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Mortality Effects of *Beauveria bassiana* and *Purpureocillium lilacinum* Isolates and Efficacy of a Wettable Formulation on *Palemona prasina* (Hemiptera: Pentatomidae) Nymphs

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ABSTRACT

In this study, the mortality effect of two *Beauveria bassiana* and two *Purpureocillium lilacinum* isolates on the 4th stage nymphs of *Palemona prassiana* was determined. The most lethal isolate was formulated as wettable powder and tested on the pest. Furthermore, Y-tube olfactometry tests were conducted to detect behavioral response of the nymphs in presence of the fungus. All the experiments were carried out under controlled conditions. The mortality varied depending on the isolates between 28.51% and 82.14% on the 12th day. *Beauveria bassiana* FAI-38 caused the highest mortality (82.14% at 1x10^{8} conidia ml^{-1}) with LC_{50} and LT_{50} estimations of 3.3x10^{6} conidia ml^{-1} and 8.4 days, respectively. According to data taken 6 and 12 days after application, the wettable powder formulation was found to be significantly more effective (89.65% at 1x10^{7} conidia ml^{-1}, LT_{50} 6.08 days). According to the Y-tube olfactometry tests, the nymphs exhibited avoidance from unformulated *B. bassiana* spores; however, once the spores were formulated as wettable powder, the behavior of the insects changed to neutral. It is concluded that *Beauveria bassiana* FAI-38 presents a potential as a control agent, and the wettable powder formulation of the fungus improves its effectiveness by increasing mortality and removing repellency effect of the fungal spores.

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Beauveria bassiana ve *Purpureocillium lilacinum* İzolatlarının *Palemona prasina* (Hemiptera: Pentatomidae) Nimflerine Ölüm Etkisi ve Islanabilir Toz Formulasyonun Etkinliği

ABSTRACT

Bu çalışmada, *Palemona prassiana*'nın dördüncü dönem nimflerine iki *Beauveria bassiana* ve iki *Purpureocillium lilacinum* izolatının ölümcül etkileri belirlenmiş, en yüksek ölüm etkisi gösteren izolatın bir ıslanabilir toz formülasyonu hazırlanarak test edilmiştir. Ayrıca, zararlının fungusa gösterdiği davranış tepkisi Y-tüpü olfaktometre testleri ile ortaya konmuştur. Tüm denemeler kontrollü şartlar altında gerçekleştirilmiştir. Ölüm oranları izolata bağlı olarak 12. günde %28.51 ile %82.14 arasında farklılık göstermiştir. En yüksek ölüm oranı *Beauveria bassiana* FAI-38 izolatında gözlenmiştir (1x10^{8} konidi/ml konsantrasyonda %82.14) ve LC_{50}, LT_{50} değerleri sırasıyla 3.3x10^{6} konidi/ml ve 8.4 gün olarak hesaplanmıştır. Uygulamadan 6 ve 12 gün sonra alınan verilere göre, ıslanabilir toz formülasyonu önemli derecede daha etkili bulunmuştur (1x10^{7} konidi ml^{-1}'de %89.65, LT_{50} 6.08 gün). Y-tüpü olfaktometre testlerine göre, nimfler, formüle edilmemiş *B. bassiana* sporlarından uzaklaşma tepkisi sergilemiştir. Fungus sporlar ıslanabilir toz olarak formüle edildiğinde böcekler herhangi bir davranış göstermemişlerdir. *Beauveria bassiana* FAI-38'in mücadele etmeni olarak potansiyel gösterdiği; ölüm oranını artırması ve sporların uzaklaştırıcı etkisini ortadan kaldırması nedeniyle ıslanabilir toz formülasyonun fungusun etkinliğini geliştirdiği sonucuna varılmıştır.

