Sandeep *et al.*
Egyptian Journal of Biological Pest Control (2022) 32:106
https://doi.org/10.1186/s41938-022-00600-z

Egyptian Journal of
Biological Pest Control

RESEARCH

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Field efficacy of *Isaria fumosorosea* alone and in combination with insecticides against *Aleurodicus rugioperculatus* on coconut

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Abstract

Background: The rugose spiraling whitefly (RSW), *Aleurodicus rugioperculatus* Martin (Hemiptera: Aleyrodidae), an exotic polyphagous pest and its infestation was documented on coconut (*Cocos nucifera* L.) for the first time in India during 2016. RSW has attained a serious pest on many economically important crop plants due to its damage severity and rapid spread across the country in a short span of time. Hence, an attempt was made to evaluate the efficiency of the entomopathogenic fungus (EPF), *Isaria fumosorosea* alone as well as in combination with a reduced dose of few insecticides against RSW on coconut to devise a sustainable integrated pest management module. Thirty-two randomly selected RSW-infested coconut palms were labelled, and eight treatments with four replications were used in a randomized complete block design during the summer months (April and May) during 2021. Two sprays were performed at 15-day intervals, at a rate of 5 L/palm, and observations on infestation/palm (%), intensity (%) and mean live colonies of RSW were recorded.

Results: The results revealed that the combination of *I. fumosorosea* (ICAR-NBAIR Pfu-5) @ 5 ml/l + profenophos 50 EC @ 2 ml/l spray significantly reduced per cent infestation of RSW (82.97%), per cent intensity of RSW (80.49%) and mean RSW live colonies (79.68%) followed by combination of *I. fumosorosea* (ICAR-NBAIR Pfu-5) @ 5 ml/l + buprofezin 25% SC @ 1.25 ml/l spray significantly reduced per cent infestation of RSW (79.35%), intensity of RSW (74.79%) and mean RSW live colonies (74.20%) over *I. fumosorosea* (ICAR-NBAIR Pfu-5) @ 5 ml/l alone and untreated control. A combination of *I. fumosorosea* (5 ml/l) + profenophos 50 EC (2 ml/l) spray twice at an interval of 15 days was found effective in reducing the RSW population on coconut.

Conclusions: The present study concludes that the RSW can be controlled effectively by *I. fumosorosea* alone and in combination with novel insecticides at a reduced rate which showed better toxicity, ovicidal action and preserve natural enemies and reduced environmental load of chemical pesticides.

Keywords: *Aleurodicus rugioperculatus*, Invasive, Rugose spiraling whitefly, Entomopathogenic fungus, Biological control, Novel insecticides

Background

Rugose spiraling whitefly (RSW), *Aleurodicus rugioperculatus* Martin (Hemiptera:Aleyrodidae), was first reported in Miami-Dade Country, Florida, USA, as a pest of gumbo limbo *Bursera simaruba* (L.) in 2009. RSW was first described by Martin (2004) and based on samples collected from coconut palm (*Cocos nucifera* L.), Arecaceae leaves in Belize. Occurrence and infestation of RSW on coconut palms was reported for the first time

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in India at Pollachi, Tamil Nadu, during 2016 (Sundararaj and Selvaraj 2017). RSW is highly polyphagous being found to feed on more than 120 plants, mainly from the families of Arecaceae, Musaceae, Anacardiaceae, Euphorbiaceae, Myrtaceae, Fabaceae and Combretaceae. In India, so far, 40 host plants have been recorded, but the pest is majorly a menace to coconut, oil palm and banana (Sundararaj et al. 2021).

In South Indian states, coconut production has been severely affected after the invasion and establishment of RSW and reached the outbreak situation during favourable weather conditions, which has led to reduction in quality of nuts and increase in input costs for cultivation (Josephrajkumar et al. 2018). Traditional pesticides are harmful to natural enemies, including predators and parasitoids, and may cause a resurgence in many sucking pests, such as whiteflies. The present incidence of RSW in India is concerning, because of its polyphagous nature and capacity to spread to new locations, and a systematic approach is necessary to manage this invasion (Selvaraj et al. 2017). To manage these groups of insects, biological control methods should be incorporated into integrated pest management strategies (Skinner et al. 2014).

