



Field Efficacy of Biocapsules of Entomopathogenic Fungi for the Management of *Amaranthus* Leaf Webber *Spoladea recurvalis* (Fabricius) Crambidae: Lepidoptera

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Author's contribution

The sole author designed, analysed, interpreted and prepared the manuscript.

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ABSTRACT

There are many insecticides are being available for pest control in the market, but traditionally focused on killing insect pests using a variety of insecticides may lead to insecticide resistance in insect pests. Biological control methods are promising alternative methods to the chemical method. In the evaluation of the efficacy of biocapsules of *Beauveria bassiana* (Balsamo) Vuillemin and *Metarhizium anisopliae* (Metschnikoff) Sorokin, for the management of amaranthus leaf webber, *Spoladea (Hymenia) recurvalis* F., it was revealed that *Metarhizium* and *Beauveria* capsules @ three L⁻¹ sprayed twice (at weekly intervals) was effective reducing the larval population of *S. recurvalis* (83.6 and 69.9 % respectively). Lower doses of two and one capsule L⁻¹ were less effective (47.3 to 66.5 % reductions). Spraying spore suspensions of these fungi @ 10⁸ mL⁻¹ resulted in 91.2 to 100 per cent reduction, while in flubendiamide 39.35 SC, it was 89.8 per cent.

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Treatment with *Metarhizium* and *Beauveria* capsules did not affect the natural enemy population significantly with 2.33 to 3.67 plant⁻¹, while the corresponding population was 1.4 in flubendiamide 39.5 SC and 3.6 in the untreated control. The yield in the plots treated with *Metarhizium* and *Beauveria* capsules @ three L⁻¹ was higher when compared to that in the untreated plot. Therefore, Need-based production of biocontrol formulations in the form of capsules, tablets, powder, etc. should be broadcasted.

Keywords: *Spoladea recurvalis*; *Metarhizium*; *Beauveria*; biocapsules.

1. INTRODUCTION

Safe-to-eat vegetable production in the state demands the use of eco-friendly tools for managing pests. Agriculture must face the destructive activities of numerous pests like fungi, weeds, and insects which have a serious effect on food production. Global crop yield is reduced by 20 to 40 per cent annually due to plant pests and diseases [1]. With the advent of chemical pesticides, this crisis was resolved to a great extent. But the over-dependence on chemical pesticides and their eventual uninhibited use has necessitated alternatives mainly for environmental concerns. Though biopesticides cover about one per cent of the total plant protection products globally, their number and growth rate have been showing an increasing trend in the past two decades, about 175 biopesticides active ingredients and 700 products have been registered worldwide [2] sufficient.

Entomopathogenic fungi (EPF), is a widely used organic tool, especially in the case of vegetable production, which renders a cultivation practice that is free from pesticide residues. They constitute a group with over 750 species from 90 genera that are known to be entomopathogenic [3]. Widely studied entomopathogenic fungi belong to genera such as *Beauveria*, *Metarhizium*, *Lecanicillium*, *Hirsutella*, *Erymia* (*Zoopththora*), *Nomuraea*, *Aspergillus*, *Aschersonia*, *Paecilomyces*, *Tolypocladium*, *Leptolegnia*, *Culicinomyces*, *Coelomomyces*, and *Lagenidium* [4], of which, *Beauveria* spp., *Metarhizium* spp., *Lecanicillium* spp., and *Isaria* spp. have been developed as successful mycoinsecticides for various groups of insect pests [5]. The main advantages of EPF are their specificity to target pests, safety for non-target organisms, high virulence, persistence, and safety for the environment and human health.

The objective of the experiment was to evaluate the comparative efficacy of *B. bassiana* and *M. anisopliae* capsules and standardize their dose in

managing defoliators, which was carried out in amaranthus. Market samples of amaranthus, the most commonly used leafy vegetable was reported to harbor pesticide residues [6]. It is attacked by several insect and non-insect pests of which its yield was reported to be hindered by major insect pests such as *Spoladea recurvalis* (F.) (beet webworm), *Spodoptera littoralis* (Boisduval) (cotton leafworm), *Hypolixus* sp. (F.) (amaranth stem weevils), *Liriomyza huidobrensis* (Blanchard) (pea leaf miner) and *Myzus persicae* (Sulzer) (green peach aphid) [7].

