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COMPATIBILITY OF *BEAUVERIA BASSIANA* (BALSAMO) VUILLEMIN WITH DIFFERENT INSECTICIDES AND FUNGICIDES

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ABSTRACT

Compatibility of *B. bassiana* was studied in laboratory conditions with commonly used twelve insecticides and five fungicides with three doses (Lower, recommended and higher) by poison food technique. The results showed that the insecticide fipronil 5 SC 0.005% was found to be the most compatible at all doses, as it was categorized as harmless (Grade 1) to this fungus. However, carbosulfan 25 EC 0.025%, emamectin benzoate 5 SG 0.00125%, dimethoate 30 EC 0.015%, imidacloprid 17.8 SL 0.0026% and chlorantraniliprole 18.5 SC 0.003% were relatively harmless (Grade 1) at lower dose. At recommended dose, fipronil 5 SC 0.010%, chlorantraniliprole 18.5 SC 0.006%, acetamiprid 20 SP 0.006% and dimethoate 30 EC 0.030% were compatible with tested fungus. Dichlorvos 76 EC and quinalphos 25 EC at all doses were categorized as harmful (Grade 4) to these fungi. In case of fungicides, chlorothalonil 75 WP was found to be the most compatible at all doses. Hexaconazole 5 EC, mancozeb 70 WP and carbendazim 50 WP were found slightly harmful (Grade 2) while propiconazole 25 EC at all doses was harmful (Grade 4) to this fungus. From the overall results it can be concluded that, among twelve insecticides, fipronil, chlorantranilliprole, acetamiprid, dimethoate, carbosulfan, novaluron and imidacloprid can be used with *B. bassiana*. Among five fungicides, chlorothalonil can be used with *B. bassiana*.

Key words: Entomopathogenic fungi, *Beauveria bassiana*, groundnut, compatibility, *Helicoverpa armigera*, *Spodoptera litura*, insecticides, fungicides, Junagadh, compatible insecticide

Preference of insecticides depends on their easy availability and applicability, but their excessive and indiscriminate use has resulted in the development of insecticidal resistance in the pests and environmental pollution (Phokela et al*.,* 1990). There is a need to explore alternatives, encompassing available pest control methods and techniques in order to reduce the sole dependence on insecticides.

The entomopathogenic fungus, *Beauveria bassiana* (Balsamo) Vuillemin (Hyphomycetes, Moniliates) is a potential biocontrol agent against approximately 700 insect species used for the management of several crop insect pests. It is also very effective and widely used bio-pesticide, which controls various pests of different crops. It can be developed in the laboratory for use as myco-insecticidal agent. The larvae (Cadavers) infected by *B. bassiana* were identified by the chalky white powdery growth which was easily dislodged by tapping from hardened cuticle and cadavers become brittle and mummified. The fungus produces a toxic substance 'Beauvericine', which causes death of larvae. Parameswaran and Sankaran (1977) have first time recorded this fungus occurring naturally in India. Likewise, Dutky (1959) stated that with its wide undefined host range, *B. bassiana* is referred as "Magnificent pathogen".

Compatibility experiments of *B. bassiana* were carried out to detect insecticides and fungicide's side effects on entomopathogenic fungi (Neves et al., 2001). In vitro studies indicated inhibition of *B. bassiana* by many pesticides (Olmert and Kenneth, 1974). Todorova et al. (1998) reinforced the importance of pesticides influence on conidial germination, vegetative growth and sporulation of fungus and some are compatible with fungus. For some products, the combination treatments showed higher dose mortality response than the sole treatments of fungal conidia (Purwar and Sachan, 2006).

