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RESEARCH ARTICLE



Pathogenicity of entomopathogenic fungus *Isaria fumosorosea* on rugose spiralling whitefly *Aleurodicus rugioperculatus* and its effect on parasitoid *Encarsia guadeloupae*

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ABSTRACT

Highly polyphagous rugose spiralling whitefly (RSW), *Aleurodicus rugioperculatus* Martin (Hemiptera: Aleyrodidae) invaded India during 2016. Pathogenicity of two strains (ICAR-NBAIR Pfu-1 & Pfu-5) of entomopathogenic fungus *Isaria fumosorosea* Wize were evaluated against RSW, however, only Pfu-5 showed virulence. Further dose mortality studies were carried out with re-isolated Pfu-5 strain with five different concentrations ranging from 1×10^4 – 1×10^8 spores/mL on *A. rugioperculatus* and its dominant parasitoid *Encarsia guadeloupae* (Hymenoptera: Aphelinidae). The Pfu-5 strain showed mean mortality of 44.03, 44.80, 36.42 and 28.82% on the eggs, first, third and fourth instar nymphs, respectively. The LC_{50} values of *I. fumosorosea* for eggs, first, third and fourth instar nymphs were 2.6×10^4 , 2.3×10^4 , 3.5×10^5 and 9.5×10^5 spores/mL, respectively, and the LT_{50} values were 50.39, 41.39, 67.54 and 68.31 h, respectively, for corresponding life stages for the concentration of 1×10^8 spores/mL. The impacts of *I. fumosorosea* on the parasitoid showed the inhibition of adult emergence was much lower in the Pfu-5 strain (5.8–19.6%) at a concentration of 1×10^8 spores/mL at different exposure times. The present study indicated the pathogenicity of Pfu-5 against *A. rugioperculatus* and warrant further studies on the compatibility of these biocontrol agents to be evaluated for the simultaneous utilisation under field conditions.

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Invasive; rugose spiralling whitefly; pathogenicity; *Isaria fumosorosea*; lethal concentration; entomopathogen

1. Introduction

Rugose spiralling whitefly (RSW), *Aleurodicus rugioperculatus* Martin (Hemiptera: Aleyrodidae) is highly polyphagous and invasive pest, recorded for the first time in India during 2016 in Tamil Nadu on coconut (Selvaraj et al., 2016; Sundararaj & Selvaraj, 2017). Subsequently, it was reported from different parts of India viz., Kerala, Karnataka, Andhra Pradesh, Assam, Goa, West Bengal, Maharashtra, Telangana, Meghalaya and Gujarat on coconut, banana, sapota, maize, oil palm, mango, cashew and many other ornamental plants (Selvaraj et al., 2019). *A. rugioperculatus* nymphs and adults not only feed

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aggressively on leaf sap resulting in depletion of nutrients and water which leads to premature leaf drop and drying but also produce wax and excretes sticky honeydew on infested areas leading to the extreme growth of black sooty mould which results in the reduction of photosynthetic efficiency in palms (Kumar et al., 2013). It has now become a regular pest of coconut and oil palm in India, warranting control measures to avoid crop losses.

Two parasitoids, *Encarsia guadeloupae* and *E. dispersa* (Hymenoptera: Aphelinidae) have been fortuitously introduced into India along with spiralling whitefly *Aleurodicus dispersus* during 1990s (Ramani et al., 2002). Among these parasitoids, *E. guadeloupae* was found to be most abundant and potential with natural parasitisation to the extent of 60–82% on *A. rugioperculatus* on coconut and other crops. In India, *E. guadeloupae* was well established by augmentation, re-distribution and various conservation strategies (Selvaraj et al., 2018). Using a single biocontrol agent to suppress the whitefly population may be difficult under severe outbreak conditions especially for an invasive pest like RSW. Many workers have investigated the combined use of insect parasitoid and entomopathogenic fungi as potential biocontrol strategy for the many insects and reported either compatible or non-significant impacts on parasitoids (Avery et al., 2008; Huang et al., 2010; Labbé et al., 2009; Ou et al., 2019).