With the increasing awareness of the eco-friendly approach of pest management, microbial control employing the application of entomopathogens particularly fungi is found to be promising. Formulations of microbial pesticides are largely talc-based products that are bulky and hence difficult to transport and use. Furthermore, the chances of contamination and loss of viability are more in these formulations. Capsule is a stable formulation wherein the bioagent is encapsulated in coatings and thus protected from extreme environmental conditions such as UV radiation, rain, and temperature. The possibility of getting contaminated is also meager as the infective propagules are encapsulated in a protective covering. Capsules have more residual stability than spray formulations. In a field experiment conducted by James et al. [8], it was observed that inoculation of *B. bassiana* (isolate Bba 5648) conidial suspension @ 1×10^8 mL⁻¹ was more virulent to larvae of *S. recurvalis* than other strains, as it caused 100 per cent mortality of larvae within five days after inoculation. They could observe mycosis in 83 per cent of the dead larvae.

Pooru [9] reported 100 per cent cessation of movement of *S. recurvalis* larvae 72 h after treatment when *B. bassiana* was sprayed @ 10^8 CFU g⁻¹ with both doses, 10g and 20g L⁻¹. In a laboratory study, Miller [10] reported that *B. bassiana* (isolate ICIPE 725) conidial suspensions @ 1×10^8 mL⁻¹ sprayed on leaves of amaranthus caused 83 per cent mortality in

second instar larvae of *S. recurvalis* after seven days of treatment. Praveena [11] reported that *M. anisopliae* isolates SP11 and Ma4 @ 28.01×10^7 spores mL^{-1} caused mortality of 63.3 to 100 per cent, against second instar larvae of *S. recurvalis*, 14 days after inoculation. A pot culture experiment was conducted by her, to evaluate seven indigenous isolates of *Fusarium solani* (Mart.) Sacc. (SP6), *M. anisopliae* (SP7, SP8, SP9, SP11, and SP13) and *Purpureocillium lilacinum* Thorn (Samson) S10 and two NBAIR isolates Bb5, Ma4 @ 1×10^8 spores mL^{-1} against leaf webbers in amaranthus variety Arun, it was revealed that the number of plants infested by the webbers, number of webbings plant^{-1} and number of larvae web^{-1} was lowest in SP11 treatment, at 14 days after treatment. The yield recorded was the highest (50.7g plant^{-1}) in this treatment compared to others ($32.2\text{-}46.7 \text{g plant}^{-1}$). In a laboratory study, Miller [10] reported that *M. anisopliae* (isolate ICIPE 30) caused 92 per cent larval mortality in second-instar larvae of *S. recurvalis* after 4.8 days of inoculation.

2. MATERIALS AND METHODS

2.1 Source and Conservation of Entomopathogenic Fungi

The entomofungal cultures maintained in the Biocontrol Laboratory, Department of Agricultural Entomology, College of Agriculture, Vellayani, were utilized for the study. *Beauveria bassiana* (Balsamo) Vuillemin isolate Bb5 and *Metarhizium anisopliae* (Metsch.) Sorokin isolates Ma4 National Bureau of Agricultural Insect Resources (NBAIR), Bengaluru. The virulence of these entomopathogens was maintained by periodically passing them through their respective host insects. *B. bassiana* was periodically revived using the grubs of banana pseudostem weevil, *Odoiporus longicollis* Oliver, and *M. anisopliae* using the grubs of rhizome weevil *Cosmopolites sordidus* Germer. Pure and subcultures of these fungi were maintained in Potato Dextrose Agar (PDA) slants.

2.2 Preparation of Biocapsules, Mass Culturing of Fungi and Preparation of Conidial Suspension

Biocapsules of *B. bassiana* and *M. anisopliae* were prepared using 14 days old cultures incubated at ambient conditions. The protocol developed by Remya and Reji [12] was followed for capsule preparation. The fungi under study

were mass multiplied by static liquid fermentation in Sabouraud Dextrose Broth (SDB) taken in a 2L fermenter flask. Upon sporulation, the conidia were harvested. Sporulating cultures of *B. bassiana* and *M. anisopliae* (14 day old) were blended in a mixer-grinder by adding a drop of tween 20. The culture was then filtered through a double-layered muslin cloth. The filtrate served as the conidial suspension for further preparation of capsules.