MATERIALS AND METHODS

The insecticides and fungicides selected for compatibility study were among those, commonly used for pest management. The effect of these insecticides and fungicides on the growth inhibition of *B. bassiana* were evaluated. The insecticides and fungicides dose were calculated on the basis of lower, recommended and higher for field application rate. Twenty insecticides and five fungicides were evaluated by poisoned food technique described by Dhingra and Sinclair (1985) in Potato Dextrose Agar (PDA) medium. Twenty mililitre of PDA medium was sterilized in individual boiling tubes and the insecticide & fungicide emulsions of required concentration (lower, recommended and higher) were incorporated into the melted sterile PDA aseptically, thoroughly mixed, poured into 9 cm diameter sterile petri dishes and allowed to solidify under laminar flow cabinet. Make a liquid suspension containing 2 x 10^6 spore per ml of *B. bassiana* in 500 ml of conical flask. Take a 20 µl of this suspension with the help of micro pipette and inoculate the center of petri plates having poisoned medium with the insecticides & fungicides and repeated thrice. Growth medium (PDA) without insecticide and fungicide but inoculated with liquid suspension of *B. bassiana* served as an untreated check.

Radial growth of the entomogenous fungus was recorded when full growth obtained with control (15th day of inoculation). The % inhibition in radial growth over control was calculated as per the following equation given by Bliss (1934), and statistical analysis was done by suitable transformation.

$$I = \frac{C - T}{C} * 100$$

Where,

I = % inhibition

C = Colony diameter of control plate

T = Colony diameter of treated plate

The growth inhibition (%) of the fungus obtained from each treatment further classified into different categories based on Hassan's classification scheme (Hassan, 1989). Grade 1 (Harmless, <50% reduction in beneficial capacity), grade 2 (Slightly harmful, 50-79%), grade 3 (Moderately harmful, 80-90%) and grade 4 (Harmful, >90%). The data obtained on the basis of above indexes in each treatment were subjected to arcsine transformation and statistically analyzed.

RESULTS AND DISCUSSION

Compatibility of *B. bassiana* with different insecticides

The compatibility between entomopathogenic fungus and chemical insecticides was tested individually in the laboratory. The optimum combined use of fungi and chemicals for pest management may be required simultaneous applications. Thus, compatibility of *B. bassiana* with commonly used insecticides against *H. armigera* and *S. litura* tested under laboratory conditions and the results are described as below.

Compatibility at lower dose

The data on growth inhibition presented in Table 1 indicated that carbosulfan 25 EC 0.025% showed significantly the lowest (5.59%) growth inhibition of *B. bassiana* at lower dose. It was followed by emamectin benzoate 5 SG 0.0013% (16.71%), dimethoate 30 EC 0.015% (17.95%), imidacloprid 17.8 SL 0.0026% (18.24%), chlorantraniliprole 18.5 SC 0.003% (19.78) and fipronil 5 SC 0.005% (19.96%). The treatments of novaluron 10 EC 0.005% (20.67%), spinosad 45 SC 0.007% (24.00%) and acetamiprid 20 SP 0.003% (30.00%) exhibited moderate growth inhibition of *B. bassiana.* The highest (51.56%) growth inhibition was recorded in dichlorvos 76 EC 0.038% which was followed by quinalphos 25 EC 0.025% (50.59%) and diafenthiron 50 WP 0.025% (38.67%). The above results indicated that among the insecticides tested, carbosulfan, emamectin benzoate, dimethoate, imidacloprid, chlorantraniliprole, fipronil, novaluron, spinosad, acetamiprid and difenthiuron were relatively harmless (Grade 1) to *B. bassiana* as they showed less than 50% growth inhibition at lower dose. Two insecticides (dichlorvos and quinalphos) were slightly harmful (Grade 2) to *B. bassiana.*

Compatibility at recommended dose

The effect of insecticides on the growth of *B. bassiana* at recommended dose is shown in Table 1. The results clearly indicated that the lowest (15.14%) growth inhibition was observed due to fipronil 5 SC 0.010% followed by chlorantraniliprole 18.5 SC 0.006% (17.35%). The insecticides acetamiprid 20 SP 0.006% (30.42%), dimethoate 30 EC 0.030% (30.93%) and spinosad 45 SC 0.014% (33.78%) were found next in order to inhibit the growth of *B. bassiana*. Carbosulfan 25 EC 0.050%, novaluron 10 EC 0.010%, imidacloprid 17.8 SL 0.005%, difenthiuron 50 WP 0.050% and emamectin benzoate 5 SG 0.0025% have moderately affected the growth of the fungus at 39.32, 39.65, 41.31, 43.56% and 50.07%, respectively. The results showed that dichlorvos 76 EC 0.076% caused the highest reduction in mycelial growth of *B. bassiana* with 87.11% inhibition, which was followed by quinolphos 25 EC 0.050% (75.93%).