The entomopathogenic fungus, *Isaria fumosorosea* Wize (formerly *Paecilomyces fumosoroseus*) is distributed worldwide and effective against insect pests mainly whiteflies (Luangsa-Ard et al., 2005) and can initiate epizootics under natural field conditions. This fungus is used as potential biocontrol agent against *A. rugioperculatus* and Bondar's nesting whitefly *Paraleyrodes bondari* on coconut (Ali et al., 2015; Kumar et al., 2016, 2018), Ficus whitefly, *Singhiella simplex* on Ficus (Avery et al., 2019) in Florida. In the preliminary studies, two strains of *I. fumosorosea* (ICAR-NBAIR Pfu-1) and (ICAR-NBAIR Pfu-5) were tested on *A. rugioperculatus* life stages at the concentration of 1×10^8 spores/mL. Based on the tested strains, ICAR-NBAIR Pfu-5 has shown fungal infection and high virulence against eggs, nymphs and adults of *A. rugioperculatus* which was re-isolated and used for present studies.

To improve the effectiveness of biological control of *A. rugioperculatus*, it is need to know if a combination of *I. fumosorosea* and *E. guadeloupae* is more effective than their individual applications. This strategy may provide farmers with an alternative option for the management of *A. rugioperculatus*. The present study was taken up to evaluate the efficacy of native isolate of *I. fumosorosea* (ICAR-NBAIR Pfu-5) and assess the effect on parasitoid, *E. guadeloupae* for management of *A. rugioperculatus* in the coconut.

2. Materials and methods

2.1. Insect culture

Aleurodicus rugioperculatus adults were collected from infested coconut fields from Bengaluru (13.0973° N, 77.3856° E), Karnataka, India. *A. rugioperculatus* culture is reared and maintained on coconut seedlings raised in potted condition in the greenhouse with a natural photoperiod at ICAR-NBAIR, Bengaluru, India. Different development stages such as eggs, first instars (crawlers), third instar and fourth instar nymphs (pupae) were used for bioassays. Similarly, *Encarsia guadeloupae* culture is maintained on

A. rugioperculatus using *Canna indica* as host plant in the greenhouse with a natural photoperiod at ICAR-NBAIR. The parasitised *A. rugioperculatus* nymphs were collected from parasitoid production facility to evaluate the effect of *I. fumosorosea* on *E. guadeloupae*.

2.2. Fungal culture

Two strains of *I. fumosorosea* (ICAR-NBAIR Pfu-1) and (ICAR-NBAIR Pfu-5) were collected from ICAR-NBAIR fungal repository for screening against *A. rugioperculatus*. ICAR-NBAIR Pfu-1 was isolated from red spider mite, *Tetranychus urticae* in Bengaluru, Karnataka, whereas ICAR-NBAIR Pfu-5 was isolated from soil samples in Banas kantha, Gujarat in India. Initial identification of the fungi was done based on phialid and conidial characters. Further, confirmed through molecular characterisation by sequencing internal transcribed spacer region of the fungal recombinant DNA operon and the sequences were deposited in NCBI GenBank database (accession No. of Pfu-1 is JF718687 & KC147664 for Pfu-5). Fungal culture of (ICAR-NBAIR Pfu-5) was produced on 100 g of sterilised rice grains by inoculating 5 mL of 4 days old shaker culture and incubated at $26 \pm 1^\circ\text{C}$ for 15 days. Spore suspension was prepared by suspending one gram of conidiated rice in sterile distilled water containing 0.01% Tween 80. The suspension was filtrated through three layers of muslin cloth to get hyphal-free spore suspension and the concentration of the spore in the suspension was adjusted to 1×10^8 spores/mL using Neubauer's improved haemocytometer.

