

Biological Approaches for The Management of Pulse Beetle *Callosobruchus chinensis* in Red Gram

Shaik Javed, S. M. A. S. Rahman, S. J. Rahman, T. Ramesh

Received 7 March 2016; Accepted 29 April 2016; Published online 16 May 2016

Abstract Pulse beetle, *Callosobruchus* spp. (Bruchidae: Coleoptera) are major pest of stored redgram, blackgram, greengram, cowpea and other pulse grains. Pulse beetle causes a potential loss in legumes by feeding on the protein content of the grain and their damage ranges from 12–30% in developing countries. By keeping this view present study were conducted to evaluate the efficacy of biopesticide formulations on percent adult mortality and fecundity of pulse beetle in redgram. The present investigations were found that among all the bio pesticide formulations wettable powder formulation of *Bacillus thuringiensis* shows the highest percent of adult

mortality 46.67% 72 hours after treatment and lowest fecundity 36.33% and highest percent ovipositional difference (POD) 86.20% followed by others.

Keywords Pulse beetle, Bio-pesticide formulations, Percent adult mortality, POD, Red gram.

Introduction

Pulses have been considered as the poor mans meat which gives an important source of dietary protein and have property of mantiaing the restoring soil fertility through biological nitrogen fixation. Pulses are conserving and improving physical properties of soil by virtue of their alef fall and after crop harvesting incorporation into the soil. Pulse crops leave behind reasonable quantity of nitrogen in the soil and add upto 30 kh N/ha [1].

The global pulse market is estimated at 60 million tonnes. In the world, pulses are grown by 171 countries. India is the world's largest producer and the largest consumer of pulses. A large amount (15–20%) of agricultural production in the world is lost every year due to insect infestation. Out of this, 8% production is lost every year due to insect infestation alone in storage. In India, losses caused by insects accounted for 6.5% of stored grain. The damage in storage of pulses is more crucial than in the field [2]. Twelve percent loss due to pulse beetle infestation in pulses stored in warehouse. There are 1,300 species of bruchids belonging to 56 genera in 5 sub-families

S. M. A. S. Rahman, S. J. Rahman, T. Ramesh
 Department of Entomology,
 College of Agriculture, Rajendranagar,
 Hyderabad, India

S. Javed*
 Harithanilayam, PJTSAU,
 College of Agriculture, Rajendranagar,
 Hyderabad, India
 e-mail : skjaved789@gmail.com

*Correspondence

Table 1. Effect of biopesticidal formulations on percent adult mortality of *Callosobruchus chinensis* in redgram at different hours after treatment. Figures in parentheses are square root transformed values. The values denoted by a common letter are not significantly different from each other as per DMRT.

Sl. No.	Treatments	Adult mortality (%) after 24 48 72 hours hours hours		
1	T ₁ <i>Metarhizium anisopliae</i> -WP	16.67 ^c (4.19)	26.67 ^c (5.26)	43.33 ^c (6.66)
2	T ₂ <i>Verticillium lecanii</i> -WP	13.33 ^c (3.77)	21.67 ^c (4.76)	31.67 ^b (5.70)
3	T ₃ <i>Beauveria bassiana</i> -WP	16.67 ^c (4.19)	26.67 ^c (5.26)	41.67 ^c (6.53)
4	T ₄ <i>Bacillus thuringiensis</i> -WP	18.33 ^c (4.39)	30.00 ^c (5.56)	46.67 ^c (6.90)
5	T ₅ <i>Bacillus thuringiensis</i> -Liq	8.33 ^b (3.03)	13.33 ^b (3.77)	30.00 ^b (5.56)
6	T ₆ <i>Metarhizium anisopliae</i> -Oil	8.33 ^b (3.03)	13.33 ^b (3.77)	26.67 ^b (5.26)
7	T ₇ <i>Verticillium lecanii</i> -Oil	6.67 ^b (2.74)	13.33 ^b (3.77)	25.00 ^b (5.08)
8	T ₈ <i>Beauveria bassiana</i> -Oil	8.33 ^b (3.03)	13.33 ^b (3.77)	30.00 ^b (5.56)
9	T ₉ Control	0.00 ^a (0.70)	0.00 ^a (0.70)	0.00 ^a (0.70)
10	CD at 5%	0.71	0.59	0.65
11	SE(m) ±	0.24	0.20	0.22

Table 2. Effect of biopesticidal formulations on fecundity and percent ovipositional difference of *Callosobruchus chinensis* in redgram at 7 days after treatment. Figures in parentheses are square root transformed values. The values denoted by a common letter are not significantly different from each other as per DMRT (Duncan's multiple range test).

