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Field assessment of delivery methods for fungal pathogens and Insecticides against cashew stem and root Borer, *Plocaederus ferrugineus* L. (Cerambycidae: Coleoptera)

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ABSTRACT

Field trials were conducted to assess the performance of *Beauveria bassiana* and *Metarhizium anisopliae* against cashew stem and root borer, *Plocaederus ferrugineus* Linn (Coleoptera: Cerambycidae) and to compare them with conventional prophylatic treatment with insecticides. Cashew trees with early phase of borer infestation were marked and the treatments were applied sequentially adopting different delivery methods *viz.*, swabbing of saturated conidial-mud slurry over tree trunk, pouring saturated aqueous suspension of conidia through larval entry holes and soil incorporation of fungal spawn, after removing the grubs and cleaning the frass materials. The effectiveness of treatments was assessed based on the extent of recovery of infested trees. All the treatments were superior to untreated control in reducing the borer infestation. Variations among the treatments could be observed with higher co-efficient of variation with 58.16 per cent. The efficacy of *B. bassiana* and *M. anisopliae* were on a par, but the delivery methods varied significantly. Pouring conidial suspension effected 33.3-36.4% recovery of infested trees followed by swabbing conidial slurry with 23.0-25.0% and soil application with 15.4-16.6% recovered trees. Conventional insecticidal treatments remained superior with 46.2-50.0% recovery of infested trees. However, implementation of fungal application in integrated control of *P. ferrugineus* should be considered, because fungi would not only safer to non-target organisms, but also be more effective in the long term pest control programme.

Key Words: Cashew, *Plocaederus ferrugineus,* fungal entomopathogens, delivery methods

INTRODUCTION

Cashewnut is a major foreign exchange earning plantation crop of India. The productivity of the crop is threatened by insect-pests. Of them, the cashew stem and root borer (CSRB), *Plocaederus ferrugineus* Linn (Coleoptera: Cerambycidae) is the most harmful one in India. The adults are dark brown longicorn beetles, and the gravid female lays eggs in the crevices of loose bark of the trunk and exposed roots. The emerging larvae bore into the living bark and causes significant damage to the vascular system by feeding and tunneling the inner layer of bark, resulting in reduced nutrient uptake, premature leaf senescence, gradual shedding of leaves and death of the tree. The intensity of infestation and extent of damage varies widely across the regions. On an average, this borer kills about 2-5% productive trees every year (Rai, 1984). As much as 6-35% trees of neglected plantations were infested in Kerala, Tamil Nadu and Orissa (Misra and Basu Choudhury, 1985; Senguttuvan, 1999; Mohapatra and Satapathy, 1998). In Guntur and Prakasam Districts of Andhra Pradesh, CSRB infestation went up to 40% in different periods (Arjuna Rao, 1978; Ayyanna and Rama Devi, 1986). Severely attacked trees die within a period of two years causing capital loss to the growers needing to uproot and replace the infested trees. Existing pest management strategies utilizing cultural, mechanical and chemical control methods have met with limited success mainly due to the cryptic life-cycle of the borer inside the trunk and roots (Samiayyan *et al.,* 1991; Punnaiah and Deva Prasad, 1995; Senguttuvan and Mahadevan, 1997; Mohapatra and Satapathy, 1998; Ambethgar, 2002). Cultural and mechanical control tend to be labour intensive, while the use of pesticides is often met with control failure which makes it necessary for repeated application. The variable response of existing control measures has prompted investigation for alternative targeting strategies. Use of fungal entomopathogens as an alternative strategy seemed to be a potential measure particularly for the concealed pests like stem borers owing to the prevalence of congenial environment within the larval tunnels for development of mycoses. The biostages of *P. ferrugineus* in dead cashew trees were found often naturally infected by *Metarhizium anisopliae* (Metsch.) Sorok., and *Beauveria bassiana* (Bals.) Vuill., at enzootic levels (Bhat and Raviprasad, 1996; Ambethgar *et al.*, 1999). Diverse isolates of *M. anisopliae* and *B. bassiana* were reported to be highly pathogenic to *P. ferrugineus* under laboratory bioassay studies (Ambethgar, 2002), but their potential following artificial introductions in fields was seldom explored due to inadequate application technology. In the present study, the efficacy of *M. anisopliae* and *B. bassiana* against *P. ferrugineus* was assessed under field conditions using different delivery methods and were compared with the recommended chemical insecticides.

