Journal of Entomology and Zoology Studies

Available online at www.entomoljournal.com

E-ISSN: 2320-7078
P-ISSN: 2349-6800
JEZS 2018; 6(6): 205-207

Received: 24-09-2018
Accepted: 26-10-2018

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Compatibility Studies of Different Systemic Insecticides with Entomopathogenic Fungus *Beauveria bassiana* (Balsamo) Vuillemin

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Abstract
Compatibility of *Beauveria bassiana* (Balsamo) Vuillemin was studied in the laboratory condition with five commonly used insecticides with their three doses by poison food technique. The results showed that the dinotefuran 20 SG 0.005%, diafenthiuron 50 WP 0.025%, spiromesifen 22.9 SC 0.11%, spinosad 45 SC 0.007% and dimethoate 30 EC 0.015% at lower dose and dinotefuran 20 SG 0.011%, diafenthiuron 50 WP 0.050%, spinosad 45 SC 0.015% and dimethoate 30 EC 0.030% at recommended dose were rated as relatively harmless (Grade 1) and it was compatible with *B. bassiana*, while dinotefuran 20 SG 0.016%, diafenthiuron 50 WP 0.075%,spiromesifen 22.9 SC 0.34%, spinosad 45 SC 0.022% and dimethoate 30 EC 0.045% at higher dose and spiromesifen 22.9 SC 0.23% at recommended dose were" slightly harmful" (Grade 2) to *B. bassiana*.

Keywords: *Beauveria bassiana* (Balsamo) Vuillemin, compatibility, entomopathogenic fungus, insecticides

Introduction

Several species of natural enemies including insects, mites, fungi, bacteria, virus and protozoa have been recovered from different ecosystem, only few are efficient in controlling the pest population. Among them, the entomopathogenic fungus, *Beauveria bassiana* (Balsamo) Vuillemin is a very potential bio-control agent for pest management. It is also very effective and widely used bio-pesticide, which controls various pests of different crops. It can be developed in laboratory for use as mycoinsecticide agent. Earlier, scientists have first time recorded this fungus occurring naturally in India [10]. This fungus effects on more than 150 insect species [12]. Likewise, due to its wide undefined host range, *B. bassiana* referred as "Magnificent pathogen" [4]. *B. bassiana* did not have an adverse impact on humans, livestock, birds, fish, beneficial insects, crops, waterways or groundwater resources [13].

There is a need to explore alternatives like Integrated Pest Management (IPM) encompassing available pest control methods and techniques in order to reduce the sole dependence on insecticides. Combined utilization of selective insecticides in association with fungus pathogens can increase the efficiency of control by reduction of the amount of applied insecticides, minimizing environmental contamination hazards and pest resistance [9, 11]. The use of insecticides with entomopathogenic fungus, *B. bassiana* has been laid earlier for the management of insect pests. So, it is a need to study the compatibility between entomopathogenic fungi and pesticides used in crop protection. Some of the earlier reports stated that *B. bassiana* was compatible with avermectin and the pyrethroids formulations which was more compatible with *B. bassiana* [2]. However, farmers used sole insecticides for the management of insect pests which shows deleterious effects on environment and human beings. So, there is need of incorporating the biological agents such as *B. bassiana* with such insecticides after their compatibility studies which will minimize these effects. These studies underline the possibility of application of less harmful insecticides together with entomopathogenic fungi in the IPM programs.

Materials and Methods

The present study was conducted at Biocontrol Research Laboratory, Department of Entomology, College of Agriculture, Junagadh Agricultural University, Junagadh, Gujarat, India during 2017-18. Five insecticides with their three doses were evaluated by poisoned food technique in Potato Dextrose Agar (PDA) medium [3]. The insecticides dose was calculated on the basis of lower, recommended and higher for field application rate and each treatment of insecticides was repeated thrice.

Twenty ml of PDA medium was sterilized in individual boiling tubes and the insecticide 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. An agar disc along with mycelium mat of *B. bassiana* was cored from the periphery of 10 days old colony of *B. bassiana* by 5 mm diameter cork borer and transferred in to the centre of the PDA plate. Growth medium (PDA) without insecticide but inoculated with mycelial disc served as untreated check.

The growth of entomogenous fungus was recorded, when full growth obtained with control (10^{th} day of inoculation). After that the growth observed on the medium was recorded and following grades were given.

Table 1

Grades	Percent growth
0	0%
1	1-20%
2	21-40%
3	41-60%
4	61-80%
5	81-100%

The grades given after the observed growth was imparted for the calculation of the growth inhibition percentage of *B. bassiana* by using following formula.

$$X = \frac{Y - Z}{Y} \times 100$$

Where,

X- Percent growth inhibition

Y- Grade obtained to the growth in control plate

Z- Grade obtained to the growth in treated plate

The growth inhibition of the fungus obtained from each treatment and it was further classified in evaluation categories of following 1- 4 scoring index in toxicity tests *in vitro* according to following Hassan's classification scheme [7].

