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SCREENING OF FUNGAL ENTOMOPATHOGENS AGAINST BANANA APHID *PENTALONIA NIGRONERVOSA* COQ

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ABSTRACT

Entomopathogenic fungal pathogens such as *Lecanicilium fusisporum*, *Metarhizium anisopliae* and *Beauveria bassiana* were screened against the banana aphid, *Pentalonia nigronervosa* under in vitro conditions. A spore concentration of 1x10^7 CFU/ ml., aphid mortality of 54, 29 and 42% was recorded 3 days after inoculation in *L. fusisporum, M. anisopliae* and *B. bassiana* respectively. Further an increased mortality of 96.38%, 77.40% and 94.0% were recorded 7 days after application in *L. fusisporum, M. anisopliae* and *B. bassiana,* respectively. Results indicated that *L. fusisporum* has the highest efficacy in controlling the pest.

Key words: Banana, aphid, pest, BBTV, *Pentalonia nigronervosa,* in vitro screening, bioefficacy, entomopathogenic fungus, *Lecanicilium fusisporum, Metarhizium anisopliae, Beauveria bassiana*

Banana aphid (*Pentalonia nigronervosa*), banana rhizome weevil, *Cosmopolites sordidus* (Germ.) and banana stem weevil (*Odoiporus longicollis* (Oliv.)) are considered as the main pest of banana (Shanker et al., 2022). Among these pests, aphid transmits a few plant viruses. The secondary infection of Banana bunch top disease is caused by the insect vector, *P. nigronervosa* (Chakraborty et al., 2021). Biological control is considered as an alternative insect pest control method that utilizes wide range of predators, parasitoids and pathogens to control pest attack in plants (Stenberg et al., 2021). *Beauveria bassiana* and *Lecanicilium longisporum*, are examples of entomopathogenic fungi that have been effectively produced as biological control agents against a variety of pests (De Faria and Wraight, 2007; Shah and Pell, 2003; Powell and Pell, 2007). *Verticillium lecanii* has been employed as a biological control agent against aphids (Hall, 1981). Kakati et al. (2018) reported that the three entomopathogens *B. bassiana, M. anisopliae* and *V. lecanii* are compatible with the common insecticide used against banana aphid such as dimethoate, azadiracchtin and imidacloprid. The present study was carried out to study the efficacy of these entomopathogens against banana aphid, *P. nigronervosa.*

MATERIALS AND METHODS

The experiment was conducted at the Department of Zoology, Jamal Mohamed College (Autonomous), Tiruchirappallli, Tamil Nadu during December 2021-February 2022. The aphids required for this study were maintained in banana plants cv. Karpuravalli grown in earthen pots. The aphid cultures were maintained on in a room with ambient temperature of (24± 2ºC) for mass multiplication. From the colony required aphids were collected using a fine Camlin hair brush. The soil samples (0.5kg) from various fields were collected at a depth of 15 cm. The entomopathogens were isolated by insect baiting method (Bedding and Akhurst, 1975; Chandler et al., 1997). To isolate the entomopathogens, 50 gm of the collected soil was filled in separate plastic container (5 cm x 4 cm) and released five 4th instar larvae of *Corcyra cephalonica* (rice moth) in to the container with perforated lid. Larval mortality was observed at 24 hr interval. The dead larvae were placed on a watch glass covered with wet Whatman filter paper and the watch glass kept inside a petri plate. The pure cultures of fungi were stored in a refrigerator at -4°C until further use on Potato dextrose agar slants. The additional multiplication (25°C) was carried out at 70± 10% RH. The individual isolates were inoculated into potato dextrose broth and the flasks were incubated in a humidity chamber for 10-14 days to obtain sporulation stage of the fungus. Then the mat was harvested and blended in a blender using sterile distilled water and the spore suspension was filtered through double layer of sterile cheese cloth. The spores were transferred to screw cap test tube. The spore suspension was prepared using serial dilution method for all the three different isolates of entomopathogens.

The fungi isolated were evaluated against banana aphid (*P. nigronervosa*) by detached leaf bioassay technique (Yokomi and Gottwald, 1988). The banana mid rib was washed in sterile distilled water, surface sterilized for five minutes with 0.05% sodium hypochlorite, and then dried by air. To get rid of mycelial mats, the spore mixture was filtered through many layers of muslin cloth. By diluting spore suspensions for the bioassay with 0.005% Triton X-100, the final concentration of fungus was adjusted to $1x10^7$ CFU/ml (Vu et al., 2007) (*B. bassiana, M. anisopliae* and *L. fusisporum*). Prior to inoculation, individual banana midribs with a comparable density of insects (30 insects/cm^2) were released. Aphid-infested leaf was submerged in the suspension of spores for ten seconds. Controls were submerged in Triton X-100 0.005%. By counting the number of diseased and controlled insects per mid rib, the daily mortality of aphids were calculated. Four replications were used for each isolate. Imidacloprid was used as a standard check and leaf sheath wiped with 0.01% Tween 80 which was used as control. The dead aphids were collected and kept inside a Petri plate containing moisture filter paper for recording the development of fungal growth on the aphid. Larval mortality data was recorded at 24 hr interval until either their demise or the adult's emergence. Using Abbott's formula, the mortality was corrected. To determine the degree of deviation among all the treatments, analysis of the various variables were done. Data on the percentage of aphid deaths were subjected to two-way ANOVA with Tukey's pairwise comparison analysis using ICAR Software-WASP 2.0.