A wide range of entomopathogenic fungi (EPF) are known to infect whiteflies naturally worldwide. Among them, *Isaria fumosorosea* Wize formerly (*Paecilomyces fumosoroseus*) (Hypocreales: Cordycipitaceae) is an effective biocontrol agent against whiteflies. It causes epizootics under natural field conditions (Luangsa-Ard et al. 2005). *I. fumosorosea* is widely used to manage *A. rugioperculatus* and *P. bondari* infesting coconut in Florida (Ali et al. 2015; Kumar et al. 2018) and *Singhiella simplex* infesting ficus (Avery et al. 2019). The blastospore formulation of *I. fumosorosea* (ICAR-NBAIR Pfu-5) has shown potential in managing RSW (Sumalatha et al. 2020). However, some insect growth regulators, biopesticides and conventional insecticides showed effectiveness against RSW (Pradhan et al. 2020).

The efficacy of *I. fumosorosea* (ICAR-NBAIR Pfu-5) in the field can be enhanced when it is used in combination with appropriate insecticides at a reduced rate for effective pest control. This technique might be an alternate choice for farmers for managing *A. rugioperculatus,* as pesticide usage can be reduced, resulting in less risk to the environment and resistance. The present study was undertaken to assess the efficacy of native isolate *I. fumosorosea* (ICAR-NBAIR Pfu-5) alone and in combination with selected insecticides insecticides and biopesticides at reduced rate for the management of *A. rugioperculatus* Martin on coconut.

Table 1 Experimental treatments

Treatments	
T_1	*Isaria fumosorosea* (ICAR-NBAIR Pfu-5) @5 ml/l alone
T_2	*Isaria fumosorosea* (ICAR-NBAIR Pfu-5) @5 ml/l + Buprofezin 25% SC @0.6 ml/l
T_3	*Isaria fumosorosea* (ICAR-NBAIR Pfu-5) @5 ml/l + Buprofezin 25% SC @1.25 ml/l
T_4	*Isaria fumosorosea* (ICAR-NBAIR Pfu-5) @5 ml/l + Pongamia soap @10 g/l
T_5	*Isaria fumosorosea* (ICAR-NBAIR Pfu-5) @5 ml/l + Fish oil rosin soap @2 ml/l
T_6	*Isaria fumosorosea* (ICAR-NBAIR Pfu-5) @5 ml/l + Azadirachtin10000 ppm @2 ml/l
T_7	*Isaria fumosorosea* (ICAR-NBAIR Pfu-5) @5 ml/l + Profenophos 50 EC @2 ml/l
T_8	Untreated control

Methods

Efficacy of *I. fumosorosea* (ICAR NBAIR Pfu-5) was studied alone and in combination with various insecticides and biopesticides against *A. rugioperculatus* on coconut palms in Kommanalu village (14° 3′ 32.56″ N 75° 32′ 47.4″ E, 610 m), Shivamogga, Karnataka, India, in a randomized complete block design (RCBD) during summer months (April and May) in 2021. Complete treatment details are presented in Table 1. For screening, *I. fumosorosea* (ICAR-NBAIR Pfu-5) blastopore oil formulation was procured from the ICAR-NBAIR fungal repository. Pongamia soap was obtained from the ICAR- IIHR, Bangalore. Buprofezin 25% SC, profenophos 50 EC, fish oil rosin soap and azadirachtin10000 ppm were obtained from local pesticides dealer in Shivamogga, Karnataka. Thirty-two randomly selected infested coconut palms (age: 3 years, variety: Arsikere tall, height: 2.0 m) were labelled, and eight treatments with four replications were used. Two sprays were performed at 15-day intervals. Fortune Agro battery-operated Knapsack high-volume sprayer with a capacity of 16 l was used. Water was used to calibrate the sprayer before each application. During the evening hrs (4–6 PM), spray solutions were sprayed at a rate of 5 l/palm.

Observations on the RSW population were taken on three parameters, infestation/palm (%), intensity of RSW (%) and mean live colonies.

Infestation of RSW per Palm (%)

Infestation of RSW made before and after spraying was determined using the formula suggested by (Visalakshi et al. 2021).

$$\text{RSW infestation/palm(\%)} = \frac{\text{Number of fronds infested by RSW}}{\text{Total fronds/palm}} \times 100$$