Sl. No.	Treatments	No. of POD eggs laid (%)	
1	T ₁ <i>Metarhizium anisopliae</i> -WP	38.67 ^a (6.29)	85.28 ^f (9.28)
2	T ₂ <i>Verticillium lecanii</i> -WP	88.00 ^{bc} (9.43)	66.55 ^d (8.21)
3	T ₃ <i>Beauveria bassiana</i> -WP	45.33 ^a (6.76)	82.65 ^f (9.14)
4	T ₄ <i>Bacillus thuringiensis</i> -WP	36.33 ^a (6.09)	86.20 ^f (9.33)
5	T ₅ <i>Bacillus thuringiensis</i> -Liq	75.00 ^b (8.71)	71.49 ^e (8.51)
6	T ₆ <i>Metarhizium anisopliae</i> -Oil	82.33 ^b (9.12)	68.69 ^{de} (8.34)
7	T ₇ <i>Verticillium lecanii</i> -Oil	115.33 ^d (10.78)	56.10 ^b (7.55)
8	T ₈ <i>Beauveria bassiana</i> -Oil	101.00 ^{cd} (10.09)	61.59 ^c (7.91)
9	T ₉ Control	263.00 ^e (16.24)	0.00 ^a (0.70)
10	CD at 5%	0.75	0.25
11	SE(m) ±	0.25	0.08

in the world and in India 97 species of bruchids in 11 genera have been recorded. Among the stored grain pests of pulses, the pulse beetle, *Callosobruchus* spp. are the economic pest [3]. In India, two species named *Callosobruchus maculatus* F. and *Callosobruchus chinensis* L. are commonly known as pulse beetles [4].

C. chinensis, known as pulse beetle, attack all pulses but chickpea and beans are significantly affected both qualitatively and quantitatively. The beetle completes its life cycle within 25–34 days during summer while it takes 40–50 days in winter [5]. Infestation starts right from the field and continues to the store, attains its peak in January and continues till February.

Many synthetic insecticides have been found effective against pulse beetle but some are hazardous due to their residues in food [6]. Therefore, there has been an increased need to explore suitable alterna-

tive methods of pest control. Several alternate studies are going through-out the world. Among that some of the entomopathogens shows effective control of pulse beetle [7] or in combination with other control treatments such as diatomaceous earth [8–11]. A number of biopesticide formulations are available which will be eco-friendly and biodegradable. Biopesticides are mostly compatible with other techniques and shows character having non-residual and non-toxic. Because of all these characters it plays an important role in integrated pest management (IPM). Hence, the present study was conducted to evaluate the efficacy of biopesticide formulation on percent adult mortality and fecundity of pulse beetle in redgram.

Materials and Methods

Insect culture

The pulse beetle, *Callosobruchus chinensis* L. (Bru-

chidae: Coleoptera) were used as a test insect, as it is the major storage pest of pigeonpea in India, attacking different pulses under field and storage conditions accounting for huge losses. The laboratory studies were conducted at AICRP on Biological Control, ARI, Rajendranagar, Hyderabad, Andhra Pradesh.

Lab culture of *Callosobruchus chinensis* L. were carried out on chick pea seed to obtain inoculum cultures. The infested seed kept in plastic container and covered with muslin cloth till the emergence of adults. Healthy adults emerging from the container were shifted to another plastic container, provided with clean chickpea seeds for oviposition and maintained at $25 \pm 5^\circ$ temperature and 65% relative humidity in BOD incubator. The containers were not disturbed till the emergence of the adults. Freshly emerging subsequent generations were used for further experiments.