MATERIALS AND METHODS

Source of Fungal pathogens

The fungal pathogens, *M. anisopliae* and *B. bassiana* were initially isolated from naturally infected cadavers of *P. ferrugineus* grubs and selected based on laboratory bioassay adopting two-way screening *viz.*, using an initial single-dose assay with a standard concentration of $1x10^8$ conidia/ml in 0.02% Tween 80[â], followed by a multiple-dose mortality assay with six different conidial concentrations *viz.*, $1x10^4$, 10^5, 10^6, 10^7, 10^8 and 10^9 conidia/ml in 0.02% Tween 80[â] as surfactant (Ambethgar, 2002).

Production and Preparation of Inoculum

The fungal isolates were initially passed through the larvae of *P. ferrugineus* and the type-isolates were re-isolated on Sabouraud Dextrose Agar medium with 0.25% w/v yeast extract (SDAY). After re-isolation, the fungi were inoculated separately in autoclavable polythene bags each containing 250g of 24h presoaked sorghum grains, and autoclaved at 15 lbs/cm² pressure for 30 minutes. The fungus inoculated grains were incubated at 28°C ± 2°C until fluffy sporulation. The conidia were harvested from 2-3 week old sporulated spawn by crushing and mixing in sterile distilled water containing 0.02 per cent Tween 80[â] as surfactant to preserve the efficacy of fungi. Viability of the conidia was determined by direct examination at 200x with a phase contrast microscope, prior to use.

Field Trial

Field trials were conducted during 2004-2008 on the efficacy of *M. anisopliae* and *B. bassiana* using three different delivery methods in controlling *P. ferrugineus* infestation and compared with the standard check monocrotophos 0.02% and neem oil 5% swab as an environment friendly component on a ten years old clonal planting of VRI 2 cashew at the Regional Research Station, Vridhachalam (11.3°N of latitude, 79.26°E of longitude and 42.67m MSL), Tamil Nadu. The trial was conducted in Completely Randomized Block Design using nine treatments *viz.*, T_1 Swabbing the trunk with saturated conidia of *M. anisopliae* in mud slurry; T_2 Swabbing the trunk with saturated conidia of *B. bassiana* in mud slurry; T_3 Pouring saturated conidial suspension of *M. anisopliae* through borer's entry holes; T_4 Pouring saturated conidial suspension of *B. bassiana* through borer's entry holes; T_5 Soil application of *M. anisopliae* spawn 250g + FYM 50Kg/tree under the canopy; T_6 Soil application of *B. bassiana* spawn 250g + FYM 50Kg/tree tree under the canopy; T_7 Spraying the trunk with Monocrotophos 0.2%; T_8 Spraying the trunk with Crude Neem oil 5% and T_9 Control (only extraction of grubs).

Early phase borer infested cashew trees with symptoms of gummosis and extrusion of frass were marked. Before imposition of the treatments, the grubs residing in the trees were removed mechanically. The expelled remains of frass and resin in the damaged portions of bark were cleaned. The treatments were applied to the dewormed trees adopting the following delivery methods described: (1) *Swabbing:* Two litres of mud slurry containing saturated conidia of respective fungus were swabbed thoroughly on the exposed roots and trunk from collar to one meter height after brushing the scaly bark to dislodge the eggs, (2) *Pouring conidial suspension:* Two litres of saturated aqueous spore suspension of respective fungus containing 0.02% Tween 80® was poured through borer's entry holes all-around the collar region of trunk until the point of saturation and (3) *Soil application:* Crushed spawn (250g) containing conidia and mycelia was mixed with 50kg of well ripened / decomposed farm yard manure and incorporated in to soil up to 20cm deep and 1.0m radius from the tree base under the canopy area.

The treatments were imposed one-by-one sequentially as and when infested trees are available (AICRP, 2004). The sequential treatments refer to T-1 on the first infested tree; T-2 on the second infested tree; T-3 on the third infested tree, like wise up to T-9 on the ninth infested tree. Again T-1 on the tenth infested tree, T-2 on the eleventh infested tree and so on. In this way, 11-13 trees were used per treatment with a total of 110 infested trees received different treatments throughout the period of study as shown in Table 1.