2.3. Bioassay on *A. rugioperculatus*

Different spore concentrations (1×10^4 – 1×10^8 spores/mL) were tested against developmental stages of *A. rugioperculatus* using a leaf-dip bioassay method. Each coconut leaf bits with 30–45 eggs, first instars, third- and fourth instar nymphs of *A. rugioperculatus* were collected separately from the greenhouse culture. These leaf bits were dipped gently for 15 s in freshly prepared respective spore concentrations along with 0.01% tween-80 and placed on Petri plate (14×2 cm) over the sterilised wet tissue paper. The experiment replicated five times and whole bioassay repeated three times. Wet cotton was kept on both sides of the leaf bits in order to maintain turgidity. The mortality data were recorded by counting the dead cadavers and nymphs with fungal growth. Observations on mortality were recorded periodically at 24-h intervals up to 7 days and mortality data were subjected to Abbott's formula (Abbott, 1925) for calculating corrected mortality.

2.4. Bioassay of *I. fumosorosea* on *Encarsia guadeloupae* parasitised nymphs

Freshly emerged *E. guadeloupae* were collected from nucleus culture and released 20 parasitoids/plant for the parasitism in caged condition when the *A. rugioperculatus* reached second instar nymphs as its preferred stage for the parasitisation. The parasitoid adults were removed from cage after 3 days of release to get uniform parasitism. Parasitised nymphs were assessed by observing the mycetome dispersal and melanisation inside the translucent nymphs (Figure 1(a,b)). To study the compatibility, 30 leaf bits (8 cm length) with each of 40–45 parasitised nymphs of 8 (Figure 1(c)) & 15 days (Figure 1

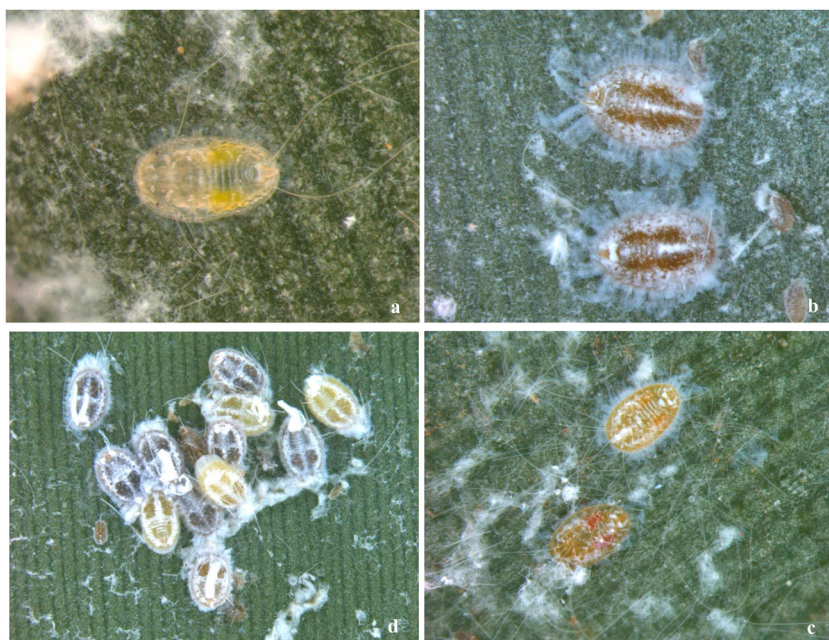


Figure 1. *Aleurodicus rugioperculatus*: Unparasitised nymph (a), parasitised nymphs with melanisation (b), Parasitised nymphs (8 days after parasitoid release) (c) and 15 days after parasitoid release (d).

(d)) after parasitoid release were removed from host plant. 8th and 15th days of parasitised nymphs were made available in separate cages for the same treatment date. These leaf bits were treated with five different concentrations (1×10^4 – 1×10^8 spores/mL) of Pfu-5. The bioassay for each concentration of *I. fumosorosea* was replicated with five times. Observations were recorded on emergence of parasitoid by presence of exit hole (Circular in shape) and mortality of parasitoid (absence of parasitoid movement inside the nymphs) and also parasitoid that did not emerge up to 15 days of treatment was assumed to have died.