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INTRODUCTION

Green shield bug, *Palomena prasina* L. (Hemiptera: Pentatomidae) is an important pest of hazelnuts causing yield and quality losses in Turkey and other hazelnut-producing countries. Exporters' associations complain about poor quality kernels mostly due to green shield bugs. This pest causes yield reduction and reduced quality (shriveled and/or spotted kernels) (Ak et al., 2017), the latter of which becomes quite significant, especially for those produced for export (Kurt, 1975). Currently, two pesticide applications are required to control this pest in hazelnut orchards in Turkey. The first is in the second half of April against overwintered adults, and the second one in the first half of July if the nymphs are detected in orchards. Unfortunately, the only available control method for this pest is spraying chemical insecticides. Appropriate selection of plant protection products is of great importance because the second application is very close to harvest imposing risks for chemical residues in export products. Besides, although chemical insecticides are effective, concerns about their negative side effects on the environment and people directed researchers to seek alternatives suitable for sustainable agriculture, organic farming, and protecting biological diversity. As an alternative approach, biological control has been used or evaluated for various insect pests. It is considered to be widespread all over the world in the future due to pest resistance to effective chemicals, difficulties in developing new ones, public concerns about food safety and genetically modified agricultural products, and increasing demand for organic products (Birişik, 2015). The International Organization for Biological Control (IOBC) estimated that the share of biological control products in the total plant protection product market will be 30-35% in 2050. Today, around 800 chemical agents are widely used in the world to control pest organisms.

There are a few studies on the biological control of hazelnut green shield bug using entomopathogenic fungi. Erper et al. (2016) tested isolates of *Beauveria bassiana*, *Isaria fumosorosea*, *Simplicillium lamellicola*, *Lecanicillium muscarium* to the nymphs in controlled conditions, and found that the *B. bassiana* and one *L. muscarium* were the most virulent with 95 and 98% mortality, respectively, in 12 days. Özdemir (2021) obtained even higher mortalities in a shorter time using isolates from seven species including *B. bassiana*, *B. pseudobassiana*, *I. fumosorosea*. Yiğit (2022) applied *B. bassiana*, *Metarhizium anisopliae* and *I. fumosorosea* against nymphs and adults under controlled conditions and in the field. They found one of the *B. bassiana* isolates as the most promising. The isolate caused 75-80% mortality in 14 days in field trials. Studies were conducted under controlled conditions with the exception of a field trial by Yiğit (2022), where fungal spores were applied without formulating. For the success of *B. bassiana* applications, the fungus should have persistence on foliage as much as high pathogenicity to the targeted pest. Once a potential entomopathogenic fungus is obtained, formulation of the fungus can help to fulfill this requirement with an even higher potency as a biological control agent. There are studies on testing formulated entomopathogenic fungi on other pest species in the family Pentatomidae. Parker et al. (2015) used wettable powder and emulsifiable suspension formulations of *B. bassiana* against nymphs of brown marmored sting bug, wettable powder formulation being more efficacious. Sosa-Gomez et al (1998) tested kaolin-based powder formulations of *M. anisopliae* and *B. bassiana* for the control of *Nezara viridula, Piezodorus guildinii*, and *Euschistus heros* in soybean plots.

One other issue for the success of a fungus is concerned with the behavior of the pest insect towards the fungus to be applied. For infection to occur, the fungus should come in contact with the insect's body during its foraging activity. Recognition of the pathogen and avoidance by the insect can hinder the success of the application of the fungus as a biological control agent (Doğan et al., 2017). The level of such behavioral response was found to depend on insect-fungus interaction and spore concentration (İncir, 2018). It is mostly demonstrated that insects show avoidance from entomopathogenic fungi (Baverstock et al., 2010; Wei et al., 2020; Avery et al., 2021; Daisy 2022; Geedi et al., 2022). However, fungal spores mixed with some compounds or ingredients for combined applications or to formulate the spores did not induce such avoidance behavior (Wang & Powell 2004; Fernandez-Grandon et al. 2020). Therefore, formulating entomopathogenic fungi at appropriate concentrations can increase the efficacy (Wang & Powell, 2004.

In this study, a potential entomopathogenic fungus was selected amongst isolates obtained from hazelnut green shield bugs, and it was formulated for higher pathogenicity. The avoidance behavior of the pest was also tested for its suitability for later field applications.

MATERIAL and METHODS

Insect Culture

Overwintered *Palomena prasina* adults were collected from hazelnut orchards to start a culture. Adults were maintained at 25±2°C and 75±5% relative humidity in 16:8 (L:D) photoperiod in a climate room. To obtain their eggs, food, water, and filter paper were placed inside glass jars. Beans that were grown in a greenhouse in the hazelnut research institution were used as food. Before using the beans were cleaned by dipping in 3% sodium bicarbonate solution for 5 minutes. As a supplement, 5-10 pieces of unsalted sunflower seeds or half a slice of apple were also provided. The green beans, apples, and water were changed every other day, while sunflower seeds were changed every 4-5 days. Egg clusters were transferred to a 150 ml plastic cup and the insect-rearing room was kept until hatching. Once they reached the second stage, the nymphs were provided with water and green beans. The nymphs at the 4th stage were used in the experiments.