2.3 Preparation of Primary Powder

Spore suspension after straining was taken in centrifuge bottles and centrifuged in a Remi R 23 centrifuge for 20 min at 4000 rpm. The spore pellet collected at the bottom of the tube was washed gently with sterile distilled water, to remove the mycelial mat adhering to it. The primary powder was prepared by mixing the spore pellet and crude chitosan in a ratio of 1:1 to obtain 10^{10} spores g^{-1} . Filling material was prepared by mixing the primary powder with chitosan in a ratio of 1:20. The empty Hydroxy Propyl Methyl Cellulose (HPMC) capsules of 0.8g were filled using a hand-operated capsule filling device as illustrated which yield 100 capsules in one set. The capsules were stored airtight under ambient conditions in plastic bottles for field evaluation studies.

3. FIELD EFFICACY OF BIOCAPSULES

The objective of the experiment was to evaluate the comparative efficacy of *B. bassiana* and *M. anisopliae* capsules and standardize their dose in managing defoliators, which was carried out in amaranthus. The experiment was carried out in the Instructional Farm College of Agriculture, Vellayani, during 2018-21, following the Package of Practices recommendations of Kerala Agricultural University (KAU, 2017), except for pest management.

Seeds of KAU variety Arun procured from the Department of Vegetable Science, College of Agriculture, Vellayani was used for the experiment. The experimental plot was laid out in Randomized Block Design (RBD) consisting of 10 treatments replicated thrice with a plot size of 2m x 2m. The treatments were as follows.

T1 - 1 *Beauveria* capsule L^{-1} , T2 - 2 *Beauveria* capsule L^{-1} , T3 - 3 *Beauveria* capsule L^{-1} , T4 - 1 *Metarhizium* capsule L^{-1} , T5 - 2 *Metarhizium* capsule L^{-1} , T6 - 3 *Metarhizium* capsule L^{-1} , T7 - *Beauveria* spore suspension @ 10^8mL^{-1} -

20 mL L⁻¹, T8 - *Metarhizium* spore suspension @ 10⁸ mL⁻¹ - 20g L⁻¹, T9 - Chemical check – flubendiamide 39.35 % SC (18.24 g a. i ha⁻¹), T10 - Untreated control.

3.1 Method of Application of Capsules and Observations

Capsules at the respective doses were dispersed in water with 0.1% tween 80. Spraying was carried out using a knapsack sprayer. The first spraying was given when 10 per cent of plants were infested and the second after one week of the first application. Observations were made on pre and post-count of larvae of the leafwebber *S. recurvalis* plant⁻¹, which was the dominant defoliator observed. For recording the larval population, three plants were selected at random from each replication and the average was worked out. The incidence of other foliage and sap-feeding insects observed throughout the crop period was also recorded. The number of natural enemy plot⁻¹ was recorded by visual counting. Average yield plot⁻¹ was noted for comparison of treatments.

4. RESULTS AND DISCUSSION

4.1 Field Efficacy of Biocapsules against Leaf Webber *S. recurvalis* in Amaranthus

4.1.1 First spraying

Analysis of data on mean larval count (Table 1) noted on the third DAS (Days After Spraying) revealed that spraying three capsules L⁻¹ of *B. bassiana* and *M. anisopliae* was equally good in reducing the population, compared to their dosages of two and one capsule L⁻¹. The mean population was 2.89 and 2.56 larvae plant⁻¹ in the first two treatments respectively, while the count was 3.22 and 3.0 when sprayed @ two capsules L⁻¹ of *Beauveria* and *Metarhizium* respectively. The mean larval count was 3.56 and 3.45 plant⁻¹ when the dosage was reduced to one capsule L⁻¹ of each of them respectively and their effect was on par with each other. Among the biocontrol treatments, it was the plots treated with a spore suspension of both the fungi that exhibited the maximum reduction in population. The mean larval count was 2.33 and 2.00 plant⁻¹ in the case of *B. bassiana* and *M. anisopliae* sprayed @ 10⁸ spores mL⁻¹. The highest reduction in population was noted in plots treated with flubendiamide 39.35 % SC @ 0.1