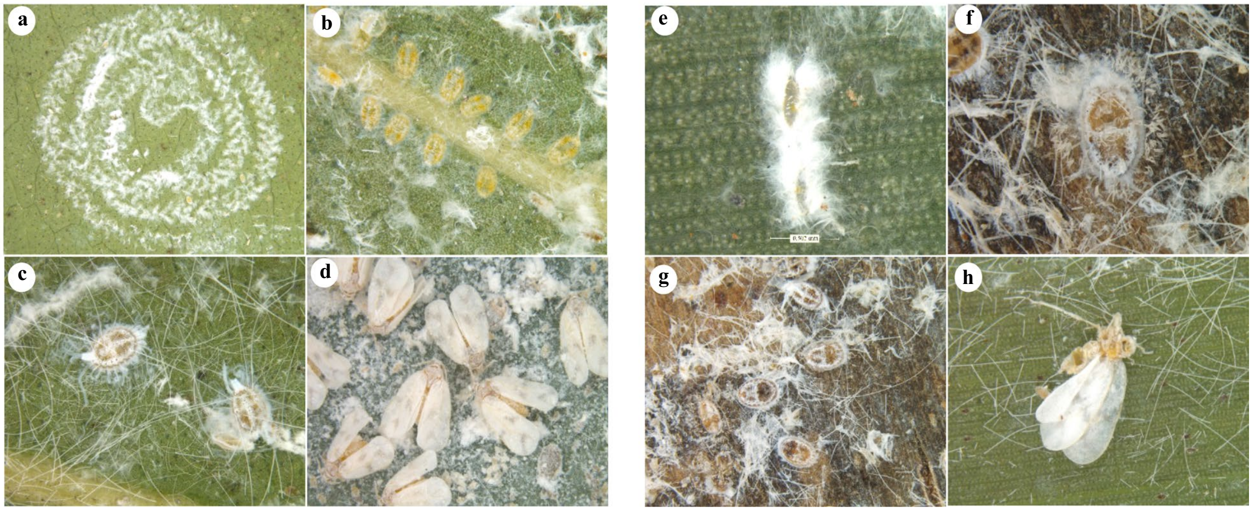

Fig. 1 *Aleurodicus rugioperculatus:* **a**–**d** Healthy life stages; **e**–**h** *Isaria fumosorosea* infested life stages after spraying

Intensity of RSW infestation (%)

Intensity of pest damage was taken from four infested fronds per palm from the outer and middle whorl representing the four directions. Per cent intensity of RSW was made using the formula (Visalakshi et al. 2021).

$$\text{Intensity of RSW infestation}(\%) = \frac{\text{Number of leaflets infested by RSW}}{\text{Total leaflets/frond}} \times 100$$

Mean live colonies of RSW/leaflet

Live colonies count was taken from 10 randomly selected leaflets from each palm before and after spraying (Visalakshi et al. 2021).

Observations on the RSW population were made one day before spraying, 7 and 15 days after spraying for each treatment. The leaf samples were brought to the laboratory and examined *I. fumosorosea* mycelium growth and ascertain the mortality RSW using a stereo binocular microscope at $10\times$ to $40\times$ magnification. Efficacy was computed based on a reduction in the number of RSW live colonies compared to the untreated check.

Statistical analysis

The data were analysed using the analysis of variance technique (ANOVA) of RCBD in SPSS software, and the results were interpreted at a 5% level of significance. For comparing treatment means, the Duncan multiple range test (DMRT) was used (Duncan 1955).

Results

The present study revealed that the EPF, *I. fumosorosea* (ICAR-NBAIR Pfu-5), had significant virulence and fungal mycelia development on RSW eggs, nymphs and adults. *I. fumosorosea* infection affects egg hatching, and nymphal mortality especially fourth-instar nymph (pupae), which led to a severe reduction in adult emergence and further perpetuation of RSW population. Substantial mycosis and deformation were seen in newly emerging adults from *I. fumosorosea*-treated nymphs, which led to a drastic reduction of adult emergence that may result in less perpetuation of the pest in the coconut ecosystem (Fig. 1). Further, the combination of *I. fumosorosea* (ICAR-NBAIR Pfu-5) @ 5 ml /l sublethal dose of profenophos 50 EC@ 2 ml/l was superior to all other treatments after 7 and 15 days, with significantly lower RSW population on coconut palms.