Table 2

Scoring Index	Toxicity
1	Harmless (<50% reduction in beneficial capacity)
2	Slightly harmful (50-79%)
3	Moderately harmful (80-90%)
4	Harmful (>90%)

Results and Discussion

The result demonstrated in Table 1 discovered that the growth inhibition percentage of fungus in different treatments was ranged from 20.00 to 73.33%. Among the different insecticides and their different doses tested, dimethoate 30 EC 0.015% showed significantly superior result from all other treatments as it recorded minimum (20.00%) growth inhibition. Dinotefuran 20 SG 0.005% and spinosad 45 SC 0.007% were found second better treatments, with growth inhibition of 26.67% recorded in each. The next treatments were diafenthiuron 50 WP 0.025% (33.33%), spinosad 45 SC 0.015% (33.33%), dimethoate 30 EC 0.03% (33.33%), spiromesifen22.9 SC0.011% (40.00%), dinotefuran 20 SG 0.011% (46.67%) and diafenthiuron 50 WP 0.05% (46.67%). The remaining treatments, spiromesifen22.9 SC0.023% (53.33%), dimethoate 30 EC 0.045% (53.33%), spinosad 45 SC 0.022% (60.00%), diafenthiuron 50 WP 0.075% (60.00%) and dinotefuran 20 SG 0.016% (60.00%) showed higher growth inhibition than prior treatments. Spiromesifen 22.9 SC 0.34% showed the highest growth inhibition of 73.33%.

Perusal of data revealed that lower and recommended doses of all the insecticides gave less than 50% growth inhibition, which was categorized as Grade 1 (Harmless), except spiromesifen which showed more than 50% growth inhibition categorized as Grade 2 (Slightly harmful). The higher dose of insecticides showed growth inhibition between 50 to 79%, which were categorized as Grade 2 (Slightly harmful).

Data showed that at lower concentration, all treatments were highly compatible (Grade 1) with *B. bassiana*, among that dimethoate showed least growth inhibition (20.00%), while spiromesifen showed maximum growth inhibition (40.00%) among all treatments. So, dimethoate was safest among all the treatments at lower concentration. At recommended concentration of insecticides, all treatments were highly compatible (Grade 1) with *B. bassiana* except spiromesifen, which showed moderately compatible result (Grade 2). Among all the treatments, spinosad and dimethoate showed minimum growth inhibition (33.33%), while spiromesifen showed maximum growth inhibition (53.33%). The insecticides, dimethoate and spinosad were found safest among all treatments at recommended concentration. At higher concentration, all insecticides showed moderately compatible result (Grade 2).

The results unveiled that lower dose of any treatments showed less growth inhibition, while as doses of insecticides increased, the growth inhibition percentage was also increased. Maximum growth inhibition was observed in higher concentration of insecticides. Dimethoate at every dose (recommended, lower and higher) found compatible with *B. bassiana* followed by spinosad. Among all insecticide treatments, dimethoate was found highly compatible with *B. bassiana* and inhibited minimum growth of fungus [5]. In present findings, the growth inhibition of spinosad 45 SC was recorded below 50%, which supports the findings several authors who observed the lowest (23.22%) per cent of inhibition of colony diameter of *B. bassiana* in sample treated with 0.0018 % concentration of spinosad 45 SC [6]. Isolates of *B. bassiana*, URPE-6 and UFPE-19 were compatible with spinosad which supports the present results [8]. However, several researchers reported that spinosad 45 SC is slightly toxic to *B. bassiana* which is not in agreement with present studies [1].

Table 1: Effect of insecticides on the growth of *B. bassiana*

Sr. No.	Treatments	Dose/litre	Growth Inhibition (%)	Grade
1	Dinotefuran 20 SG 0.005%	0.26 g	*31.00 (26.67)	1
2	Dinotefuran 20 SG 0.011%	0.53 g	43.09 (46.67)	1
3	Dinotefuran 20 SG 0.016%	0.79 g	50.77 (60.00)	2
4	Diafenthiuron 50 WP 0.025%	0.50 g	35.23 (33.33)	1
5	Diafenthiuron 50 WP 0.050%	1.00 g	43.08 (46.67)	1
6	Diafenthiuron 50 WP 0.075%	1.50 g	50.77 (60.00)	2
7	Spiromesifen22.9 SC0.011%	0.50 ml	39.23 (40.00)	1
8	Spiromesifen22.9 SC 0.023%	1.00 ml	46.92 (53.33)	2
9	Spiromesifen22.9 SC 0.034%	1.50 ml	59.21 (73.33)	2
10	Spinosad 45 SC 0.007%	0.15 ml	31.07 (26.67)	1
11	Spinosad 45 SC 0.015%	0.30 ml	35.25 (33.33)	1
12	Spinosad 45 SC 0.022%	0.45 ml	50.77 (60.00)	2
13	Dimethoate 30 EC 0.015%	0.50 ml	26.57 (20.00)	1
14	Dimethoate 30 EC 0.030%	1.00 ml	35.25 (33.33)	1
15	Dimethoate 30 EC 0.045%	1.50 ml	46.92 (53.33)	2
16	Control	-	16.43 (0.00)	-
S.Em.±			1.6111	-
C.D. at 5 %			4.5113	-
C.V. %			12.21	-

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

Grades*: 1 = harmless (<50% reduction in beneficial capacity), 2 = slightly harmful (50-79%), 3 = moderately harmful (80-90%), 4 = harmful (>90%)

Conclusion

From the present we came to know that almost all the insecticides are compatible at their lower dose with *B. bassiana*. However, dinotefuran 20 SG 0.011%, diafenthiuron 50 WP 0.050%, spinosad 45 SC 0.015% and dimethoate 30 EC 0.030% were found relatively harmless (Grade 1) i.e. comparatively compatible with tested fungus at recommended dose but spiromesifen 22.9 SC 0.23% was recorded slightly harmful at same dose. In case of higher dose, dinotefuran 20 SG 0.016%, diafenthiuron 50 WP 0.075%, spiromesifen 22.9 SC 0.34%, spinosad 45 SC 0.022% and dimethoate 30 EC 0.045% were reported slightly harmful to the fungus. Overall it can be concluded from the present investigation that *B. bassiana* is compatible with half dose of dimethoate 30 EC 0.015% and spinosad 45 SC 0.006% and can be used for the management of insect pests in field condition.

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