mL⁻¹, where the mean population was 1.67 larvae plant⁻¹. After one week, there was a narrow decline in population. Among the capsule-treated plots, the lowest population was recorded in plots treated with three capsules L⁻¹ of *M. anisopliae* (2.11 larvae plant⁻¹), which was on par with the effect of the same dose of *B. bassiana* (2.39). With the lower dose of two capsules L⁻¹, the population was 2.56 and 2.78 respectively with *Metarhizium* and *Beauveria*, which did not differ significantly. Single capsule L⁻¹ was the inferior treatment where the mean larval count recorded was 3.0 and 3.11 respectively with *Metarhizium* and *Beauveria*. The lowest larval count was recorded in spore suspension of *Metarhizium* @ 10⁸ spores mL⁻¹ which was closely followed by that of *Beauveria* @ 10⁸ spores mL⁻¹. Population in flubendiamide 39.35% SC @ 0.1 mL⁻¹ treated plot was the lowest among treatments (1.11 larvae plant⁻¹).

4.1.2 Second spraying

After three days of second spraying, among the capsule treatments, the dosage of three capsules L⁻¹ of *M. anisopliae* was the most effective treatment which was closely followed by *B. bassiana* treatment @ three capsules L⁻¹. The mean population noted was 1.44 and 1.44 and 1.67 respectively. With the lower dose of two capsules of L⁻¹, the population recorded with both fungi was on par (1.89 and 2.0 respectively). Single capsule treatment with *Metarhizium* recorded 2.5 larvae plant⁻¹ which differed significantly from the corresponding dose of *Beauveria* (2.78 larvae plant⁻¹). The mean larval count noted after 7 days was 1.34 1.89 and 1.67 1.34 2.00 plant⁻¹ respectively with the dosages of two capsules L⁻¹ of *Metarhizium* and *Beauveria* capsules which were on par with others. The corresponding larval count noted in plots treated @ one capsule L⁻¹ was 2.22 and 2.0 plant⁻¹ respectively with *Metarhizium* and *Beauveria* which were in parity with each other. Spore suspensions were found to be superior to capsules in bringing down the larval population. There was no larval population at all in the *Metarhizium*-treated plots while it was negligible in *Beauveria*-treated plots (0.33 larva plant⁻¹). The corresponding count in flubendiamide 39.35% SC treated plot was 0.44 larva plant⁻¹.

4.2 Effect on Natural Enemy Population

Table 2 reveals the mean natural enemy population in the experimental plot. The natural enemies comprised spiders such as *Tetragnatha*

sp. and *Mantis* sp. Analysis of data on the total count of natural enemies per plant revealed that there was no significant variation in their count before and after treatment. Their population did not vary significantly among the treatments even after two sprayings. It varied from 1.89 2.33 to 3.89 3.45 plot⁻¹ in biocontrol treatments, while it was 1.56 in plots treated with flubendiamide 39.35% SC, three DAT. At the end of the experimental period, the biocontrol plots recorded a population of 2.33 44 to 3.67 39 plot⁻¹ while that in flubendiamide 39.35% SC treatment it was 1.44 plot⁻¹. The natural enemy population noted in the untreated plot varied from 2.56 to 4.56 plot⁻¹ during the experimental period.

4.3 Effect of Biocapsules Treatments on the Yield of Amaranthus

Analysis of data on yield recorded from the 2x2m² plot revealed that there was significant variation among treatments. Among the biocapsules highest yield of 2.67 kg was obtained from plots treated with *Metarhizium* capsules @ three L⁻¹, which was significantly lower than the yield obtained from plots treated with its spore suspension (3.23 kg) @ 10⁸ spores mL⁻¹ as well as from plots treated with flubendiamide 39.35% SC (2.9 kg). The yield recorded from plots treated with two *Metarhizium* capsules L⁻¹ was 2.10 kg which was significantly higher than its single capsule treatment (1.20 kg) as well as from yield recorded from plots treated with two capsules L⁻¹ of *Beauveria* (1.87 kg) and one capsule of *Beauveria* (1.3 kg). The yield from the untreated plot was significantly lower (0.8 kg).