Table 1. Effect of different insecticides on the growth of *B. bassiana* at lower, recommended and higher dose

Sl. No.	Treatments	Lower dose			Recommended dose			Higher dose		
		Conc. (%)	Dose/lit	Growth inhibition (%)	Conc. (%)	Dose/lit	Growth inhibition (%)	Conc. (%)	Dose/lit	Growth inhibition (%)
1.	Dimethoate 30 EC	0.015	0.50 ml	25.04 (17.95)	0.030	1.00 ml	33.77 (30.93)	0.045	1.50 ml	44.92 (49.86)
2.	Carbosulfan 25 EC	0.025	1.00 ml	13.66 (5.59)	0.050	2.00 ml	38.83 (39.32)	0.075	3.00 ml	67.29 (85.10)
3.	Diafenthiuron 50 WP	0.025	0.50 g	38.45 (38.67)	0.050	1.00 g	41.27 (43.56)	0.075	1.50 g	46.72 (53.00)
4.	Fipronil 5 SC	0.005	1.00 ml	26.53 (19.96)	0.010	2.00 ml	22.89 (15.14)	0.015	3.00 ml	33.13 (29.87)
5.	Acetamiprid 20 SP	0.003	0.15 g	33.18 (30.00)	0.006	0.30 g	33.44 (30.42)	0.009	0.45 g	60.03 (75.04)
6.	Imidacloprid 17.8 SL	0.0026	0.15 ml	25.28 (18.24)	0.005	0.30 ml	39.98 (41.31)	0.008	0.45 ml	57.31 (70.82)
7.	Quinalphos 25 EC	0.025	1.00 ml	45.34 (50.59)	0.050	2.00 ml	60.64 (75.93)	0.075	3.00 ml	64.08 (80.89)
8.	Dichlorvos 76 EC	0.038	0.5 ml	45.89 (51.56)	0.076	1.0 ml	69.27 (87.11)	0.114	1.5 ml	76.37 (94.44)
9.	Novaluron 10 EC	0.005	0.50 ml	26.79 (20.67)	0.010	1.00 ml	39.02 (39.65)	0.015	1.50 ml	37.73 (37.44)
10.	Spinosad 45 SC	0.007	0.15 ml	29.33 (24.00)	0.014	0.30 ml	35.51 (33.78)	0.021	0.45 ml	51.81 (61.78)
11.	Chlorantraniliprole 18.5 SC	0.003	0.15 ml	26.09 (19.78)	0.006	0.30 ml	24.59 (17.35)	0.009	0.45 ml	37.93 (37.78)
12.	Emamectin benzoate 5 SG	0.0013	0.25 g	22.95 (16.71)	0.0025	0.50 g	45.04 (50.07)	0.00375	0.75 g	49.00 (56.95)
13.	Control	-	-	12.92 (0.00)	-	-	12.92 (0.00)	-	-	12.92 (0.00)
S.Em.±				1.63			1.05			1.00
C.D. at 5 %				4.63			2.98			2.84
C.V. %				12.76			6.14			4.55

Figures in parenthesis are original values, while outside values are angular transformed. Local strain of *B. bassiana* @ 2×10^6 cfu/g was used.