2.5. Statistical analysis

Mortality/hatching inhibition of eggs, first instars, third and fourth instar nymphs was statistically analysed. The dose and time dependent mortality studies to kill 50% of the population (LC_{50} and LT_{50}) were determined by probit analysis (Finney, 1971). The dose mortality studies (LC_{50}) were conducted using five spore concentrations (1×10^4 , 1×10^5 , 1×10^6 , 1×10^7 and 1×10^8 spores/mL) against *A. rugioperculatus* life stages. For time mortality studies, the higher concentration @ 1×10^8 spores/mL and the lower concentration 1×10^4 spores/mL were used to determine LT_{50} of Pfu-5 against *A. rugioperculatus* life stages. The statistical analysis was done using SPSS windows version 20.0. The arcsine transformation (% data) was used for normalisation of data before conducting analysis of variance (ANOVA) and analysis was undertaken on the transformed data and untransformed means $\pm$ standard error were presented. *I. fumosorosea* concentration, time and their interactive effects on mortality percentage



Figure 2. *Isaria fumosorosea* infection on *Aleurodicus rugioperculatus*. (A) Eggs, (B,C) Third & fourth instar nymphs, (D) Adult.

data were analysed using PROC GLM (SAS version 9.3; S. A. S Institute, 2011). When ANOVA indicated a significant F value ($\alpha < 0.05$) treatment effects further separated using Tukey post-hoc test.

3. Results

The present studies showed that strain Pfu-5 of *I. fumosorosea* able to affect the egg hatching and even it caused mortality in all the four nymphal stages and adults of *A. rugioperculatus* (Figure 2). Though *I. fumosorosea* not treated on adult, mortality/mycosis and deformation was observed in the adult emerged from treated nymphs.

3.1. Ovicidal effect of *I. fumosorosea* on *A. rugioperculatus*

The overall egg mortality induced by *I. fumosorosea* ranged from 2.63 to 31.78% at two days after treatment in different concentrations tested (Figure 3(a)). The highest egg mortality was noticed when *I. fumosorosea* treated at a concentration of 1×10^8 spores/mL. The egg mortality increased significantly with the increase in the spore concentrations ($F = 23$; $df = 4, 20$; $P < 0.0001$). At 5 days after treatment, the highest egg mortality (99.57%) was observed at 1×10^8 spores/mL and the lowest egg mortality (35.25%) was at 1×10^4 spores/mL ($F = 183.87$; $df = 4, 20$; $P < 0.0001$). The egg mortality of *A. rugioperculatus* was significantly increased with time after inoculation in each treatment ($F = 1217$; $df = 1, 40$; $P < 0.0001$) (Figure 3(a)). A significant interaction was observed between spore concentrations and time ($F = 196.74$; $df = 4, 40$; $P < 0.0001$).