In the field experiment, the major pest observed was the leaf webber *S. recurvalis* F. the destructive defoliator pest of amaranthus. Results of this experiment revealed that spraying of *Metarhizium* capsules @ three L⁻¹ was more effective, causing an 83.69 per cent reduction in the population of larvae (Fig. 1) than *Beauveria* capsules @ three L⁻¹ (69.97 per cent). Two and one capsules of *Metarhizium* and *Beauveria* caused 66.5, 47.39, 59.36, and 52.60 per cent reduction in larval population, respectively.

Although the evaluation of capsule formulations of entomopathogenic fungi for vegetable pests is the first of its kind, various researchers have proved by now, the efficacy of *B. bassiana* and *M. anisopliae* in managing *S. recurvalis* using spore suspensions and talc-based formulations.

In a field experiment conducted by James et al. [8], it was observed that *B. bassiana* (isolate Bba 5644) conidial suspension @ 1x10⁸ mL⁻¹ caused 100 per cent mortality of the leaf webber *S. recurvalis*, where 83 per cent dead larvae showed fungal sporulation, while the isolates Bba5653 and Bba5654 caused 97 per cent mortality each and 33 per cent of the dead larvae manifested the sporulation. They also reported that Bba 5644 was virulent to larvae of *P. basalilis* larvae resulting in 100 per cent mortality of larvae within five days. In a similar study conducted by Pooru [9], there was a 100 per cent cessation of movement of *S. recurvalis* larvae 72 h after treatment when *B. bassiana* was sprayed @ 10⁸ CFU g⁻¹ with both the doses, 10g, and 20g L⁻¹.

Praveena [11] while studying the efficacy of the same isolates used in the present study viz. Bb5 and Ma4 reported that the efficacy of the indigenous isolate SP 11 of *M. anisopliae* (from Vellayani, Kerala, India) was superior as it reported the lowest number of plants infested by the webbers, number of webbings plant⁻¹, and number of larvae web⁻¹, 14 days after treatment. The mortality reported by her on *S. recurvalis* was 63.33 to 100 per cent under laboratory conditions. In her study, Bb 5 and Ma4 @ 1 x 10⁹ spores mL⁻¹ caused 46.66 and 100 per cent mortality to *S. recurvalis* larvae respectively, seven days after treatment.

In concurrence with the present study, Miller [10] reported that *M. anisopliae* (isolate ICIPE 30) @ 1x10⁸ mL⁻¹ caused 92 per cent larval mortality in the second instar larvae of *S. recurvalis* after 4.8 days of spraying which was more effective than *B. bassiana* (isolate ICIPE 725) that caused 83 per cent mortality, seven days after treatment.

Efficacy of flubendiamide 39.5% SC the chemical check used in this study is a proven insecticide for the management of defoliators of leafy vegetables such as amaranth and cabbage. Muralikrishna et al. [13] observed 100 per cent mortality of second instar larvae of *S. recurvalis* 36 h after treatment in amaranthus. So also, Sambathkumar [14] reported that flubendiamide 39.5% SC (0.1 mL L⁻¹) caused 100 per cent mortality in cabbage leaf webber, *Crociodolomia binotalis* Zeller 96 h after treatment.

Even though precisely targeted formulations of flubendiamide are expected to be safe for non-target organisms, several recent studies have shown its toxic potential on many non-target