Identification of the test insect

The adults are 32–3 mm long brownish beetles. Antennae are pectinate in male and slightly serrate in female. Abdomen is not covered by the elytra in case of females.

Different biopesticides which are used

Four biopesticided, viz. *Beauveria bassiana*, *Metarhizium anisopliae*, *Verticillium lecanii* and *Bacillus thuringiensis* both wettable powder and oil formulations were used for the experimental study. Details of the treatments and their concentrations are given below : 1. *Beauveria bassiana* wettable powder-8 g/kg seed, 2. *Beauveria bassiana* oil based-8 ml/kg seed, 3. *Metarhizium anisopliae* Wettable powder-8 g/kg seed, 4. *Metarhizium anisopliae* oil based-8 ml/kg seed, 5. *Verticillium lecanii* wettable powder-8 g/kg seed, 6. *Verticillium lecanii* oil based-8 ml/kg seed, 7. *Bacillus thuringiensis* powder from-8 g/kg seed, 8. *Bacillus thuringiensis* liquid form-8 ml/kg seed, 9. Untreated control.

Bio assay studies were conducted treatment wise in transparent plastic containers with three replica-

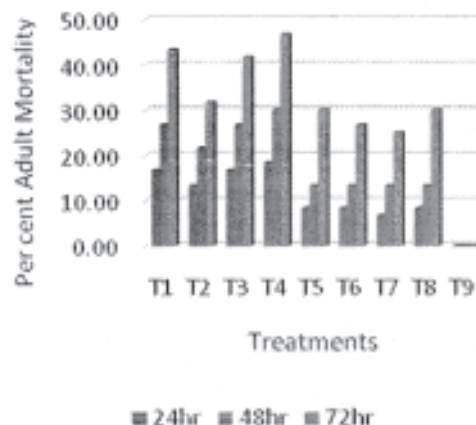


Fig. 1. Impact of biopesticidal formulations on percent adult mortality of *Callosobruchus chinensis* in redgram at different hours after treatment.

tions for each treatments. The treatmental formulations were mixed thoroughly with seeds by using plastic polythene bags before the initiation of experiment.

Mortality studies

Different biopesticides were evaluated at doses of 0 (control), 8 g/kg seed in wettable powder formulations and 8 ml/kg seed in oil based formulations in separate transparent plastic containers having 500 ml capacity. The required amount of biopesticides were mixed thoroughly with 100 g of seeds of different pulses. Ten pairs of 0–24 hour's old adults were released into each container and the containers were capped. The number of dead beetles were recorded after 24, 48 and 72 hours of treatment respectively. Mortality was considered when the beetle did not respond to gentle pressure using a fingertip. After each counting, dead bruchids were removed.

Fecundity studies

About 100 g of the treated seeds of different pulses as per the treatment concentrations of test bio pesticides were kept on transparent plastic containers. Ten pairs of 0–24 hour's old adults (10 males + 10 females) were released in each container and kept undisturbed

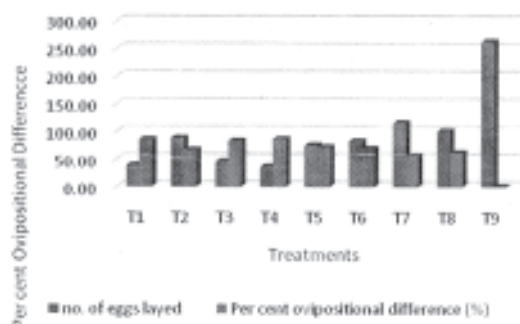


Fig. 2. Impact of biopesticidal formulations on fecundity and percent ovipositional difference of *Callosobruchus chinensis* in redgram at 7 days after treatment.

for seven days allowing to lay the eggs. After 7 days number of eggs laid were (POD) was calculated by using the formula.

$$POD = \frac{C_s - T_s}{C_s} \times 100$$

C_s – No of eggs laid on control seeds, T_s – No of eggs laid on treated seeds.