Observation recorded

Observations on active holes of stem borer were recorded one day prior to treatment and post observations were made three months after imposing the treatments. Stoppage of frass extrusion/gum exudation from borer's entry hole after treatment was treated as dead holes, and holes with fresh frass and continuous gum exudation was treated as active holes. The effectiveness of treatments was assessed based on the extent of recovery of infested trees out of total number of trees treated per treatment over the entire period of the study. The recovery per cent for each treatment was worked out using the following formula:

$$\text{Recovery Per Cent} = \frac{\text{Number of trees recovered}}{\text{Total number of trees treated}} \times 100$$

Mean and coefficient of variations were worked out, and the data were analyzed statistically following Gomez and Gomez (1984).

RESULTS AND DISCUSSION

The data on the efficacy of *M. anisopliae* and *B. bassiana* against *P. ferrugineus* are presented in Table 1 and Fig.1. All the treatments were superior to untreated control in reducing the borer infestation at varying levels. Variations among the treatments was evident as indicated by higher co-efficient of variation (58.16 %), exhibiting superiority to the untreated control.

Eventhough the efficacy of *M. anisopliae* and *B. bassiana* was on a par with each other, the performance of delivery methods varied. Among three delivery methods, direct pouring of saturated suspension of conidia through

Table 1. Effect of fungal pathogens and insecticides on *Plocaederus ferrugineus* under field condition (2004-2008)

Treatments	No. of trees	2004	2005	2006	2007	2008	Total no. of trees treated	Total no. of trees recoverd	Recovery (%)
T-1 Swabbing with *Metarhizium anisopliae* in mud slurry	Treated Recovered	3 0	3 1	2 1	2 0	2 1	12	3	25.0
T-2 Swabbing with *Beauveria bassiana* in mud slurry	Treated Recovered	3 0	3 1	3 1	2 1	2 0	13	3	23.0
T-3 Pouring *M. anisopliae* saturated conidial suspension through borer's entry holes	Treated Recovered	3 0	2 1	2 1	2 1	2 1	11	4	36.4
T-4 Pouring *B. bassiana* saturated conidial suspension through borer's entry holes	Treated Recovered	3 0	3 1	2 1	2 1	2 1	12	4	33.3
T-5 Soil application of *M. anisopliae* spawn 250g + FYM 50 Kg/tree	Treated Recovered	3 0	3 1	2 0	2 0	2 1	12	2	16.6
T-6 Soil application of *B. bassiana* spawn 250g + FYM 50 Kg/tree	Treated Recovered	3 0	3 0	3 1	2 1	2 0	13	2	15.3
T-7 Spraying with Monocrotophos 0.2%	Treated Recovered	3 1	3 2	2 1	2 1	2 1	12	6	50.0
T-8 Spraying with Crude Neem oil 5%	Treated Recovered	3 1	3 2	3 1	2 1	2 1	13	6	46.2
T-9 Control (only removal of grubs)	Treated Recovered	3 0	3 0	2 0	2 0	2 0	12	0	00.0
Total	-	-	-	-	-	-	110	30	-
Mean	-	-	-	-	-	-	-	-	27.31
S.Ed	-	-	-	-	-	-	-	-	05.29
C. V (%)	-	-	-	-	-	-	-	-	58.16

Note: Treatments 1-8 were also imposed after mechanical removal of grubs and cleaning the decayed bark portions borer's entry holes registered maximum of 36.4 and 33.3% recovery of infested trees for *M. anisopliae* and *B. bassiana* respectively, which were on a par with each other; but significantly superior to rest of the two delivery methods *viz.*, swabbing the tree trunk with mud slurry containing saturated concentration of conidia, (with 25.0 and 23.0 per cent recovery) and soil application of crushed fungal spawn under tree canopy, with 16.6 and 15.4 per cent recovery of trees for *M. anisopliae* and *B. bassiana* respectively. None of the untreated trees recovered (Fig. 1). The result of the present study agrees with the findings of the recent field studies. Pouring spore suspension of *M. anisopliae* and *B. bassiana* through bored holes was effective compared to swabbing and soil application of spores (Saminathan *et al.*, 2004). Soil application of *M. anisopliae* and *B. bassiana* spawn @250g in combination with 500 g neem cake during October-November minimized the CSRB infestation to 7.40 and 11.10% respectively as against 20.35% infestation in the untreated control (Sahu and Sharma, 2008). However, swabbing conidial suspension of *M. anisopliae* as prophylactic measure was not satisfactory and resulted less effect against the borer (Mohapatra and Jena, 2008). In the present study, phytosanitation by mechanical removal of grubs followed by pouring infested portions with fungal inoculum have improved the efficiency of *M. anisopliae* and *B. bassiana* by realizing 33.3-46.4% recovery of infested trees. The overall recovery per cent in each of the treatment confirmed the performance of the delivery methods in the following descending order: pouring through infestation holes > swabbing the trunk and exposed roots > soil application under the tree canopy.