Table 1. Efficacy of biocapsules against *Spoladea recurvalis* in amaranthus

| Treatments (L ⁻¹) | No. of capsules/ spore suspension | No. of larvae plant ⁻¹ * | | | | |
|--|-----------------------------------|-------------------------------------|-----------------------------|----------------------------|----------------------------|----------------------------|
| | | First spray | | | Second spray | |
| | | DBT | 3 DAT | 7 DAT | 3 DAT | 7 DAT |
| <i>Beauveria</i> capsule | 1 | 4.22 (2.04) | 3.56 (1.87) ^{ab} | 3.11 (1.74) ^{ab} | 2.78 (1.64) ^{ab} | 2.00 (1.56) ^{ab} |
| <i>Beauveria</i> capsules | 2 | 4.11 (2.01) | 3.22 (1.76) ^{ab} | 2.78 (1.64) ^{abc} | 2.00 (1.38) ^{bc} | 1.67 (1.45) ^{bc} |
| <i>Beauveria</i> capsules | 3 | 3.33 (1.76) | 2.89 (1.63) ^{abcd} | 2.39 (1.47) ^{bcd} | 1.67 (1.22) ^{cd} | 1.00 (1.19) ^{de} |
| <i>Metarhizium</i> capsule | 1 | 4.22 (2.05) | 3.45 (1.82) ^{ab} | 3.00 (1.69) ^{ab} | 2.50 (1.53) ^{abc} | 2.22 (1.61) ^{ab} |
| <i>Metarhizium</i> capsules | 2 | 4.00 (1.99) | 3.00 (1.71) ^{abc} | 2.56 (1.57) ^{abc} | 1.89 (1.34) ^{bc} | 1.34 (1.32) ^{bcd} |
| <i>Metarhizium</i> capsule | 3 | 4.11 (2.01) | 2.56 (1.59) ^{abcd} | 2.11 (1.44) ^{bcd} | 1.44 (1.19) ^{cd} | 0.67 (1.07) ^{de} |
| <i>Beauveria</i> spore suspension (biocontrol check 1) | 10 ⁸ mL ⁻¹ | 3.78 (1.93) | 2.33 (1.51) ^{bcd} | 1.78 (1.30) ^{cde} | 0.94 (0.93) ^{def} | 0.33 (0.90) ^{ef} |
| <i>Metarhizium</i> spore suspension (biocontrol check 2) | 10 ⁸ mL ⁻¹ | 3.89 (1.94) | 2.00 (1.38) ^{cd} | 1.50 (1.17) ^{de} | 0.39 (0.62) ^f | 0.00 (0.71) ^f |
| Flubendiamide 39.35 % SC (chemical check) | 18.24 g a.i ha ⁻¹) | 4.33 (2.08) | 1.67 (1.29) ^d | 1.11 (1.04) ^e | 0.67 (0.80) ^{ef} | 0.44 (0.97) ^{def} |
| Untreated control | - | 4.11 (2.02) | 3.89 (1.96) ^a | 3.67 (1.90) ^a | 3.67 (1.88) ^a | 3.06 (1.87) ^a |
| CD (0.05) | - | NS | (0.38) | (0.37) | (0.40) | (0.35) |

NS - Not Significant. Values in the parentheses are square root transformed,

* Mean of three replications, DBT: Day before treatment: DAT - Days after treatment

Table 2. Effect of entomopathogenic fungi on natural enemy population in amaranthus