Infestation of RSW per palm (%)

Pre-treatment per cent infestation was uniform in all the treatments as indicated by non-significant differences, ranging from 43.44 to 51.99% (Fig. 2). However, after 7 days of topical application, *I. fumosorosea* (ICAR-NBAIR Pfu-5) @ 5 ml/l + profenophos 50 EC @ 2 ml/l

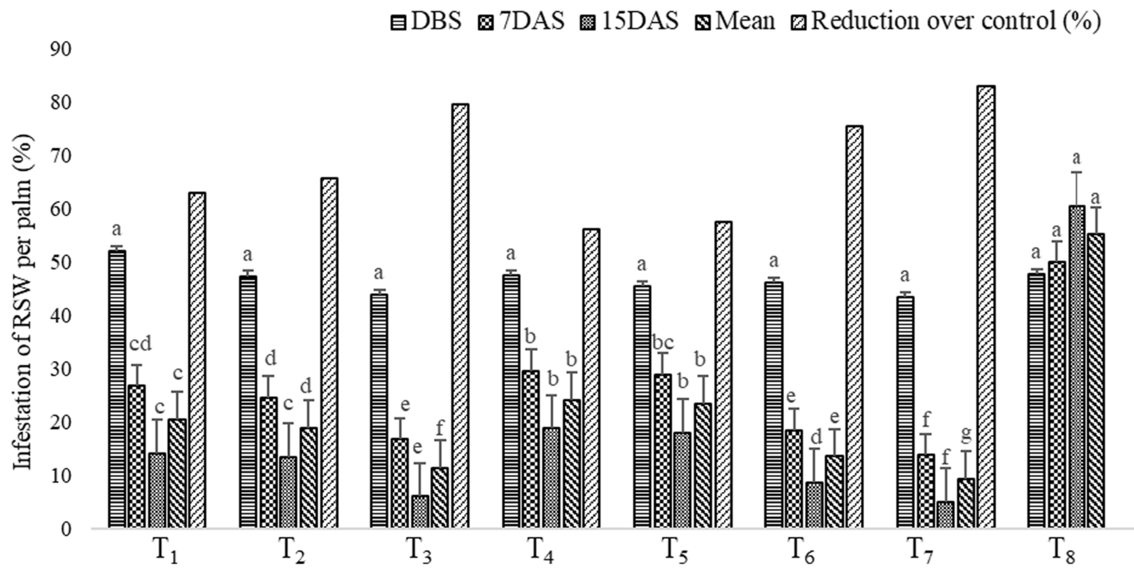

Fig. 2 Efficacy of *Isaria fumosorosea* alone and in combination with selected insecticides and biopesticides on per cent infestation of RSW on coconut in summer (first and second spray pooled data). Means followed by the same letter do not differ significantly by DMRT (P = 0.05); DBS = day before spray; DAS = day after spray; RSW = rugose spiraling whitefly; infestation of RSW/palm (%): (number of fronds infested by RSW/total fronds per palm) × 100

(T_7) showed the lowest infestation (13.84%), followed by *I. fumosorosea* (ICAR-NBAIR Pfu-5) @ 5 ml/l + buprofezin 25% SC @ 1.25 ml/l (T_3) with (16.72%), *I. fumosorosea* (ICAR-NBAIR Pfu-5) @ 5 ml/l + azadirachtin10000 ppm @ 2 ml/l (T_6) with (18.47%) and *I. fumosorosea* (ICAR-NBAIR Pfu-5) @ 5 ml/l + buprofezin 25% SC @ 0.6 ml/l (T_2) with (24.53%) over *I. fumosorosea* (ICAR-NBAIR Pfu-5) @ 5 ml/l alone (T_1) with (26.74%), while the highest RSW infestation was recorded in *I. fumosorosea* (ICAR-NBAIR Pfu-5) @ 5 ml/l + pongamia soap @10 g/l (T_4) with (29.60%) followed by *I. fumosorosea* (ICAR-NBAIR Pfu-5) @ 5 ml/l + fish oil rosin soap @ 2 ml/l (T_5) with (28.89%), whereas untreated control, (T_8) with (49.90%) infestation (Fig. 2).

Similarly, after 15 days, T_7 dominated all other treatments with the lowest infestation (4.95%), followed by T_3 with (6.06%), T_6 with (8.75%) and T_2 with (13.35%) over *I. fumosorosea* (ICAR-NBAIR Pfu-5) alone (T_1) with (14.16%), while the highest RSW infestation was recorded in T_4 with (18.77%) followed by T_5 with (17.98%), whereas untreated control, T_8 with (60.44%) infestation (Fig. 2).

However, after topical application of T_7 showed the lowest mean infestation (9.40%), followed by T_3 with (11.39%) and T_6 with (13.61%) over *I. fumosorosea* (ICAR-NBAIR Pfu-5) alone (T_1) with (20.45%), while the highest RSW infestation was recorded in T_4 (24.18%) followed by T_5 (23.43%), whereas untreated control, T_8 with 55.17% infestation (Fig. 2).