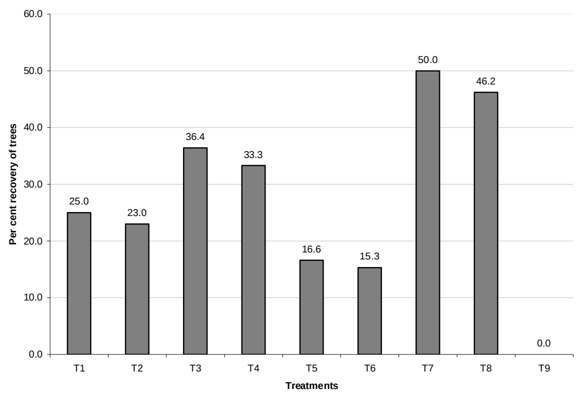

Fig.1 Effect of fungal pathogens on *P. ferrugineus* under field condition

T_1: Swabbing the trunk with saturated conidia of *Metarhizium anisopliae,* T_2: Swabbing the trunk with saturated conidia of *Beauveria bassiana,* T_3: Pouring saturated *Metarhizium* suspension through bore holes, T_4: Pouring saturated *Beauveria* suspension through bore holes, T_6: Soil application of *Beauveria* spawn 250 g + FYM 50Kg/tree, T_7: Spraying the trunk with Monocrotophos 0.2 % , T_8: Spraying the trunk with Crude Neem oil 5 %, T_9: Control (only extraction of grubs), T_5: Soil application of *Metarhizium* spawn 250g + FYM 50 Kg/tree. The conventional chemical treatments remained superior to the fungal pathogens with different delivery methods (Table 1). Post-prophylatic treatment with monocrotophos

@0.2% registered maximum of 50% recovery of infested trees followed by crude neem oil @5% with (46.2% recovery) as against nil recovery of trees in the control, where only mechanical removal of grubs was maintained. The complete failure met with mechanical removal of grubs from the infested trees was owing to reinfestation resulting from volatile chemicals, particularly of kairomones released from the wounded portions of trees, which might have served as promoter factor to attract gravid beetles for egg laying and subsequent re-infestation (Chakraborti, 2006). It is seemed that the insecticides (monocrotophos and neem oil) performed as maskers for the tree volatiles and thereby prevented reinfestation by 50 per cent; but mortality of remaining 50% of treated trees might be due to progression of larval tunneling resulting of non-reaching of toxicants to the target sites where the residual larval population resided inside the trees (or) the trees might have in the state of beyond recovery phase when the treatment was imposed. Prior reports indicated that infested trees in the early phase of borer's attack respond to insecticidal treatments, but succumb in the advanced phase of attack (Samiayyan *et al.*, 1991; Senguttuvan and Mahadevan, 1997; Mohapatra *et al.*, 2008).

Despite more convincing results favouring the chemical insecticides were arrived in the present study, absolute control of the pest could not be achieved with neither conventional insecticides or nor fungal entomopathogens. Compared to insecticides entomofungi were least effective; but still, implementation of fungal application in the integrated control programme of *P. ferrugineus* should be considered, since there could be great logistic advantages; repeated introduction of fungi could kill as many larvae as possible over the years that would reduce the pest incidence in that locality in long run. Mixing the fungal spawn with organic carriers like farm yard manure, vermicompost and cashew apple chunk increase the spore load and improve the persistence for at least three months under field conditions. If these entomofungi are made to be naturalized in cashew plantings, they might represent a practical and safe method of suppressing the pest complex, because fungi would not only safer to non-target organisms, but also be more effective in the long term by displaying self-replicative mechanisms in the cropped ecosystems.

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REFERENCES

AICRP (All India Coordinated Research Project) on Cashew. 2004. Proceedings of National Group Meeting of Scientists of AICRP on Cashew, 4-6 June 2004. National Research Centre for Cashew, Puttur, Karnataka-574 202, India.