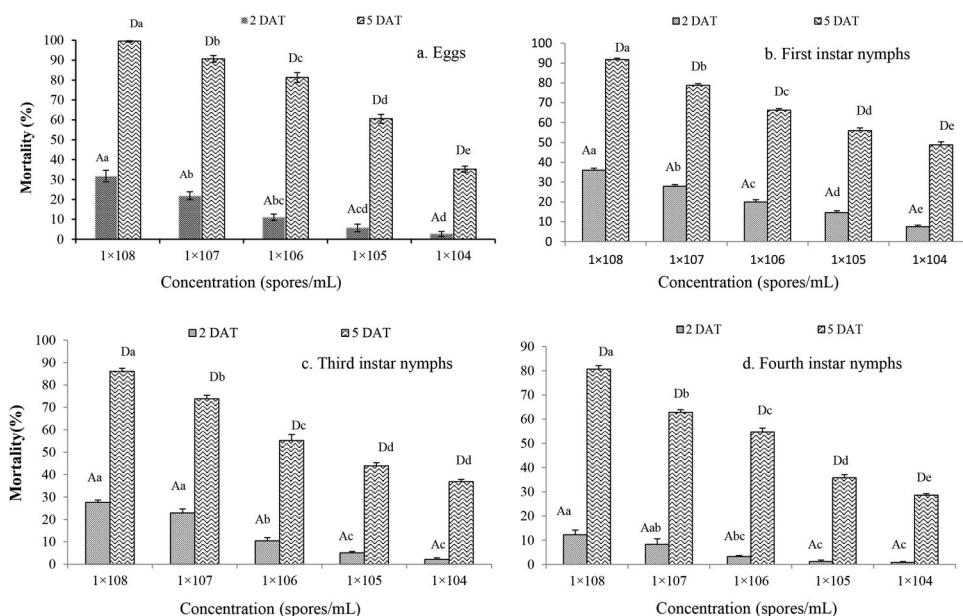


Figure 3. Corrected mortality/inhibition of hatching (% $\pm$ SE) of eggs (A), first instar nymphs (B), second & third instar nymphs (C), pupae (D) of *A. rugioperculatus* treated with *Isaria fumosorosea*. Bars with different uppercase letters on the top of error bars indicate significant differences among the different days after treatment and with different lowercase letters indicate significant differences for different treatments i.e. concentration of *I. fumosorosea* ($P < 0.05$, Tukey's test).

3.2. Pathogenicity of *I. fumosorosea* on early instar nymphs

Isaria fumosorosea caused highest mortality (36.10%) at a concentration of 1×10^8 spores/mL and least mortality (7.58%) at concentration of 1×10^4 spores/mL on first instar nymphs at two days after treatment (Figure 3(b)). The percent mortality was increased significantly with the increase in the spore concentrations ($F = 130.43$; $df = 4, 20$; $P < 0.0001$) (Figure 3(b)). Similarly, first instar nymphal mortality was greatest (91.81%) when treated with spore concentration of 1×10^8 spores/mL and the lowest percent mortality was (48.83%) at spore concentration of 1×10^4 spores/mL at five days after treatment ($F = 274.59$; $df = 4, 20$; $P < 0.0001$). The mortality of first instar nymphs was significantly increased with time after inoculation in each treatment ($F = 4467.17$; $df = 1, 40$; $P < 0.0001$) (Figure 3(b)). A significant interaction was observed between spore concentrations and time ($F = 675.24$; $df = 4, 40$; $P < 0.0001$).

The overall mortality caused by *I. fumosorosea* in third instar nymphs ranged from 27.58% to 2.22% at two days after treatment (Figure 3(c)). As noticed in case of eggs and first instars, similar trend of increased mortality with increased in spore concentration of *I. fumosorosea* was also recorded in third instar nymphs ($F = 64.71$; $df = 4, 20$; $P < 0.0001$). The highest third instar nymphal mortality (86.13%) was recorded at 1×10^8 spores/mL and the lowest mortality (36.91%) was at 1×10^4 spores/mL at 5 days after treatment ($F = 142.84$; $df = 4, 20$; $P < 0.0001$). The percent mortality in third instar nymphs of *A. rugioperculatus* was significantly increased with time after the treatment ($F = 1753.78$;

df = 1, 40; $P < 0.0001$) (Figure 3(c)). A significant interaction was observed between spore concentrations and time ($F = 281.31$; df = 4, 40; $P < 0.0001$).

3.3. Pathogenicity of *I. fumosorosea* on late instar nymphs

At two days after treatment, the percent fourth instar nymphal mortality was 12.24% when treated with at a concentration of 1×10^8 spores/mL and very low mortality (0.80%) were recorded at a concentration of 1×10^4 spores/mL (Figure 3(d)). The pupal mortality increased significantly with the increase in the spore concentrations ($F = 10.49$; df = 4, 20; $P < 0.0001$). The highest pupal mortality was (80.65%) after treating eggs with 1×10^8 spores/mL and lowest mortality was (28.57%) when treated with 1×10^4 spores/mL at 5 days after treatment ($F = 245.68$; df = 4, 20; $P < 0.0001$). The mortality of fourth instar nymphs were significantly increased with time after inoculation in each treatment ($F = 1271.60$; df = 1, 40; $P < 0.0001$) (Figure 3(d)). A significant interaction was observed between spore concentrations and time ($F = 177.90$; df = 4, 40; $P < 0.0001$).