| Treatments (L ⁻¹) | No. of capsules/ spore suspension | No. of natural enemies plot ⁻¹ * | | | | |
|---|-----------------------------------|---|-------------|-------------|--------------|-------------|
| | | First spray | | | Second spray | |
| | | DBT | 3 DAT | 7 DAT | 3 DAT | 7 DAT |
| <i>Beauveria</i> capsule | 1 | 2.44 (1.56) | 2.67 (1.78) | 2.45 (1.54) | 2.67 (1.62) | 2.67 (1.61) |
| <i>Beauveria</i> capsules | 2 | 3.11 (1.76) | 2.50 (1.72) | 3.05 (1.74) | 2.78 (1.66) | 3.67 (1.91) |
| <i>Beauveria</i> capsules | 3 | 3.44 (1.85) | 3.45 (1.98) | 3.11 (1.75) | 1.89 (1.28) | 2.44 (1.52) |
| <i>Metarhizium</i> capsule | 1 | 3.67 (1.91) | 3.17 (1.90) | 2.33 (1.52) | 2.33 (1.49) | 2.78 (1.64) |
| <i>Metarhizium</i> capsules | 2 | 2.89 (1.68) | 2.00 (1.47) | 3.89 (1.96) | 3.22 (1.79) | 3.39 (1.83) |
| <i>Metarhizium</i> capsule | 3 | 3.00 (1.73) | 3.22 (1.93) | 3.33 (1.81) | 3.28 (1.79) | 2.33 (1.52) |
| <i>Beauveria</i> spore suspension (biocontrol check 1) | 10 ⁸ mL ⁻¹ | 2.39 (1.51) | 2.89 (1.82) | 3.22 (1.79) | 3.11 (1.76) | 3.61 (1.90) |
| <i>Metarhizium</i> spore suspension (biocontrol check 2) | 10 ⁸ mL ⁻¹ | 2.89 (1.64) | 2.33 (1.66) | 2.78 (1.66) | 2.67 (1.63) | 2.67 (1.63) |
| Flubendiamide 39.35 % SC (chemical check) | 18.24 g a.i ha ⁻¹ | 1.89 (1.37) | 1.56 (1.43) | 2.67 (1.61) | 2.17 (1.46) | 1.44 (1.18) |
| T10-Untreated control | - | 2.78 (1.61) | 2.56 (1.73) | 4.56 (2.11) | 3.89 (1.96) | 3.67 (1.91) |
| CD (0.05) | - | NS | NS | NS | NS | NS |

*Plot size 2m x 2m. The mean of three replications. Figures in parentheses are square root transformed values. DBT: Day before treatment; DAT - Days after treatment. NS - Nonsignificant

Table 3. Effect of biocapsules on yield of amaranthus

| Treatments (L⁻¹) | Yield (kg plot⁻¹) * |
|---|---------------------------------------|
| 1 <i>Beauveria</i> capsule | 1.30 ^g |
| 2 <i>Beauveria</i> capsules | 1.87 ^f |
| 3 <i>Beauveria</i> capsules | 2.30 ^d |
| 1 <i>Metarhizium</i> capsule | 1.20 ^g |
| 2 <i>Metarhizium</i> capsules | 2.10 ^e |
| 3 <i>Metarhizium</i> capsules | 2.67 ^c |
| <i>Beauveria</i> spore suspension @ 10 ⁸ mL ⁻¹ (biocontrolcheck 1) | 3.10 ^a |
| <i>Metarhizium</i> spore suspension @ 10 ⁸ mL ⁻¹ (biocontrol check 2) | 3.23 ^a |
| Flubendiamide 39.35 % SC (18.24 g a.i ha ⁻¹) (chemical check) | 2.90 ^b |
| Untreated control | 0.80 ^h |
| CD (0.05) | 0.20 |

*Plot size 2m x 2m. The mean of three replications.

Values sharing the same alphabets in superscript are statistically on par based on ANOVA