Ambethgar, V. Lakshmanan, V., Dinakaran, D. and Selvarajan, M. 1999. Mycosis of cashew stem and root borer, *Plocaederus ferrugineus* L. (Coleoptera: Cerambyciade) by *Metarhizium anisopliae* (Metsch.) Sorokin (Deuteromycotina: Moniliales) from Tamil Nadu (India), *Journal of Entomological Research,* **23**(1): 81-83.

Ambethgar, V. 2002. Screening and selection of entomopathogenic fungi for management of cashew stem and root borer, *Plocaederus ferrugineus* L. (Cerambycidae: Coleoptera). *Pest Management in Horticultural Ecosystems,* **9**(1): 19-31.

Arjuna Rao, P. 1978. Pest complex of cashew (*Anacardium occidentale*) in Guntur and Prakasam Districts of Andhra Pradesh. *Cashew Bulletin*, **15**(1): 7-11.

Ayyanna, T. and Rama Devi. 1986.A study of the distribution and status of stem and root borer, *Plocaederus ferrugineus* dreadful pest on cashew in the Coastal districts of Andhra Pradesh and its control. *Cashew Causerie,* **8** (1): 6-8.

Bhat, P. S. and Raviprasad, T. N. 1996. Pathogenicity of entomopathogenic fungi against cashew stem borer, *Plocaederus ferrugineus* Linn. (Coleoptera: Cerambycidae). *Journal of Plantation Crops,* **24**: 265-271.

Chakraborti, S. 2006. Search for maskers of damaged tree volatiles and adult management of cashew stem and root borer, *Journal of Applied Zoological Research,* **17** (1): 72-76.

Gomez, K. A. and Gomes, A. A. 1984. Statistical Procedures for Agricultural Research, John Wily and Sons, New York.

Misra, M. P. and Basu Choudhury, J. C. 1985. Control of *Plocaederus ferrugineus* L. (Coleoptera: Cerambycidae) through field hygiene. *Indian Journal of Agricultural Science,* **55**: 290-293.

Mohapatra, L. N. and Satapathy, C. R. 1998. Effect of prophylactic measures in management of cashew stem and root borer, *Plocaederus ferrugineus*. *Indian Journal of Entomology,* **60** (3): 257-261.

Mohapatra, R. N. and Jena, B. C. 2008. Effect of prophylactic measures in management of cashew stem and root borer (*Plocaederus ferrugineus*). *Journal of Plantation Crops,* **36** (2): 140-141.

Mohapatra, R. N., Jena, B. C. and Mohapatra, L. N. 2008. Population dynamics, survey and behaviour of cashew stem and root borer *Plocaederus ferrugineus* in Orissa. *Journal of Plantation Crops,* **36** (2): 145-148.

Punnaiah, K. C. and Devaprasad, V. 1995. Management of cashew stem and root borer, *Plocaederus ferrugineus* L. *Cashew,* **9** (3): 17-23.

Rai, P. S. 1984. Handbook on Cashew Pests. Research Co Publications. 199. East Azad Nagar, Delhi, 103 **PP**.

Samiayyan, K, Palanisamy, K. P., Ahmed Shah, H. and Manivannan, K. 1991. Effect of prophylactic measures against cashew stem and root borer *Plocaederus ferrugineus*: *Cashew,* **5**(1): 16-17.

Saminathan, V. R., Mahalingam, C. A., Ambethgar, V., Vanitha, J., Ananthanayaki, D. and Prabaharan, G. 2004. Efeect of fungal pathogens on cashew stem and root borer, *Plocaederus ferrugineus* L. (Coleoptera: Cerambycidae) under field condition. *Pest Management in Horticultural Ecosystems,* **10** (1): 61-65.

Senguttuvan, T. 1999. Distribution and intensity of infestation by cashew stem and root borer, *Plocaederus ferrugineus* L. in Tamil Nadu. *Pest Management in Horticultural Ecosystems,* **5** (2): 154-156.

Senguttuvan, T. and Mahadevan, N. R. 1997. Integrated Management of Cashew stem and root borer *Plocaederus ferrugineus* L. *Pest Management in Horticultural Ecosystems,* **3** (2): 79-84.

Sahu, K. R. and Sharma, D. 2008. Management of cashew stem and root borer, *Plocaederus ferrugineus* L. by microbial and plant products. *Journal of Biopesticides,* 1 (2): 121-123.

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