In overall statistics, significant differences in the pathogenicity of *I. fumosorosea* were observed between spore concentration ($F = 527.74$, df = 4, 160, $P < 0.001$), time interval ($F = 5997.24$, df = 1, 60, $P < 0.001$) and stage of host insect ($F = 182.35$, df = 3, 160, $P < 0.001$). Significant interactions were observed between treatment * stage of host insect ($F = 8.0$, df = 12, 160, $P < 0.001$) and treatment * stage of host insect * time ($F = 2.91$, df = 12, 160, $P < 0.001$). Among the developmental stages of *A. rugioperculatus*, irrespective of spore concentration, first instar nymphs were significantly ($P < 0.05$) more susceptible compared to other life stages.

3.4. Determination of the lethal concentration and lethal time

The data on dose- and time mortality response of *I. fumosorosea* against different life stages of *A. rugioperculatus* showed significant difference in LC_{50} and LT_{50} values (Tables 1 and 2). The LC_{50} values of *I. fumosorosea* against the *A. rugioperculatus* eggs, first instar, third instar nymphs and fourth instar nymphs were 2.6×10^4 , 2.3×10^4 , 3.5×10^5 and 9.5×10^5 spores/mL, respectively. The LT_{50} values of *I. fumosorosea* at the highest concentration tested (1×10^8 spores/mL) on eggs, first instar, third instar and fourth instar nymphs were 50.39, 41.39, 67.54 and 68.31 h, respectively and its was 145.21, 119.10, 123.79 and 121.57 h, respectively, at the lowest concentration of 1×10^4 spores/mL.

3.5. Effect of *I. fumosorosea* on *Encarsia guadeloupae* parasitised nymphs

The inhibition rates of adult emergence from parasitised nymphs in the Pfu-5 strain were much lower (5.8%) at highest concentration of 1×10^8 spores/mL when treated them at 15

Table 1. Dose mortality response of *Isaria fumosorosea* against rugose spiralling whitefly.

Developmental life stages of RSW	LC_{50} spores/mL	95% FL	Slope $\pm$ SE	χ^2	P value	DF
Eggs	2.6×10^4	0.6×10^4 – 7.0×10^5	0.56 ± 0.03	5.09	0.11	3
First instar nymphs	2.3×10^4	1.2×10^4 – 3.9×10^4	0.53 ± 0.04	3.80	0.28	3
Third instar nymphs	3.5×10^5	1.7×10^5 – 6.8×10^5	0.33 ± 0.03	0.19	0.97	3
Pupae	9.5×10^5	1.09×10^5 – 7.9×10^6	0.30 ± 0.03	6.04	0.11	3

Note: FL, fiducial limits; SE: standard error; χ^2 – indicates a good fit of the line to the data.

Table 2. Time mortality response of *Isaria fumosorosea* against rugose spiralling whitefly.

Developmental life stages of RSW	LT ₅₀ hours	95% FL	Slope $\pm$ SE	χ^2	P value	DF
Concentration @ 1×10^8 spores/mL						
Eggs	50.39	40.60–59.10	1.16 ± 0.43	3.47	0.06	3
First instar nymphs	41.39	36.75–47.21	2.71 ± 0.36	2.53	0.28	3
Third instar nymphs	67.54	57.02–88.06	3.02 ± 0.46	0.43	0.51	3
Pupae	68.31	65.73–71.43	11.89 ± 1.42	0.04	0.98	3
Concentration @ 1×10^4 spores/mL						
Eggs	145.21	120.24–193.88	2.06 ± 0.24	1.43	0.48	3
First instar nymphs	119.10	99.63–157.23	2.00 ± 0.26	0.72	0.69	3
Third instar nymphs	123.79	108.12–154.90	3.84 ± 0.55	1.80	0.40	3
Pupae	121.57	107.19–152.28	4.72 ± 0.77	3.38	0.18	3

