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Evaluation of efficacy of entomopathogens against *Solenopsis geminata* (Fabricius) (Hymenoptera: Formicidae)

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ABSTRACT

Solenopsis geminata is a non – native ant species often colonizes disturbed habitats. Experiments were carried out at Annamalai University to test the efficacy of *Beauveria bassiana* and *Metarhizium anisopliae* against *S. geminata* mounds. One day after the treatment of *B. bassiana* there was reduction of original mounds in three doses viz., 10, 7 and 5g. New mounds appeared in doses of 10 and 7g. Disappearances of new mounds were found in doses of 7 and 10g at 2 and 3 days after treatment. Dead *S. geminata* workers were observed two days after treatment in the dose of 5g. Maximum number of new mounds was found in the 10g dosage. In *M. anisopliae* treated mounds 3 days after treatment control was evident in 10g dose on *S. geminata* original mounds. Also 7 and 5g doses obtained control at 5 days after treatment while 3g dose did not showed any effect on original mounds. No new mounds were found in 3, 5, 7 and 10g doses.

Key words: *Solenopsis geminata, Mound treatment, Beauveria bassiana, Metarhizium anisopliae*

Introduction

Invasive species is the present threat to global agriculture. They are the major cause of crop loss and can negatively affect food security in world (Paini *et al*., 2016). Alien species are exotic organisms that occur outside their natural adapted habitat and dispersal potential. Many alien species support our farming and forestry systems in a considerable manner. However, some of the alien species become invasive when they are introduced deliberately or unintentionally outside their natural habitats into new areas where they express the capability to establish, invade and compete native species. (Sujay *et al*., 2010). *S. geminata* often colonizes disturbed habitats (Perfecto, 1991). It is capable of colonizing most types of soils. It occurs in shaded orchards and woods (Wilson and Brown, 1958), as well as open areas. Habitat types vary greatly (Way *et al*., 1998). However, comparing to the management programmes developed for the red imported fire ant, *S. invicta*, only fewer management programmes have been specifically developed for *S. geminata* (George and Narendran, 1987; Porter *et al*., 1997).

Management of fire ants are usually accomplished with broadcast or individual mound treatments by using baits or traditional contact insecticides. Individual mound treatments generally provide the most rapid control of colonies (Vogt and Appel, 1996); however, mound relocations frequently occur (Lemke and Kissam, 1987). Even though many insecticide products are available for fire ant control, most are synthetic organic insecticides. These chemical applications however tend to create public health and environmental concerns. To resolve that it is important that new eco-friendy strategies for controlling the tropical fire ant should be developed (Kafle and Shih, 2013). Keeping in mind the importance of *S. geminata* management in major crops the present investigation was initiated.

Materials and Methods

Fire ant nests are composed of multiple mounds and underground tunnels. Therefore, delimiting the boundaries of a colony in the field with certainity is difficult. Disturbances to colonies can cause the fire ants to either move the entire nest or split into multiple mounds. A fire ant mound was defined as an experimental unit. All mounds were dated and mapped. Active mounds present in the experimental plots before treatments were classified as original mounds; mounds forming after the treatment date were classified as new mounds. A mound was rated as an active when ≥ 30 ants were present within 20 seconds after the mound was disturbed; a mound was rated as inactive when <30 ants were observed after disturbance. Mounds were disturbed by shaking vegetation growing out of the mound or blowing on the mound, or both. Activity ratings of original and new mounds were recorded at daily intervals for the duration of each study. The commercial formulations of fungal cultures, *Beauveria bassiana* and *Metarhizium anisopliae* were used for the experiment was purchased from Perunthalaivar Kamaraj Krishi Vigyan Kendra (PKKVK), Puducherry. Ungrazed pasture with clayey soil located at Annamalai University was used for the surface application study. The pasture was divided into control and treatment areas, with a 1-m-wide untreated buffer zone separating the two areas. Ten mounds, each of which were a minimum of 15 cm in basal diameter and contained brood, were used in each area. The mounds were> 1 m apart. Immediately before each mound application in the fungus treatment area, a mound was disturbed to a depth of ≈10 cm with a trowel to expose thousands of *S. geminata* workers. A rice-fungal formulation was then scattered onto the surface of each of the ten mounds. The formulation consisted of 100 g of cooked rice with a sporulating culture of *B. bassiana* and *M. anisopliae* containing about four different concentration viz., 3g, 5g, 7g and 10g conidia by weight. In the control area, 100 g of cooked rice without fungus was applied to the surface of each of 10 mounds. Application to the control area was made in the morning, and the *B. bassiana* and *M. anisopliae* formulation was applied in the afternoon of the same day to limit exposure to heat and sunlight. The study was done from third January to eighth January 2018, when mean maximum and minimum temperatures in the study area were 28.5 ± 3 and 26.3 ± 4°C, respectively, and rainfall total 155.3 mm. Percentage of original and new mounds were calculated for both *B. bassiana* and *M. anisopliae* separately (Modified Alves and Pereira, 1989).