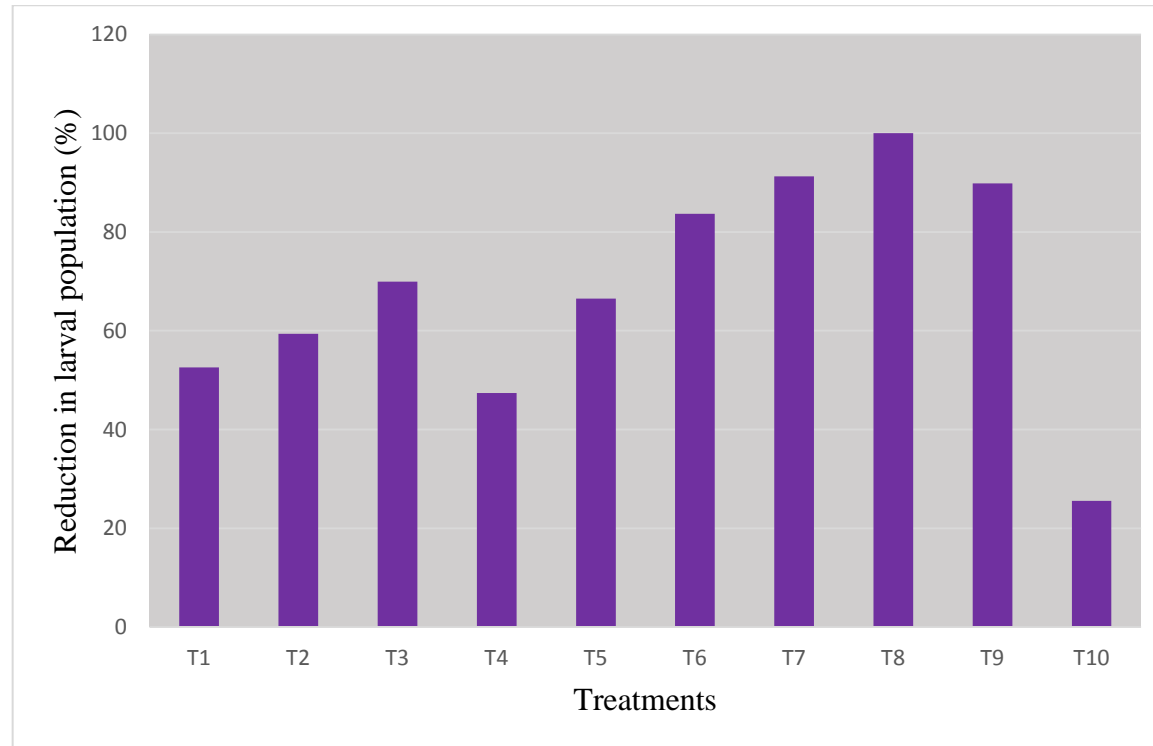


Fig. 1. Effect of biocapsules against *Spoladea recurvalis* in amaranthus

T1 - *Beauveria capsule* @ $1 L^{-1}$ T6 - *Metarhizium capsule* @ $3 L^{-1}$
T2 - *Beauveria capsule* @ $2 L^{-1}$ T7 - *Beauveria spore suspension* @ $10^8 mL^{-1}$
T3 - *Beauveria capsule* @ $3 L^{-1}$ T8 - *Metarhizium spore suspension* @ $10^8 mL^{-1}$
T4 - *Metarhizium capsule* @ $1 L^{-1}$ T9 - *Flubendiamide 39.35 SC* ($18.24 g a.i ha^{-1}$)
T5 - *Metarhizium capsule* @ $2 L^{-1}$ T10 - *Untreated control*

organisms. In a study by Sarkar et al. [15] it was observed that treatment concentrations (0.5, 1, 2, 5, 10, and 20 μgml^{-1}) of flubendiamide, inhibited acetylcholinesterase activity in third-instar larvae of *Drosophila melanogaster* Meigen indicating its neurotoxic potential. In addition, larvae exposed to flubendiamide also manifested increased amounts of stress protein hsp 70. The larvae expressing such stress response when allowed to emerge as adults displayed severe eye structure. Another study by Yan et al. [16], reported that extensive application of flubendiamide has led to increasingly prominent resistance in diamondback moth, *Plutella xylostella* (L.), where they detected a point mutation (G4946E) that caused flubendiamide resistance in it. Acute and joint toxicity of flubendiamide was reported by Wei et al. [17] on Chinese tiger frog *Hoplobatrachus chinensis* Wiegmann tadpoles. Furthermore, alterations in the protein metabolism of freshwater fish *Labeorohita* F. Hamilton have been reported by Nirmalakallagadda and Rathnamma [18]. Disruption of enzyme activity in tropical soil after flubendiamide application was reported by [19]. Consistently, Liu et al., [20] reported that exposure to flubendiamide could cause oxidative stress and DNA damage in earthworms *Eisenia fetida* Savigny. Chronic flubendiamide exposure could induce oxidative stress in water buffalo *Bubalus bubalis* L. calves [21].

5. CONCLUSION

Eco-friendly management methods for controlling insect pests are needed to reduce the continued application of insecticides that are presently used as the main method for pest control. Safe and sustainable methods using bioagents, predators, parasitoids, and physical and mechanical methods should be developed to target numerous insect pest species in such a way that it is obtainable to the common man. Need-based production of biocontrol formulations in the form of capsules, tablets, powder, etc. should be broadcasted.

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COMPETING INTERESTS

Author has declared that no competing interests exist.

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