Fungi

In this study, two *Beauveria bassiana* and two *Purpureocillium lilacinum* isolates were tested to evaluate their pathogenicity against P. prasina. The fungi were previously isolated from *Palomena prasina* adults and identified with morphological and molecular studies (unpublished data). B. bassiana FAI38 and FAI22 were isolated from hosts collected from the Central District of Giresun Province and Perşembe District of Ordu Province, respectively. P. lilacinum FAI1 and FAI70 were obtained from hosts gathered from the Central District of Giresun Province and Gülyalı District of Ordu Province, respectively.

Preparing conidial suspensions

First, the fungi were inoculated on PDA and kept in the dark at 26±2 °C for 4 weeks. The conidia on the surface were collected with a sterile polystyrene spreader within 0.02% tween80 solution. This suspension was passed through sterile gauze after vortexing and the concentration was determined by using a hemocytometer. The density was adjusted by diluting to the concentration to be used in the test. Conidia viability was determined by germination test. 100 µl suspension prepared at 1×10^5 conidia ml^{-1} was spread on 1.5% aqueous agar and incubated in a closed Petri dish at 26±2°C in the dark for 24 hours. During examination under a light microscope, conidia with germ tubes equal to or longer than spore length or diameter were considered germinated.

Fungus Screening Bioassay

Each experimental unit was set by using two 155 ml plastic cups, one upside down on top of the other one, forming 310 ml space for the insects. The bottom of the cup on the top was cut off and covered with tulle for aeration prior to the setup. In each experimental unit, a piece of twig with a three-leaved fruit peel was used as nutrition for the insects and 10 ml of distilled water was added to keep the plant alive. The husk fruits with leaves were collected from hazelnut trees where foliar plant protection products had not been applied in Giresun Hazelnut Research Institute. In order to keep the coniferous leaf fresh, 1 mm was cut from the petiole every other day. Ten 4th instar hazelnut green shield bug nymphs were placed in each unit. Two ml conidial suspension (1×10^8 conidia ml^{-1}) of designated fungus was applied to the nymphs by a hand sprayer. The nymphs in control units were sprayed with sterile %0.02 tween80 without fungal spores. The experiment had three replications and was carried out at 26±2 °C temperature, 75±5% relative humidity in a climate cabinet with 16 h light / 8 h dark photoperiod. The insects were checked on the 6th and 12th days, dead insects were counted, recorded, and removed from the experimental units. The Abbott formula was used to arrange the control measurements. The arcsine transformation was applied to the mortality rates. They were subjected to one-way ANOVA and Duncan multiple comparison tests using IBM SPSS statistics 23 program.

Preparation and efficacy of *Beauveria bassiana* FAI38 Formulation

A wettable powder formulation of *Beauveria bassiana* FAI38 was prepared and tested against *P. prasina*. The fungus conidia were obtained using the solid fermentation method described by Barış & Er (2021). Rice was used as a substrate for the fermentation process. The spores obtained in powder form were stored at +4 °C until the formulation was prepared. The formulation included 68% rice flour, 20% conidia (4×10^{11} conidia kg^{-1}), and 12% adjuvants and surfactant. The formulation was used at the ratio of 2.5 gr per 1 lt water for applications. The experiment was conducted and the data were analyzed as described above for Fungus Screening Bioassays. The formulation was used at the above-mentioned ratio (1X) and its 10-fold (10X) together with two corresponding concentrations (1×10^6 and 1×10^7 conidia ml^{-1}) of the fungal conidia without formulating for comparison. The nymphs in control units were sprayed with %0.02 tween, sterile distilled water, and without fungal spores. The insects were checked on 6th and 12th days, dead insects were counted, recorded, and removed from the experimental units. An additional experiment was carried out to calculate the LT_{50} of the formulation for 10X application according to the method described above for the estimation of LT_{50} value of unformulated spores.