The application of T_7 resulted in the highest reduction of RSW infestation (82.97%) over *I. fumosorosea* (ICAR-NBAIR Pfu-5) alone, T_1 with (62.93%) and untreated control. Following that, T_3 with (79.35%) and T_6 with (75.33%) (Fig. 2).

Intensity of RSW infestation (%)

Pre-treatment RSW intensity was uniform across all treatments, as evidenced by non-significant differences ranging from 47.32 to 59.79% (Fig. 3). After applying *I. fumosorosea* (ICAR-NBAIR Pfu-5) @ 5 ml/l + profenophos 50 EC @ 2 ml/l (T_7), the percentage intensity of RSW was drastically reduced.

After 7 days of topical application, T_7 showed the lowest (16.64%) intensity, followed by T_3 with (20.39%), T_6 with (23.37%) and T_2 with (24.97%) over *I. fumosorosea* (ICAR-NBAIR Pfu-5) alone (T_1) with (27.88%), while the highest RSW intensity (30.59%) was recorded in T_4 followed by T_5 with (30.01%), whereas untreated control, T_8 with (51.35%) intensity (Fig. 3).

Similarly, after 15 days, T_7 showed the lowest (4.05%) intensity, followed by T_3 with (6.34%), T_6 with (7.30%) and T_2 with (10.60%) over *I. fumosorosea* (ICAR-NBAIR Pfu-5) alone (T_1) with (11.84%), while the highest RSW intensity was recorded in T_4 with (15.62%) followed by T_5 with (14.08%), whereas untreated control, T_8 with (54.71%) intensity (Fig. 3).

Palms treated with T_7 surpassed all other treatments with the lowest (10.34%) mean per cent intensity

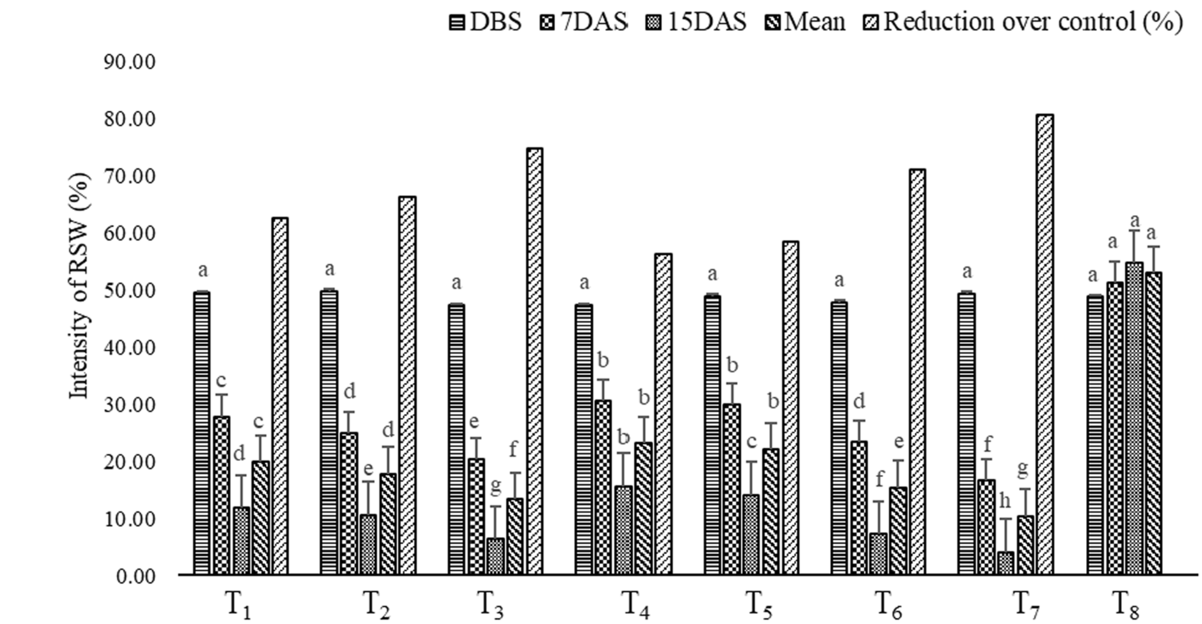

Fig. 3 Efficacy of *Isaria fumosorosea* alone and in combination with selected insecticides and biopesticides on per cent intensity of RSW on coconut in summer (first and second spray pooled data). Means followed by the same letter do not differ significantly by DMRT (P = 0.05); DBS = day before spray; DAS = day after spray; RSW = rugose spiraling whitefly; intensity of RSW (%): (number of leaflets infested by RSW/total leaflets per frond) × 100

followed by T_3 with (13.37%) and T_6 with (15.33%) over *I. fumosorosea* (ICAR-NBAIR Pfu-5) alone, T_1 with (19.86%). While RSW intensity in T_5 with (22.04%) and T_4 with (23.11%) was recorded. Whereas, untreated control T_8 with (53.03%) intensity (Fig. 3).