Results and Discussion

Efficacy of *Beauveria bassiana* on *Solenopsis geminata* mounds were presented in Table 1. The pre treatment percentage of *S. geminata* mounds in the experiment of surface application of *B. bassiana* was 100 per cent in all the four doses (3, 5, 7 and 10g of *B. bassiana*). One day after treatment percentage at original mounds were 80, 90, 90 and 100 % in the doses like 10, 7, 5 and 3g of *B. bassiana* were observed respectively. Percentage of new mounds were 20 and 10% in the doses of 10 and 7g respectively at one day after treatment of *B. bassiana* . At two days after treatment percentage of original mounds were 50, 60, 50 and 70% in 10, 7, 5 and 3g doses of *B. bassiana* respectively was noticed and the percentage of new mounds were 20% in the dose of 10g. Zero percent of new mounds were observed in all the four doses (3, 5, 7 and 10g of *B. bassiana)* after 3 days after treatment. Percentage of original mounds at three days after treatment was 20, 40, and 60 in doses of 7, 5 and 3grams of *B. bassiana* respectively. At five days after treatment percentage of original mounds were 20 in the case of 3g dose of *B. bassiana* while in 5, 7 and 10g doses it was zero per cent. In control, percentage of mounds did not reduce in case of original mounds and no new mounds were formed (Table 1).

Efficacy of *Metarhizium anisopliae* on *Solenopsis geminata* mounds were presented in Table 2. The pre-treatment percentage of *S. geminata* mounds in the experiment of surface application of *M. anisopliae* was 100 per cent in all the four doses (3, 5, 7 and 10g of *M. anisopliae*). During one and two days after treatment, percentage at original mounds was also 100.00 % in all the four doses (3, 5, 7 and 10 grams) of *M. anisopliae*. At three days after treatment percentage of original mounds were 100.00, 100.00, 100.00 and 90.00% in 3, 5, 7 and 10 gram doses of *M.*

Table 1. Efficacy of surface application of *Beauveria bassiana* against *Solenopsis geminata* mounds.

Treatment No	Dose	0 DAT		1 DAT		2 DAT		3 DAT		4 DAT		5 DAT	
		Original mounds[a] (%)	New mounds[b] (%)	Original mounds[a] (%)	New mounds[b] (%)	Original mounds[a] (%)	New mounds[b] (%)	Original mounds[a] (%)	New mounds[b] (%)	Original mounds[a] (%)	New mounds[b] (%)	Original mounds[a] (%)	New mounds[b] (%)
T1	3g	100.00	0.00	100.00	0.00	70.00	0.00	60.00	0.00	30.00	0.00	20.00	0.00
T2	5g	100.00	0.00	90.00	0.00	50.00	0.00	40.00	0.00	10.00	0.00	0.00	0.00
T3	7g	100.00	0.00	90.00	10.00	60.00	0.00	20.00	0.00	0.00	0.00	0.00	0.00
T4	10g	100.00	0.00	80.00	20.00	50.00	20.00	0.00	0.00	0.00	0.00	0.00	0.00
T5	Control	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00

[a] percentage of original mounds that remained active. (No. of active original mounds/no.of original mounds treated) × 100
[b] percentage of original mounds that are active at a new site. (No. of active new mounds/no.of original mounds treated) × 100

Table 2. Efficacy of surface application of *Metarhizium anisopliae* against *Solenopsis geminata* mounds

Treatment No	Dose	0 DAT		1 DAT		2 DAT		3 DAT		4 DAT		5 DAT	
		Original mounds[a] (%)	New mounds[b] (%)	Original mounds[a] (%)	New mounds[b] (%)	Original mounds[a] (%)	New mounds[b] (%)	Original mounds[a] (%)	New mounds[b] (%)	Original mounds[a] (%)	New mounds[b] (%)	Original mounds[a] (%)	New mounds[b] (%)
T1	3g	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00
T2	5g	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	90.00	0.00
T3	7g	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	80.00	0.00
T4	10g	100.00	0.00	100.00	0.00	100.00	0.00	90.00	0.00	80.00	0.00	60.00	0.00
T5	Control	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00