Olfactometer Bioassays

In order to determine the behavioral response of *P. prasina* nymphs to the presence of *B. bassiana* spores, four sets of Y-tube olfactometry tests were carried out. In each test, the insects were presented with two choices of food (3 fruit husks and one hazelnut leaf) with different treatments. All the treatments were achieved by spraying 2 ml of designated suspension onto the food while the controls received the same amount of %0.02 tween and distilled water. Four sets of Y-tube olfactometry tests were (1) unformulated FAI-38 spores vs control, (2) FAI-38 formulation vs control, (3) FAI-38 formulation vs formulation ingredients without spores, and (4) unformulated FAI-38 vs FAI-38 formulation. Unformulated spores were applied at 1×10^8 conidia ml^{-1} concentration and the formulation at 10X application ratios.

The lengths of the main arm and the side arms of the olfactometer were 10 cm and 24 cm, respectively. It was made of glass with 0.4 cm thickness. The air pressure was kept constant and was monitored with barometers connected to the entrance and exit of the olfactometer. The air was cleaned before entering the olfactometer by passing it through a glass balloon containing activated charcoal and then through a microfilter. The experiment was conducted under constant light at 26±2 °C temperature in a room. Between each trial, the Y-tube olfactometer assembly was cleaned with acetone, washed with distilled water, and dried in an oven at 150 °C for 40 minutes. In each test, thirty 4th instar nymphs were used individually. The nymphs were kept without food for 10 hours prior to the tests. Fifteen minutes were given for each insect to choose one of two arms after placing the insect at the entrance. Those that moved at least 5 cm into one arm were considered to make a decision. When an individual did not enter either of the arms before the end of the period, it was considered with no choice. Each experiment was repeated after the choices were replaced on opposite arms of the Y-tube olfactometer to eliminate any possible effects of the placement.

The chi-square test was applied to data once the preferences. The t-test was used to determine whether changing places of choice in the olfactometer makes a difference.

RESULTS and DISCUSSION

For fungus screening test, two *Beauveria bassiana* and two *Purpureocillium lilacinum* isolates were tested against 4th instar hazelnut green shield bug nymphs. Mortality data 6 and 12 days after treatment are presented in Figure 1.

Variation amongst 6 days post-treatment mortalities were statistically insignificant except for *P. lilacinum* FAI-1 causing lower nymph mortality than the rest. The mortality rate at the end of the 12th day varied between 28.51% and 82.14%. *Beauveria bassiana* FAI-38 provided the highest mortality 1×10^8 conidia ml^{-1} (82.14%) while the other isolates resulted in significantly lower nymphal mortalities. Therefore, *B. bassiana* FAI-38 was chosen as the most promising isolate and used in later experiments. Additionally, the mortality effect of *B. bassiana* FAI-38 was investigated further by concentration and time dependent probit analyses. Probit line equation for concentration-mortality relation was y=-4.434+0.680x (Figure 2) with LC_{50}, LC_{95} estimations of 3.3×10^6 conidia/ml (95% c.i.: 1×10^6 – 8.53×10^6) and 8.66×10^9 conidia ml^{-1} (95% c.i.: 2.18×10^8 – 9.61×10^9), respectively. The equation for time-mortality relation was y=-2.039+0.243x (Figure 3) with LT_{50} and LT_{95} values 8.403 days (95% c.i.: 7.781 – 9.117) and 15.183 days (95% c.i.: 13.750 – 17.280), respectively.

Erper et al. (2016) achieved 95% nymphal mortality at the end of the 12th day in an experiment where they applied *B. bassiana* at 1×10^8 conidia ml^{-1} concentration at 25°C temperature and 90±5% relative humidity. Ozdemir (2021), Yigit (2022) reached up to 100% mortality at the end of the 14 days after spraying *B. bassiana* at 1×10^8 conidia ml^{-1} at 25°C temperature. Since Ozdemir (2021) carried the experiment in sealed petri dishes and Yigit (2022) in closed cups, the air humidity was most likely to be rather high favoring initiation of fungal infection faster and by more spores. Similarly, the experiment of Erper et al. (2016) was conducted at high ambient humidity. Ambient temperature and humidity, ventilation, light, air and the condition of the host itself have important effects on the pathogenicity of entomopathogenic fungi (Padmini & Padmaja, 2010). As many *Beauveria* isolates show host specificity (Thakur & Sandhu, 2010), at least some of the differences in mortality levels could be due to the isolates themselves.