The treatment of T_7 resulted in the greatest reduction (80.49%) of RSW intensity over *I. fumosorosea* (ICAR-NBAIR Pfu-5) alone, T_7 with (62.55%) and untreated control. The next best treatments are T_3 with (74.79%) and T_6 with (71.09%) (Fig. 3).

Mean live colonies of RSW/leaflet

The pre-treatment mean live colonies of RSW were uniform in all the treatments as indicated by non-significant differences, ranging from 11.92 to 12.56 colonies/ leaflet (Fig. 4). The RSW mean live colonies were drastically reduced after palms were treated with *I. fumosorosea* (ICAR-NBAIR Pfu-5) @ 5 ml/l + profenophos 50 EC @ 2 ml/l.

After 7 days, T_7 showed the lowest (4.25 mean live colonies/leaflet), followed by T_3 with (5.04 mean live colonies/leaflet), T_6 with (5.60 mean live colonies/leaflet) and T_2 with (5.88 mean live colonies/leaflet) over *I. fumosorosea* (ICAR-NBAIR Pfu-5) alone (T_1) with (6.15 mean live colonies/leaflet). However, the highest RSW mean live colonies/leaflet were recorded in T_4 with (6.80 mean live colonies/leaflet) followed by T_5 with (6.48 mean live colonies/leaflet), whereas untreated control, T_8 with 12.84 mean live colonies/leaflet (Fig. 4).

After 15 days, T_7 showed the lowest (1.23 mean live colonies/leaflet) followed by T_3 with (1.91 mean live colonies/leaflet), T_6 with (2.40 mean live colonies/leaflet) and T_2 with (2.78 mean live colonies/leaflet) over *I. fumosorosea* (ICAR-NBAIR Pfu-5) alone (T_1) with (3.23 mean live colonies/leaflet), while the highest RSW mean live colonies were recorded in T_4 with (4.11 mean live colonies/leaflet) followed by T_5 with (3.80 mean live colonies/leaflet) and, however, untreated control T_8 with 14.10 mean live colonies/leaflet (Fig. 4).

Palms treated with T_7 were significantly superior to all other treatments with the lowest (2.74 overall mean live colonies/leaflet) followed by T_3 with (3.48 mean live colonies/leaflet), T_6 with (4.00 mean live colonies/leaflet) over *I. fumosorosea* (ICAR-NBAIR Pfu-5) alone (T_1) with (4.69 mean live colonies/leaflet), while T_5 with (5.14 mean live colonies/leaflet) and T_4 with (5.38 mean live colonies/leaflet) were recorded and, however, untreated control (T_8) with 13.47mean live colonies/leaflet (Fig. 4).

In general, palms treated with T_7 showed the greatest reduction (79.68%) mean number of RSW live colonies over *I. fumosorosea* (ICAR-NBAIR Pfu-5) alone (T_1) with (65.20%) and untreated control. This was followed by T_3 with (74.20%) and T_6 with (70.30%) reduction (Fig. 4).