RESULTS AND DISCUSSION

The trapping of soil samples indicated the presence of entomopathogens. *L. fusisporum, Metarhizium anisopliae* and *Beauveria bassiana* were evaluated for the efficacy against the banana aphid, *P. nigronervosa.* The aphid mortality due to the three entomopathogens indicated a gradual increase in the mortality over different days after imposing the treatment. The mortality reached >90% on 6^{th} and 7^{th} days after the treatment in *L. fusisporum* and *B. bassiana* and 100% imidacloprid. A maximum mortality of 96.38 and 94.0, 100.0 and 10.0% mortality was recorded with *L. fusisporum*, *B. bassiana, M. anisopliae*, imidacloprid and control respectively (Table 1). The pathogenicity of fungus over insects has been reported to cause insect population death (Hesketh et al., 2008) and hence, the development of biological management for a number of pests, including aphid (De Faria and Wraight, 2007). Another study using *B. brongniartii* and *M. anisopliae* showed the significance to use as potential biocontrol agents against banana fruit scarring beetle *Basilepta subcostata* (Viswakethu et al., 2021). The distributions of aphid pesticide susceptibility differ depending on whether the bioassays were conducted in a lab or a glasshouse (Fournier and Brodeur, 2000). Previous study has reported the management of *P. nigronervosa* infesting banana under controlled field conditions (Amsavalli et al., 2022). Another study reported the compatibility of entomopathogenous fungi with commonly used insecticides against the *P. nigronervosa* (Kakati et al., 2018). Askary et al. (1998) reported that the *V. lecanii* has a detrimental effect on aphid fertility. The morphological state and developmental stage have an impact on an insect's susceptibility to a fungus infection. Hall (1981) reported that the spore germination, growth, and sporulation rates of *M. anisopleae* were slower than that of *L. fusisporumi*,

Table 1. *Invitro* evaluation of selected entomopathogenic fungi against *P. nigronervosa*

Treatments	Mortality (in days)						
	I day	II day	III day	IV day	V day	VI day	VII day
Lecanicillium fusisporum	17.02± 9.4^{b}	35.96± 15.4^{b}	54.04± 13.9^{b}	65.56± 6.8^{b}	81.54± 13.4^{b}	92.46± 4.5 (91.62)^{b}	96.38 ± 3.7 (95.97)^{ab}
Metarhizium anisoplea	11.94± 5.7^{bc}	19.26± 4.4^{c}	29.26± 3.7^{c}	43.88± 6.1^{c}	62.90± 8.7^{c}	70.68± 4.3 (67.42)^{c}	77.40 ± 3.6 (74.88)^{c}
Beauveria bassiana	15.68± 8.1^{b}	34.82± 7.4^{b}	42.82± 20.3^{bc}	69.74± 11.9^{b}	76.22± 14.4^{b}	92.82± 7.3 (92.02)^{b}	94.00± 6.4 (93.33)^{b}
Imidacloprid 17.8% SL	71.34± 14.1^{a}	91.32± 9.3^{a}	100.00± 0.0^{a}	100.00± 0.0^{a}	100.00± 0.0^{a}	100.00± 0.0 (100.00)^{a}	100.00± 0.0 (100.00)^{a}
Control	2.66± 2.8^{c}	2.66± 2.8^{d}	4.00± 2.8^{d}	5.34± 3.8^{d}	6.00± 4.4^{d}	10.00± 2.3^{d}	10.00± 2.3^{d}
Level of significance CD (5%)	9.337	10.828	15.578	7.843	11.474	4.953	3.782

and this fungus spread more slowly throughout the population of aphids. Further studies will be conducted under field conditions to control the banana aphid*, P. nigronervosa* to prevent the spread of BBTV.

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AUTHOR CONTRIBUTION STATEMENT

KKA and BP conceived and designed research. KKA conducted experiments. KKA and IJ analyzed data. KKA wrote the manuscript. All authors read and approved the manuscript. No ethical issue related to animal is involved in the experimentation.

CONFLICT OF INTEREST

No conflict of interest.

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