The wettable powder formulation of *B. bassiana* FAI38 was tested against *P. prasina* nymphs at 1X and 10X and the results are given in Figure 4 along with the results of corresponding unformulated concentrations of the spores. Variation in mortality was significant. The efficacy of the formulations was significantly higher than their corresponding unformulated conidia treatments both on the 6th and 12th days post application. The formulation at 1X delivered statistically the same result with 10 fold unformulated conidia, indicating a requirement of about 10-fold less conidia with formulation to reach the same mortality level. The equation for the time-mortality relation was y=-1.139+0.187x (Figure 5) with LT_{50} and LT_{95} values 6.076 days (95% c.i.: 5.287 – 6.825) and 14.849 days (95% c.i.: 13.131 – 17.518), respectively. Both LT_{50} and LT_{95} values for formulation were lower than those for unformulated conidia even though unformulated conidia were used at higher spore concentration (10X formulation application corresponds to 1×10^7 conidia

ml^{-1}, unformulated spore concentration was 1×10^8 conidia ml^{-1}). While the confidence intervals of the LT_{95} values coincide, the confidence intervals for the LT_{50} values are well separated, showing faster effect of the formulation comparing to unformulated conidia.

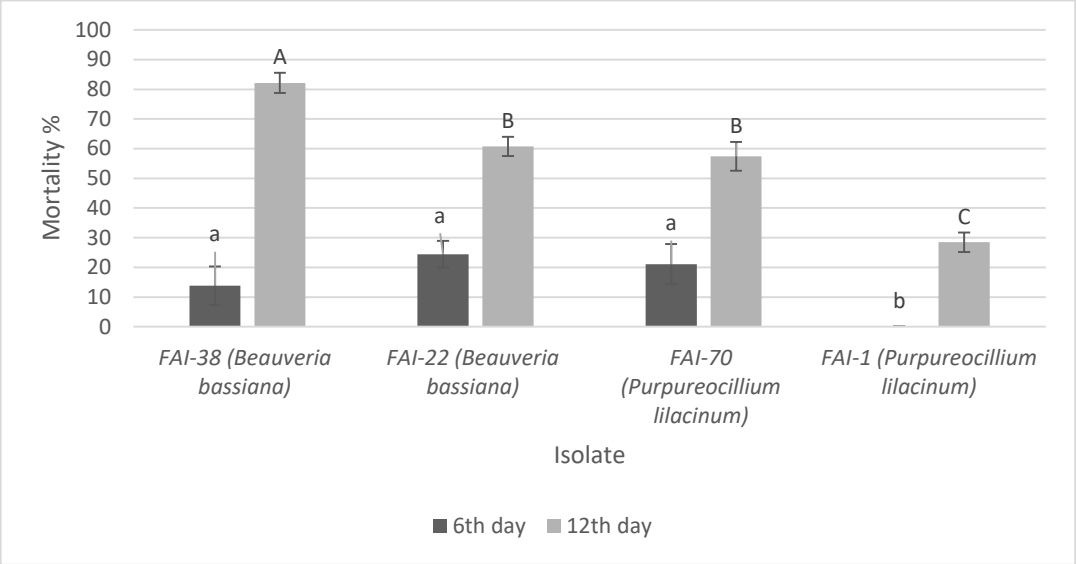

Figure 1. Corrected mortality of *Palomena prasina* nymphs after application of entomopathogenic fungi at the concentration of 1×10^8 conidia/ml (Data were subjected to Abbott's correction formula; control mortalities were 0-6,66%, one-way ANOVA was applied to the data and the differences between treatments were determined by Duncan test (n=3), error bars indicate standard errors, different letters within each time indicate statistically important differences, 6th day: F3,8=13.383 P=0.002 and 12th day: F3,8=31.374 P=0.000).