Results and Discussion

Mortality studies

At 24 hours after treatment

During the experimentation it has been found that among all the treatments wettable powder (WP) formulation of *B. thuringiensis* fared better in controlling *Callosobruchus chinensis* with percent adult mortality (18.33%) is at par with WP formulations of *V. lecanii*, *M. anisopliae* and *B. bassiana* (Table 1). This was followed by WP formulation of *M. anisopliae* and *B. bassiana* where the percent adult mortality is 16.67% which is at par with WP formulation *B. thuringiensis* (Fig. 1). Among all the treatments oil

formulation of *V. lecanil* is found to be least effective with percent adult mortality (6.67%) which is significantly different from untreated control (0.00%).

At 48 hours after treatment

The data revealed that after treatment application it has been found that among all the treatments wettable powder (WP) formulation of *B. thuringiensis* is better in controlling *Callosobruchus chinensis* with percent adult mortality (30.00%) at par with WP formulations of *V. lecanii*, *M. anisopliae* and *B. bassiana*. This was followed by WP formulation of *M. anisopliae* and *B. bassiana* where the percent adult mortality is 26.67% which is at par with WP formulation *B. thuringiensis*. Among all treatments liquid (liq) formulation of *B. thuringiensis*, oil formulations of *V. lecanii*, *M. anisopliae* and *B. bassiana* are found to be least effective with percent adult mortality (13.33%) which is almost significantly different from untreated control (0.00%).

At 72 hours after treatment

The results revealed that among all the treatments wettable powder (WP) formulation of *B. thuringiensis* fared better in controlling *Callosobruchus chinensis* with percent adult mortality (46.67%) is at par with WP formulations of *M. anisopliae* and *B. bassiana*. This was followed by WP formulation of *M. anisopliae* where the percent adult mortality is 43.33%, which is at par with WP formulation *B. thuringiensis*. Among all treatments oil formulation of *V. lecanii* is found to be least effective with percent adult mortality (25.00%) which is almost significantly different from untreated control (0.00%).

Fecundity studies

During the experimentation it has been found that among all the treatments WP formulation of *B. thuringiensis* fared better in controlling *Callosobruchus chinensis* with lowest fecundity (36.33) is at par with WP formulations on *M. anisopliae*, *B. bassiana* (Table 2). This was followed by WP formulation of *M. anisopliae* where the fecundity is (38.67)

which is at par with WP formulation *B. bassiana*. Among all treatments oil formulation of *V. lecanii* is found to be least effective with fecundity (115.33) which is almost significantly different from untreated control (263.00) (Fig. 2).

During the experimentation it has been found that among all the treatments WP formulation of *B. thuringiensis* fared better in controlling *Callosobruchus chinensis* with percent ovipositional difference (POD) (86.20%) which is at par with WP formulations of *M. anisopliae*, *B. bassiana*. This was followed by WP formulation of *M. anisopliae* where the POD (85.28%) which is at par with WP formulation *B. thuringiensis*. Among all treatments oil formulation of *V. lecanii* is found to be least effective with POD (56.10%) which is almost significantly different from untreated control (0.00%).

Among the different treatments of bio pesticide formulations are used to evaluate the percent adult mortality of pulse beetle at different hours after treatment and by comparing the number of eggs laid and percent ovipositional difference (POD) of pulse beetle, *C. chinensis* at 7 days after treatment among all the bio pesticides, WP formulation of *B. thuringiensis* shows the highest percent adult mortality and lowest fecundity and highest POD followed by others, this may be due to the binding affinity of Bt toxin to targeted sites (as this toxin jeopardize the alimentary system) of the pulse beetle or may be due to the presence of novel crystal proteins which exhibit insecticidal activity against the pulse beetle. These findings are in conformity with works of Masoud and Bahar [12] in which they comparing the isolates of *B. bassiana* and *M. anisopliae*, out of which *Beauveria bassiana* showed more reduction of oviposition in the *Orycteselegans* and also conformity with works of Batta [13], they reported mortality in F_1 adults was 28.9–47.5% in rice weevil. Cherry et al. [7] concluded that *B. bassiana* 0362 was consistently more virulent than *M. anisopliae* 0351. Neda et al. [14] found that combinations between diatomaceous earth and the fungal isolates at lethal and sub-lethal doses achieved high mortality rates in pulse beetle.