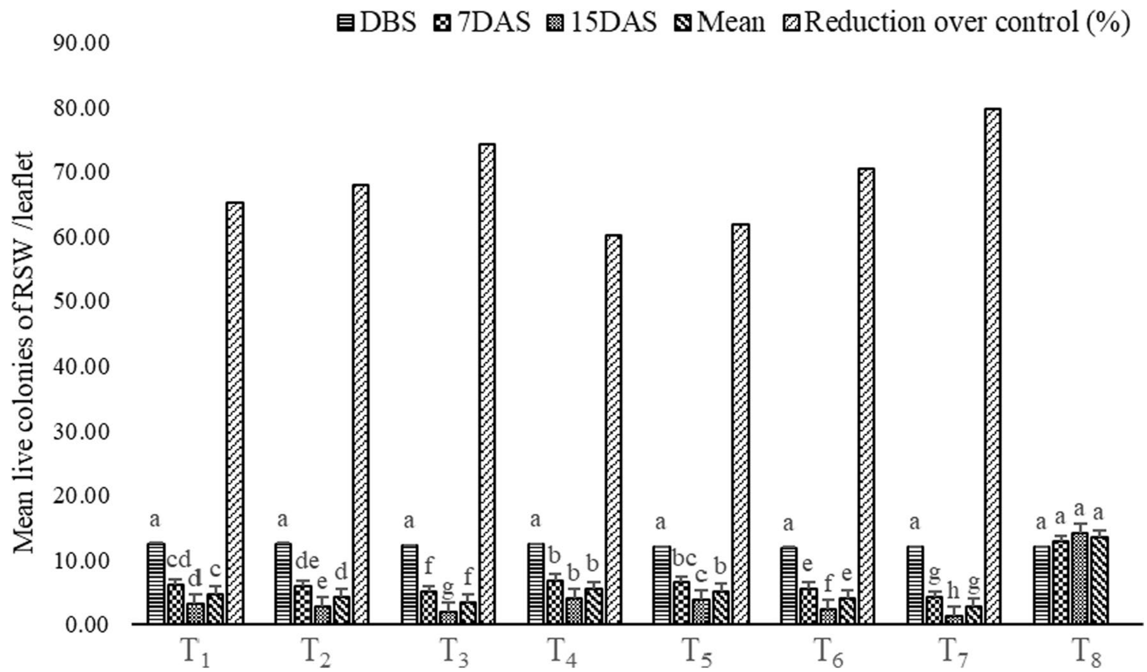

Fig. 4 Efficacy of *Isaria fumosorosea* alone and in combination with selected insecticides and biopesticides on mean live colonies of RSW on coconut in summer (first and second spray pooled data). Means followed by the same letter do not differ significantly by DMRT (P = 0.05); DBS = day before spray; DAS = day after spray; RSW = rugose spiraling whitefly; mean number of whitefly live colonies/leaflet: (ten leaflets from each palm)

Discussion

In pest management, the use of mixtures of botanical and microbial insecticides is more stable and cost-effective with less non-target effects (Yi et al. 2012). Even though microbial organisms and chemical insecticides can be used together, they must be compatible. In general, fungicides are found to harm myco-insecticides, while insecticides do not have any impact on EPF, when the chemicals are used at the appropriate levels (Chen and Feng 2003).

It is well known that *I. fumosorosea* and chemical insecticides have different modes of action. The fungal pathogen infection process is dependent on many biological events, including the adhesion of fungi to insect cuticles, spore germination and hyphal growth. In order to penetrate the insect cuticle, the fungal appressorium exerts mechanical pressure and excretes enzymes to degrade the cuticle (Altre et al. 1999).

I. fumosorosea is a widely used and studied fungus attacking nymphs and adults of whiteflies such as *Bemisia* spp. (Lacey et al. 2008), *A. dispersus* (Sanchez and Castillo, 2008), *Trialeurodes vaporariorum* (Boopathi et al. 2013), *A. rugioperculatus* (Sumalatha et al. 2020), *P. bondari* (Ali et al. 2015), *P. minei* and *A. atratus* (Sumalatha et al. 2020) and *Singhiella simplex* (Avery et al. 2019).

The present findings are in agreement with Visalakshi et al. (2021), who reported a significant reduction of mean coconut rugose spiraling whitefly (RSW) live colonies, ranged from (40.76 to 67.57%). However, Kumar et al. (2018) observed that *I. fumosorosea* significantly suppressed *A. rugioperculatus* eggs (90.00%) than the control at various sampling intervals two and eight DAT in Florida. Sanchez and Castillo (2008) reported that *I. fumosorosea* reduced egg hatching by 50% in a similarly related whitefly species, *A. dispersus*. Boopathi et al. (2013) reported *Metarhizium anisopliae* (M2 strain) and *P. fumosoroseus* (P1 strain) caused 37.3 and 22.6% of egg mortality at 8 days after treatment (DAT). One of the most significant characteristics of EPF is the ability to control pests during the initial stage. As a result, following insect population growth and crop damage will be reduced.

Profenophos 50 EC belongs to the organophosphate insecticides group, which inhibits the acetylcholinesterase enzyme, leading to paralysis and death of pest organisms (Elbert et al. 2008). Profenophos showed a significant ovicidal effect on *A. rugioperculatus* five days after treatment as the colour of eggs changed to dark brown (Pradhan et al. 2020).