Şekil 1. Entomopatojen fungus izolatlarının 1×10^8 konidi ml^{-1} konsantrasyonda uygulandıktan sonra Palemona prasina nimflerinin düzeltilmiş ölüm oranları (Verilere Abbott'un düzeltme formülüne uygulanmıştır; kontrol ölümleri %0-6,66 arasında değişmektedir, verilere tek yönlü ANOVA uygulanmış ve uygulamalar arasındaki farklar Duncan testi ile belirlenmiştir (n=3), hata çubukları standart hataları göstermektedir, aynı gündeki farklı harfler istatistiksel olarak önemli farklılıkları göstermektedir, 6th day: F3,8=13.383 P=0.002 and 12th day: F3,8=31.374 P=0.000)

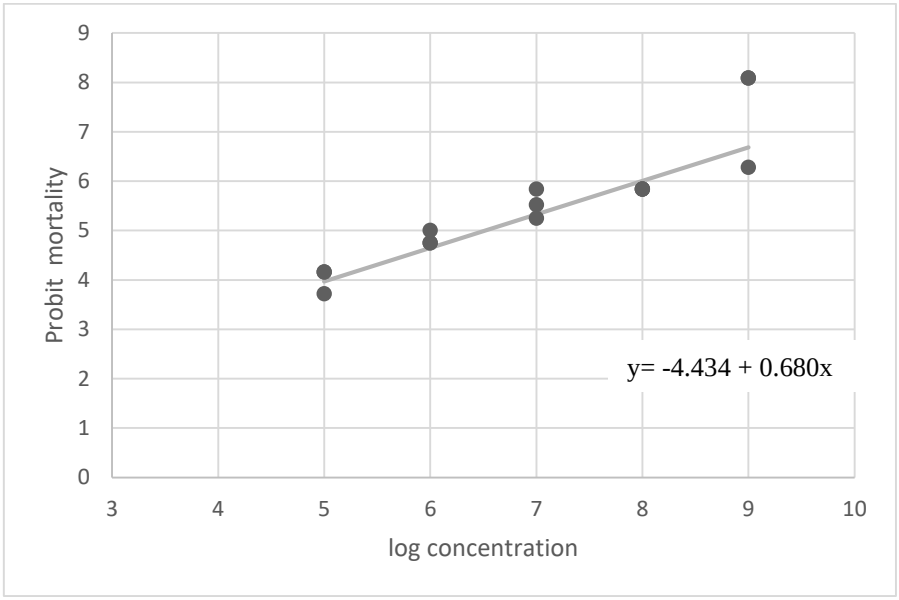

Figure 2. Concentration-dependent mortality of *Palomena prasina* nymphs 12 days after *Beauveria bassiana* FAI-38 applications

Şekil 2. Beauveria bassiana FAI-38 uygulamalarından 12 gün sonra Palomena prasina nimflerinin konsantrasyona bağlı ölümleri

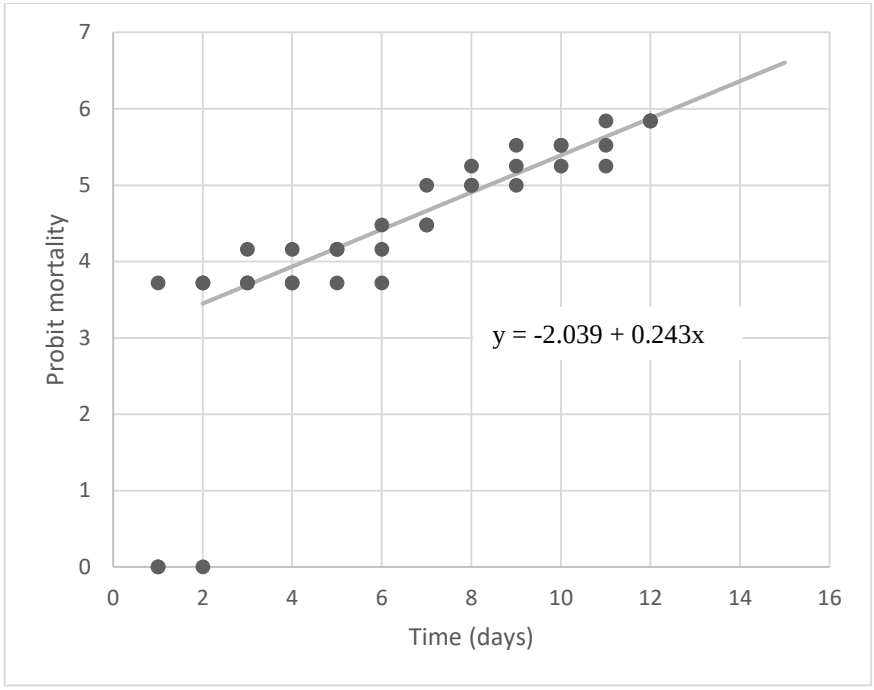

Figure 3. Time-dependent mortality of *Palomena prasina* nymphs due to *Beauveria bassiana* FAI-38 application at the concentration of 1×10^8 conidia ml^{-1}.

