)	1.319	$Y=0.39x+2.855$	3.3	$8.6 \times 10^4 - 1.2 \times 10^6$

* All lines are significantly good fit @ $P < 0.05$

Median Lethal Time (LT_{50})

At the concentration of 10^8 spores/mL, the lethal time values required to 50% mortality (LT_{50}) of the isolates Bb111 and Bb101 were 104.91h and 107.98 h respectively. Recorded LT_{50} values suggest that virulence of Bb111 and Bb101 were not significantly different at

$P < 0.05$ (Table 3). The LT_{50} values in other isolates like, B2, BbSBI, BbNBAIL, MaSBI, MaNBAIL, M2, Mf, L1NBAIL and L2 were higher and ranged between 109.02 h and 244.25 h and were not significantly different in their virulence as indicated by the overlapping fiducial limits.

Table 3. Time mortality response of fungal isolates against thrips in tomato

Fungal isolates	Heterogeneity (2)*	Regression equation	LT ₅₀ ** (h)	95% Fiducial Limits (h)
<i>Beauveria bassiana</i> (Bb 101)	0.348	Y= 2.803x-0.697	107.985	92.238-126.422
<i>Beauveria bassiana</i> (Bb 111)	3.763	Y=2.746x-0.550	104.912	87.824-125.325
<i>Beauveria bassiana</i> (BbSBI)	2.238	Y=2.67x-0.647	128.732	108.386-152.901
<i>Beauveria bassiana</i> (BbNBAlI)	2.438	Y=2.703x-0.558	113.996	96.038-135.312
<i>Beauveria bassiana</i> (B2)	2.120	Y=2.680x-0.460	109.017	90.017-131.007
<i>Metarhizium anisopliae</i> (MaSBI)	2.019	Y=2.110x-0.170	244.249	158.823-375.620
<i>Metarhizium anisopliae</i> (MaNBAlI)	2.309	Y=3.084x-1.505	130.647	108.942-156.676
<i>Metarhizium anisopliae</i> (M2)	1.260	Y=2.339x-0.109	154.319	119.042-200.051
<i>Metarhizium flavoviridae</i> var minus (BPH)	5.531	Y=2.660x-0.534	122.496	99.025-151.531
<i>Verticillium lecanii</i> (VINBAlI)	1.997	Y=2.356x-0.179	154.869	121.323-197.689
<i>Verticillium lecanii</i> (L2)	3.213	Y=2.531x-0.663	185.954	131.433-263.091

* All lines are significantly good fit @ P 0.05; ** LT50 values recorded at the highest concentration of 10⁸ spores mL⁻¹

The effectiveness of a fungal isolate is measured in terms of its pathogenicity (LC₅₀) and the speed (LT₅₀) with which it kills the target pest (Negasi *et al.*, 1998). In the present study, eleven fungal isolates assayed against *Thrips tabaci* Lindeman adults caused infection under laboratory conditions with considerable variation between the isolates. Comparing the LC₅₀ and LT₅₀ values, two fungal isolates Bb111 and Bb101 were superior and highly virulent. B2 and BbNBAlI were next in the order of efficiency in terms of virulence. For each bioassay, the corrected mortalities were transferred to probit units (y) then regressed against the log₁₀-transformed conidial concentrations (x), yielding a well-fitted linear relationship. All the fungal pathogens tested showed that the mortality of adults increased with increase in concentration. Gillespie (1986) and Fransen (1990) reported that in laboratory studies, *T. tabaci* was susceptible to *M. anisopliae*, *B. bassiana*, *P. fumosoroseus* and *V. lecanii*. In the present study, the genera and species varied in their pathogenicity. Similarly, Ekesi *et al.* (1998) reported that the differences in virulence were more pronounced for *B. bassiana* strains than for *M. anisopliae*. Malee Thungrabeab *et al.* (2006) have reported that, *B. bassiana* recorded the highest mortality with 95.5% among the fungal isolates.

Singh *et al.* (2011) have reported that, among the entomopathogenic fungi, *B. bassiana* performed better in respect of reducing thrips population as well as increasing yield. In the present study, the isolate Bb111 was found to be more virulent against *T. tabaci* and hence there is a possibility to recommend the isolate as promising candidate for use in tomato thrips management.

Currently, farmers rely heavily on systemic insecticides for the management of thrips. Continuous usage of chemicals lead not only to the resistance problem but also to several other problems including health hazards. The microbial control aimed in the proposed study, using fungal pathogens, can result in the successful management of thrips in an economic and ecofriendly manner. Also, the biodiversity of beneficial fauna will be conserved.

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