The results on compatibility of *B. bassiana* with different insecticides at their recommended dose revealed that the insecticides viz., fipronil, chlorantraniliprole, acetamiprid, dimethoate, spinosad, carbosulfan, novaluron, imidacloprid, difenthiuron were relatively harmless (Grade 1) to *B. bassiana* as they showed less than 50% growth inhibition at recommended dose. The growth inhibition was rated slightly harmful (Grade 2) with the insecticide, emamectin benzoate, while quinalphos and dichlorvos was rated moderately harmful (Grade 3) to the *B. bassiana*.

Compatibility at higher dose

All the insecticides tested have different compatible levels with *B. bassiana* at higher dose (Table 1). Perusal of data on growth inhibition sign posted that fipronil 5 SC 0.015% recorded the lowest inhibition (29.87%). The treatments, novaluran 10 EC 0.015% and chlorantraniliprole 18.5 SC 0.009% remained next in order, which registered 37.44 and 37.78% growth inhibition, respectively. The growth inhibition was moderately increased in dimethoate 30 EC 0.045% (49.86%), difenthiuron 50 WP 0.075% (53.00%), emamectin benzoate 5 SG 0.00375% (56.95%) and spinosad 45 SC 0.021% (61.78%). The least growth was obtained in dichlorvos 76 EC 0.114% as it showed 94.44% growth inhibition of *B. bassiana*. The remaining insecticides, viz., imidacloprid 17.8 SL 0.008%, acetamiprid 20 SP 0.009%, quinalphos 25 EC 0.075% and carbosulfan 25 EC 0.075% confirmed 70.82, 75.04, 80.89 and 85.10% growth inhibition, respectively.

The higher dose of tested insecticides showed more effect on the growth inhibition of tested fungus as compared to lower and recommended doses. The findings of present investigation showed that the insecticides, viz., fipronil, novaluran, chlorantraniliprole and dimethoate remained harmless (Grade 1) to *B. bassiana*. The rating of the insecticides, difenthiuron, emamectin benzoate, spinosad, imidacloprid, and acetamiprid remained slightly harmful (Grade 2) as their growth inhibition was found between 50 to 79%. Quinalphos and carbosulfan have registered as moderately harmful (Grade 3) whereas dichlorvos have highly inhibited the growth of *B. bassiana* and are distinguished as harmful (Grade 4).

The insecticide, fipronil was found compatible in the studies, which is in complete conformity with the results found by Batista et al. (1996) who tested the compatibility of fipronil with *B. bassiana* and reported that the insecticide did not affect spore production and viability of conidia but caused a slight decrease in the diameter of the colony. Haseeb (2009) reported that the insecticides strongly inhibited the growth of *B. bassiana* was quinalphos (60.9-64.6% reduction) while diamethoate affected least (32.3%), which supports the result of present studies.

Compatibility of *B. bassiana* with different fungicides

Compatibility at lower dose

The data on growth inhibition presented in Table 2 and indicated that chlorothalonil 75 WP 0.100% showed the significantly lowest (63.33%) growth inhibition of *B. bassiana* at lower dose. Hexaconazole 5 EC 0.003%, mancozeb 70 WP 0.093% and carbendazim 50 WP 0.025% showed 80.00%, 82.64% and 82.64% growth inhibition, respectively. The highest (94.44%) mycelial growth inhibition of *B. bassiana* was recorded in propiconazole 25 EC 0.013%.

The above results indicated that among the fungicides tested, chlorothalonil was slightly harmless (Grade 2) to *B. bassiana*. Whereas hexaconazole, mancozeb and carbendazim were moderately harmful (Grade 3) to the mycelia growth of *B. bassiana*. Propiconazole proved to be the strongest growth inhibitor of *B. bassiana* (Grade 4).

Compatibility at recommended dose

The effects of fungicides on the growth of *B. bassiana* at recommended dose are shown in Table 2. The results clearly indicated that the lowest (60.00%) growth inhibition was observed in chlorothalonil 75 WP 0.200%. Mancozeb 70 WP 0.187% and hexaconazole 5 EC 0.006% showed 85.28% growth inhibition of *B. bassiana* whereas, carbendazim 50 WP 0.050% showed 87.92% growth inhibition. Propiconazole 25 EC 0.026% caused the complete reduction (100.00%) in mycelial growth of *B. bassiana*.