Notes: FL, fiducial limits; SE, standard error, χ^2 indicates a good fit of the line to the data.

days after parasitisation (DAP) (Figure 4). However, the emergences of parasitoid adults from *I. fumosorosea* infected nymphs were affected to the extent of 19.6% when nymphs were treated on or before 8 days of parasitisation. Effect of *I. fumosorosea* on adult emergence showed concentration dependence and adult emergence inhibition rates were increased with increase in time of treatment and spore concentration.

4. Discussion

Isaria fumosorosea is one of the most promising and extensively studied entomopathogenic fungus attacking nymphs and adults of whiteflies such as *Bemisia* spp., *Aleurodicus dispersus* and *Trialeurodes vaporariorum* (Boopathi et al., 2013; Lacey et al., 2008; Sanchez & Castillo, 2008); *A. rugioperculatus* and *P. bondari* on coconut (Ali et al., 2015; Kumar et al., 2016, 2018) and *S. simplex* on ficus (Avery et al., 2019) in Florida. The indigenous fungal isolate (Pfu-5) caused significant egg mortality (99.57%) on *A. rugioperculatus* in the present study. Kumar et al. (2016, 2018) reported significant in suppression of eggs (90.00%) of *A. rugioperculatus* by *I. fumosorosea* treatments than control at different

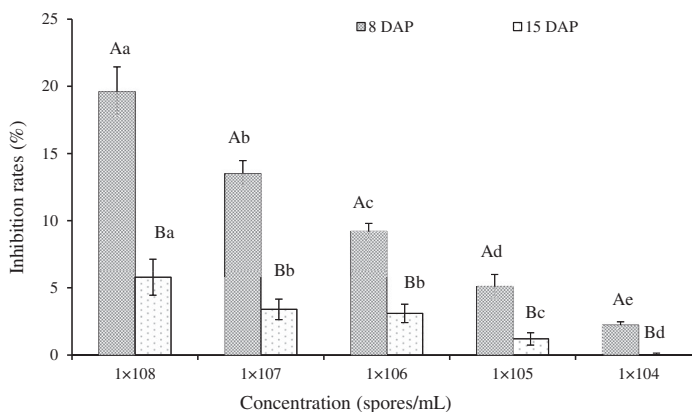


Figure 4. Inhibition rates (% $\pm$ SE) of adult parasitoid, *E. guadeloupeae* emergence when parasitised nymphs (8 & 15 days after release) treated with *I. fumosorosea*. Bars with different uppercase letters on the top of error bars indicate significant differences among the different days after parasitism and with different lowercase letters indicate significant differences for different treatments i.e. concentration of *I. fumosorosea* ($P < 0.05$, Tukey's test).

sampling periods (2 and 8 DAT) in Florida. On the other hand, Sanchez and Castillo (2008) reported only 50% reduction in egg hatching by this entomopathogen on its closely related whitefly species, *Aleurodicus dispersus*. Boopathi et al. (2013) reported that 37.3% and 22.6% of egg mortality with *Metarhizium anisopliae* (M2 Strain) and *P. fumosoroseus* (P1 strain) at 8 DAT. The egg mortality is one of the important attributes of entomopathogenic fungus by which pest is suppressed at initial stage. So that subsequent pest population build up and crop damage is reduced.

Results in the present study indicate the highest cumulative mortality (91.81%) of first instar nymphs at the concentration of 1×10^8 spores/mL at 5 DAT. Similar percent mortalities of the first instar nymphs of *A. rugioperculatus* (41–89%) and *A. dispersus* (97.8–100%) were observed with *I. fumosorosea* in Florida by Kumar et al. (2016, 2018) and in India by Boopathi et al. (2013, 2015). Highest mortality observed on first instar nymphs compared to subsequent nymphal instars may be due to lesser wax coverage on first instar nymphs.