[a] percentage of original mounds that remained active. (No. of active original mounds/no.of original mounds treated) × 100
[b] percentage of original mounds that are active at a new site. (No. of active new mounds/no.of original mounds treated) × 100

anisopliae respectively. Percentage of original mounds at four days after treatment were 100.00, 100.00, 100.00 and 80.00% in doses of 3, 5, 7 and 10 grams of *M. anisopliae* respectively. At five days after treatment percentage of original mounds was 100.00, 90.00, 80.00 and 60.00% in doses of 3, 5, 7 and 10 g of *M. anisopliae* respectively. Zero percent of new mounds were observed up to five days after treatment in all the four doses 3, 5, 7 and 10g of *M. anisopliae*. In control, percentage of mounds did not reduce in case of original mounds and no new mounds were formed (Table 2).

Even at one day after treatment of *B. bassiana* there was reduction of original mounds in three doses viz., 10, 7 and 5g. Also appearance of new mounds which is an important sign of control was found in doses like 10 and 7g of *B. bassiana*. Two days after treatment considerable reduction of original mounds were seen in 10, 7 and 5g of *B. bassiana*. Disappearances of new mounds were found in doses like 7 and 10g of *B. bassiana* at 2 and 3 days after treatment respectively. Dead *S. geminata* workers were observed two days after treatment in the dose of 5g of *B. bassiana* which confirms the efficacy of *B. bassiana* at lower dose itself. Maximum number of new mounds in the highest dosage of *B. bassiana* (10g) revealed immediate response of *S. geminata* even at one day after treatment. In control no change was observed in case of original mounds.

In the present study only at

3 days after treatment control on *S. geminata* original mounds was evident in 10g of *M. anisopliae*. Also 7 and 5g doses of *M. anisopliae* showed control on *S. geminata* original mounds at 5 days after treatments while 3g dose of *M. anisopliae* did not showed any effect on original mounds. No new mounds were found in all the four doses. In control no change was observed. Kafle *et al*. (2011) revealed that the direct application of *B. bassiana* was more efficient in killing *Solenopsis invicta* workers than the bait application. The direct application of 1.6×10^7 mL^{-1}1 *Bb* conidia successfully killed 3.25 times more *S. invicta* workers than the percentage of *S. invicta* workers killed by B-bait with 1.6×10^7 mL^{-1} *Bb* conidia. They also found, a smaller number of *S. invicta* workers died by the *B. bassiana* bait formulation than by the direct application of *B. bassiana*. This might be due to the specific behaviour of the *S. invicta* workers. This is partially similar to the present study results.

Lofgren *et al*. (1975) reported that the *B. bassiana* caused 80% mortality to *S. invicta* workers within 10 DAT, but Brinkman and Gardner (2001) observed only 55% mortality as a result of being exposed to *B. bassiana* at 10 DAT. Therefore, the efficacy of *B. bassiana* against *S. invicta* may vary depending on the fungal strain and local environment. These study results are in accordance with the present observations. Oi and Pereira (1993) reported that *S. invicta* workers use a special behavioural defense against microorganisms to reduce the mortality of their workers by *B. bassiana*. The *S. invicta* worker behaviours include grooming, secretion of antibiotics, nest hygiene, avoidance and dispersal behaviours. These behaviours affect the infection rates of *S. invicta* workers exposed to *B. bassiana*; they affect the dissemination of *B. bassiana* among nest mates and the dispersal of *B. bassiana* outside the nest.

Oi *et al*. (1994) also reported that rice with Bb447 applied to the tops of mounds of *S. invicta* resulted in a maximum infection of 55% of the live ants sampled; 70% of the treated mounds remained active or formed active new mounds within 8 wk. Injections of conidial powder formulations of Bb447 in late fall and early summer resulted in peak infections of 60 and 52% of live ants sampled, respectively. All of the injected mounds remained active or formed active new mounds within 8 wk after treatment. Injection of Bb447 mixed with a hydrophobic silica carrier resulted in a 52% reduction in active mounds. Injection of the silica carrier alone resulted in a 41% reduction. Foraging by the red imported fire ant was reduced significantly in areas within which mounds were injected with fungal formulations. This is in confirmation with the present study results.

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