Buprofezin 25 SC was the first selective insect growth regulator developed to control *B. tabaci* on cotton (Horowitz and Ishaaya 1992). It is a chitin synthesis inhibitor that interferes with cuticle formation during immature developmental stages leading to pest mortality (Palumbo et al. 2001). However, field studies showed that the efficacy of buprofezin treatments (alone or mixed with *I. fumosorosea*) was higher than *I. fumosorosea* alone. A significant reduction in the RSW population was reported for more than five weeks in buprofezin alone and more than seven weeks in the combination treatments (Kumar et al. 2018).

However, azadirahtin, a primary active ingredient obtained from the seeds of the plant *Azadirachta indica*, acts as a growth regulator, antifeedant and repellent against insects from different genera including species that feed by sucking plant juices (Copping and Duke 2007). A combination of azadirachtin and *I. fumosorosea* against the whitefly *Bemisia argentifolii* in laboratory bioassays resulted in up to 90% nymphal mortality. However, the combined effects were less than additive and azadirachtin had a moderate inhibitory effect on germination and growth of *I. fumosorosea* (James 2003). Hence, the present findings supplement the efficacy of neem against other species of whiteflies.

Huang et al. (2006) reported that combinations of *I. fumosorosea* with imidacloprid exhibited a strong synergistic effect against *B. tabaci.* Oil-based emulsifiable formulations of *I. fumosorosea* alone and combined with imidacloprid were applied against *T. vaporariorum* on lettuce grown in the greenhouse. Feng et al. (2004) found that the fungus was highly effective alone and combined with imidacloprid at around 15% of its recommended rate.

The different modes of action might contribute to the synergistic effects of insecticide mixtures. It is probably that the slower pest killing speed of EPF in combination with the faster pest-killing chemical insecticides might also contribute to the synergistic interaction. However, among the different chemical insecticides and biopesticides tested, profenophos, buprofezin and azadirachtin were significantly effective in the RSW population.

This finding is consistent with that of other researchers who have studied similar pathogens and pesticides with fungal entomopathogens such as *I. fumosorosea* and aleyrodid pests (Huang et al. 2010). According to the obtained findings, there are positive interactions between *I. fumosorosea* and profenophos 50 EC and these interactions may serve as an effective control strategy for *A. rugioperculatus* in the coconut orchards.

Conclusion

The present study revealed that a combination of *I. fumosorosea* (ICAR-NBAIR Pfu-5) @ 5 ml/l + profenophos 50 EC @ 2 ml/l had the potential to manage *A. rugioperculatus*. Spraying twice in 15-day interval significantly reduced RSW infestation (82.97%), intensity (80.49%) and mean live colonies (79.68%). It is necessary to investigate the negative effects of this combination on parasitoids and predators under field conditions to develop the biointensive integrated pest management of RSW in coconut.

Abbreviations

ICAR: Indian Council of Agricultural Research; NBAIR: National Bureau of Agricultural Insect Resources; RSW: Rugose spiraling whitefly; Pfu-5: *Paecilomyces fumosoroseus* Strain-5; L: Litre; EC: Emulsifiable concentrates; SC: Suspension concentrate; IIHR: Indian Institute of Horticultural Research; G: Gram; ppm: Parts per million; SPSS: Statistical Package for the Social Sciences.

Acknowledgements

The authors gratefully acknowledge the facilities provided by Department of Entomology, KSNUAHS, Shivamogga, as well as the Indian Council of Agricultural Research, New Delhi, for Junior Research Fellowship for M.Sc. We would also to thank the director, ICAR-NBAIR, Bangalore, for providing the entomopathogenic fungus *Isaria fumosorosea* for conducting the experiment.

Author contributions

AS carried out the experiments for his postgraduate research; CMK, KS, BCH conceptualized the study and wrote the paper; KS, BCH, HBM provided research material and helped in conducting the experiments and analysed the data. All authors have read and approved the final manuscript.

Funding

Not applicable.

Availability of data and materials

Not applicable.

Declarations

Ethics approval and consent to participate

Not applicable.

Consent for publication

All the authors give their consent to publish the submitted manuscript as "Original paper" in EJBPC.

Competing interests

The authors whose names are listed immediately below certify that they have NO affiliations with or involvement in any organization or entity with any financial interest (such as honoraria; educational grants; participation in speakers' bureaus; membership, employment, consultancies, stock ownership, or other equity interest; and expert testimony or patent-licensing arrangements), or non-financial interest (such as personal or professional relationships, affiliations, knowledge or beliefs) in the subject matter or materials discussed in this manuscript.

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Received: 26 May 2022 Accepted: 18 August 2022
Published online: 03 September 2022

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