Isaria fumosorosea also showed high pathogenicity (80.65%) on fourth instar nymphs (pupae) which leading to drastic reduction of adult emergence that may result in less perpetuation of the pest in the coconut ecosystem. Besides, high mycosis was also observed in newly emerged adults. The percentage mortality of different stages of *A. rugioperculatus* significantly increased with the increase in the spore concentrations. This indicates that higher concentration of fungal spores resulted in faster control of the targeted pest. Two commercial formulations of *I. fumosorosea* (Preferal® WP & NoFly® WP) on late instar stages of RSW and Bondar's nesting whitefly showed significant reduction (40–80%) in *A. rugioperculatus* nymphal counts (Ali et al., 2015). The strain ICAR-NBAIR Pfu-5 showed relatively lower pathogenicity against pupae compared to eggs, first, third instar nymphs. This low mortality could be due to high wax coverage and harder cuticle on pupae as compared to early instar nymphs.

Several workers reported that the efficacy of *I. fumosorosea* varies with insect species and also different developmental stages of the insect (Ali et al., 2015; Kumar et al., 2016, 2018). However, in the present study, considering the higher susceptibility of the first instar nymphs of *A. rugioperculatus* to *I. fumosorosea* than subsequent instar nymphs, *I. fumosorosea* should be applied against first instar nymphs for effective and enhanced control of *A. rugioperculatus* and also to prevent/or third-instar nymphs becoming more damaging in the field.

In the present study, the lowest LC_{50} was recorded by *I. fumosorosea* as 2.3×10^4 spores/mL for *A. rugioperculatus* early instar nymphs indicating higher virulence as compared to other stages. LC_{50} value obtained in the present study was lower than that of reported by Boopathi et al. (2013) for *A. dispersus* (8.189×10^7 conidia/mL). The difference in the LC_{50} values might be due to the difference in virulence of *I. fumosorosea* isolates and the host species. Moreover, wax coverage on nymphal stages of *A. dispersus* is higher as compared to *A. rugioperculatus*. In the present study, the lowest LT_{50} value was recorded for first instar nymphs while the highest for fourth instar. Thus, indicating the higher virulence of *I. fumosorosea* against first instar nymphs as compared to other life stages. Eslamizadeh et al. (2013) found that LT_{50} at the dose of 1×10^4 conidia/mL was 6.38 days for *Bemisia tabaci* and 5.91 days at 1×10^5 conidia/mL.

The result of present study suggests Pfu-5 as potential biopesticide against *A. rugioperculatus* with very slight negative effects on beneficial parasitoid,

E. guadeloupae. The compatibility of two different types of biological control agents is very important for sustained and successful pest management. To study compatibility, second instar nymphs of *A. rugioperculatus* were selected for the study because females of *E. guadeloupae* preferred to deposit eggs in early rather than in late instars of *A. dispersus*, and oviposition rates were highest in the second instar larvae (Mollot et al., 2015). This compatibility finding is consistent with other researchers that have studied similar parasitoid-pathogen with fungal entomopathogens including *I. fumosorosea* and aleyrodid pests (Avery et al., 2008; Huang et al., 2010; Ou et al., 2019). It is important to assess the interactions between biological control agents in the field to determine what optimal fungal concentration would maximise whitefly mortality and minimise parasitoid infection. Further studies have to be carried out to study on compatibility of the two biocontrol agents and environmental effects on efficacy of each agent alone as well as in combination for management of *A. rugioperculatus* need to be evaluated under field conditions. It is evident from present findings that there are positive interactions between the *I. fumosorosea* and *E. guadeloupae* and these interactions could serve as an effective control strategy for *A. rugioperculatus* in coconut and oil palm ecosystem.

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