The results on compatibility of *B. bassiana* with different fungicides at recommended dose revealed

Table 2. Effect of different fungicides on the growth of *B. bassiana* at lower, recommended and higher dose

Sl. No.	Treatments	Lower dose			Recommended dose			Higher dose		
		Conc. (%)	Dose/lit	Growth inhibition (%)	Conc. (%)	Dose/lit	Growth inhibition (%)	Conc. (%)	Dose/lit	Growth inhibition (%)
1.	Chlorothalonil 75 WP	0.100	1.34	52.76 (63.33)	0.200	2.67	50.81 (60.00)	0.300	4.01	52.76 (63.33)
2.	Mancozeb 70 WP	0.093	1.34	65.46 (82.64)	0.187	2.67	67.46 (85.28)	0.280	4.01	72.10 (90.55)
3.	Hexaconazole 5 EC	0.003	0.50	63.51 (80.00)	0.006	1.00	67.45 (85.28)	0.009	1.50	74.91 (93.19)
4.	Carbendazim 50 WP	0.025	0.50	65.46 (82.64)	0.050	1.00	69.82 (87.92)	0.075	1.50	72.22 (90.55)
5.	Propiconazole 25 EC	0.013	0.50	76.36 (94.44)	0.026	1.00	90.00 (100.00)	0.039	1.50	90.00 (100.00)
6.	Control	-	-	12.92 (0.00)	-	-	12.92 (0.00)	-	-	12.92 (0.00)
S.Em.±				0.90			0.95			0.71
C.D. at 5 %				2.62			2.79			2.07
C.V. %				3.58			3.57			2.63

Figures in parenthesis are original values, while outside values are angular transformed that the fungicide, chlorothalonil was slightly harmless (Grade 2) to *B. bassiana*. The growth inhibition was rated moderately harmful (Grade 3) with the fungicides mancozeb, hexaconazole and carbendazim. Propiconazole proved to be the strongest growth inhibitor of *B. bassiana* (Grade 4).

Compatibility at higher dose

All the fungicides tested have different compatible levels with *B. bassiana* at higher dose (Table 2). Perusal of data on growth inhibition sign posted that chlorothalonil 75 WP 0.300% recorded the lowest (63.33%) growth inhibition. The treatments, mancozeb 70 WP 0.280% and carbendazim 50 WP 0.075% remained next in order, which registered 90.55% growth inhibition. Hexaconazole 5 EC 0.009% exhibited (93.19%) growth inhibition where, propiconazole 25 EC 0.039% inhibited the complete growth of *B. bassiana.*

The above results indicated that among the fungicides tested at higher dose, chlorothalonil was moderately harmless (Grade 3) to *B. bassiana,* while remaining fungicides viz*.,* mancozeb, carbendazim, hexaconazole and propiconazole were harmful (Grade 4) to *B. bassiana.*

The results of present finding are in close agreement with Khan et al. (2012), who recorded that least inhibition (%) of vegetative growth and spore germination was recorded in chlorothalonil 0.1%. Other fungicides viz*.,* mancozeb 0.2%, hexaconazole 0.1% and propiconazole 0.1%, were not compatible with *B. bassiana* and caused complete or strong inhibition of vegetative growth and spore germination. Growth inhibition of propiconazole 25 EC have recorded 100% growth inhibition, which supports the findings of Pandey and Kanaujia (2009) who observed fungicides propiconazole was proved to be highly detrimental for *B. bassiana.*

The above study purpose was checking the compatible action between different pesticides (insecticides and fungicides) and entomopathogen (*B. bassiana*). Compatibility between pesticides and *B. bassiana* means there are no detrimental effects of pesticides on entomopathogen (*B. bassiana*), so we can use entopathogenic fungi with pesticides for better management of insect pest in the field without any harmful effect on the environment.

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