Further research is required to explore some new bio pesticide formulations, which can more efficiently

be utilized for the food-safety purpose and to overcome the dilemma of health hazards and environmental pollution.

References

1. Pokharkar PK, Chauhan NR (2010) Bio efficacy of botanicals material against *Callosobruchus chinensis* (Linnaeus) in stored chickpea. Int J Pl Prot 3 : 72–76.
2. Soundararajan RP, Chitra N, Geetha S, Poorani J (2012) Biological control of bruchid *Callosobruchus maculatus* (F.) in blackgram. J Biopest 5 : 192–195.
3. Ahmed KS, Itino T, Ichikawa T (2003) Duration of developmental stages of *Callosobruchus chinensis* (Coleoptera: Bruchidae) on azuki bean and the effects of neem and sesame oils at different stages of their development. Pak J Biol Sci 6 : 332–335.
4. NRI (Natural Resource Institute) (2003) Manual for handling and storage of food grains in tropical and sub-tropical areas of *Sitophilus* spp. on stored sorghum. MSc thesis. Alemaya Univ, Ethiopia.
5. Ghosh SK, Dubey SL (2003) Integrated management of stored grain pests. Int Book Distributing Co, Lucknow, India, pp 263.
6. Jolli RB, Karabrantanal SS, Jayaprakash TC (2005) Influence of pesticides and storage methods in the management of pulse beetle (*Callosobruchus* spp.) and their effects on seed viability in moth bean. J Ent Res 29 : 159–162.
7. Cherry AJ, Abalo P, Hell K (2004) A laboratory assessment of the potential of different strains of the entomopathogenic fungi *Beauveria bassiana* (Balsamo) Vuilleum and *Metarhizium anisopliae* (Metschnikoff) to control *Callosobruchus maculatus* F. (Coleoptera: Bruchidae) in stored cowpea. J Stored Prod Res 41 : 295–309.
8. Akbar W, Lord JC, Nechols JR, Howard RW (2004) Diatomaceous earth increases the efficacy of *Beauveria bassiana* against *Triboliumcastaneum* larvae and increases conidial attachment. J Econ Entomol 97 : 273–280.
9. Athanassiou CG (2004) Feasibility of using *Beauveria bassiana* plus diatomaceous earth against three stored-product beetle species. In: Stengard-Hansen L, Wakefield M, Lukas J, Stejskal V (eds). Proc the 4th Meeting of COST Action 842, Working Group 4, Athens, 2004, pp 50–52.
10. Smith SM, Moore D, Oduor GI, Wright DJ, Chandi EA, Agano JO (2006) Effect of wood ash and conidia of *Beauveria bassiana* (Balsamo) Vuillemin on mortality of *Prostephanus truncatus* (Horn). J Stored Prod Res 42 : 357–366.
11. Vassilakos TN, Athanassiou CG, Kavallieratos NG, Vayias BJ (2006) Influence of temperature on the insecticidal effect of *Beauveria bassiana* in combination with diatomaceous earth against *Rhyzoperthdominica* and *Sitophilusoryzae* on stored wheat. Biol Control, pp 55.
12. Masoud L, Bahar R (2012) Pathogenicity of the

- entomopathogenic fungi *Beauveria bassiana* (Balsamo) Vuillmin, *Beauveria brongniartii* Saccardo and *Metarhizium anisopliae*. Metsch to adult *Orycteselegans* Prell and effects on feeding and fecundity. Int J Agric Crop Sci 4 : 1026—1032.
13. Batta YA (2003) Production and testing of novel formulations of the entomopathogenic fungus *Metarhizium anisopliae* (Metschinkoff) Sorokin (Deuteromycotina: Hyphomycetes). Crop Prot 22 : 415—422.
14. Neda N, Ali M, Moosa S (2012) Efficacy of entomopathogenic fungi in combination with diatomaceous earth against *Callosobruchus maculatus* (Coleoptera: Bruchidae). Asian Entomologica Sinica 55 : 1282—1289.