Şekil 3. 1×10^8 spor ml^{-1} konsantrasyonunda Beauveria bassiana FAI-38 uygulaması sonrasında Palomena prasina nimflerinin zamana bağlı ölümleri

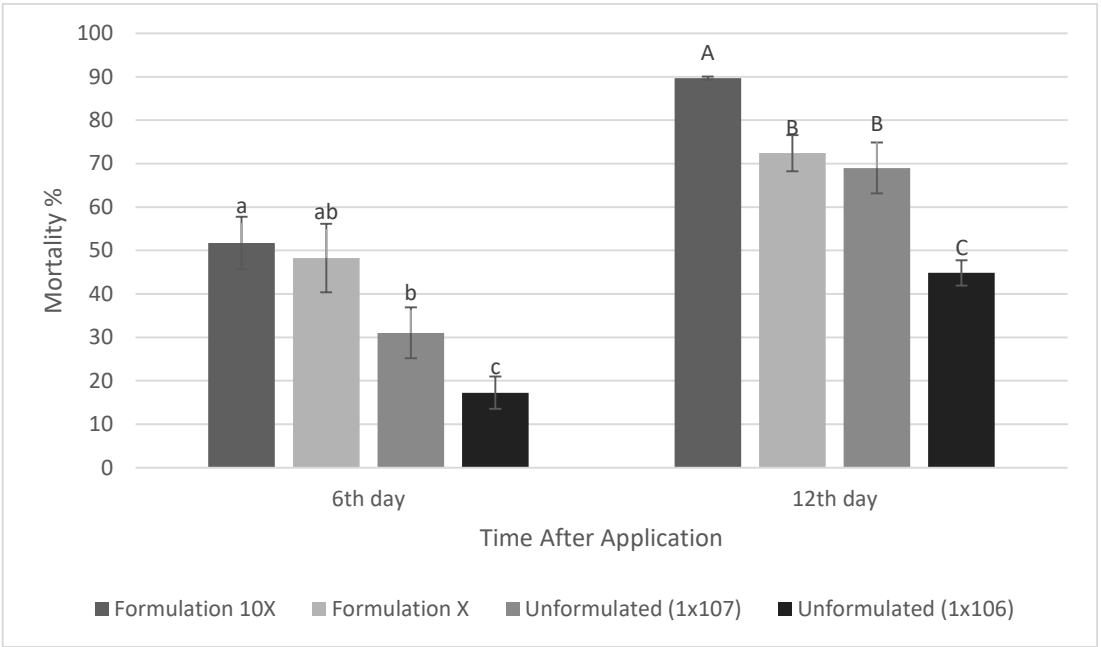

Figure 4. Mortality of *Palomena prasina* nymphs after application of WP formulation at 1X and 10X and two corresponding concentrations (1×10^6 and 1×10^7 conidia ml^{-1}) of the fungal conidia without formulating, (Data were subjected to Abbott's correction formula; error bars indicate standard errors control mortalities: 0-3.33%, n=3, different lowercase and capital letters indicate statistical differences on the 6th and 12th days respectively according to Duncan multiple comparison test 6th day: F3,8=7.094 P=0.012 and 12th day: F3,8=23.743 P=0.000)

Şekil 4. Palomena prasina nimflerinin 1X ve 10X WP formülasyonu ve konidinin formülasyonsuz karşılık gelen iki konsantrasyonu (1×10^6 ve 1×10^7 konidi ml^{-1}) uygulanmasından sonraki ölümleri. (Verilere Abbott'un düzeltme formülüne uygulanmıştır; hata çubukları standart hataları göstermektedir,kontrol ölümleri: %0-3.33, n=3, Duncan çoklu karşılaştırma testine göre farklı küçük ve büyük harfler sırasıyla 6. ve 12. günlerdeki istatistiksel farklılıkları göstermektedir, 6th day: F3,8=7.094 P=0.012 and 12th day: F3,8=23.743 P=0.000)

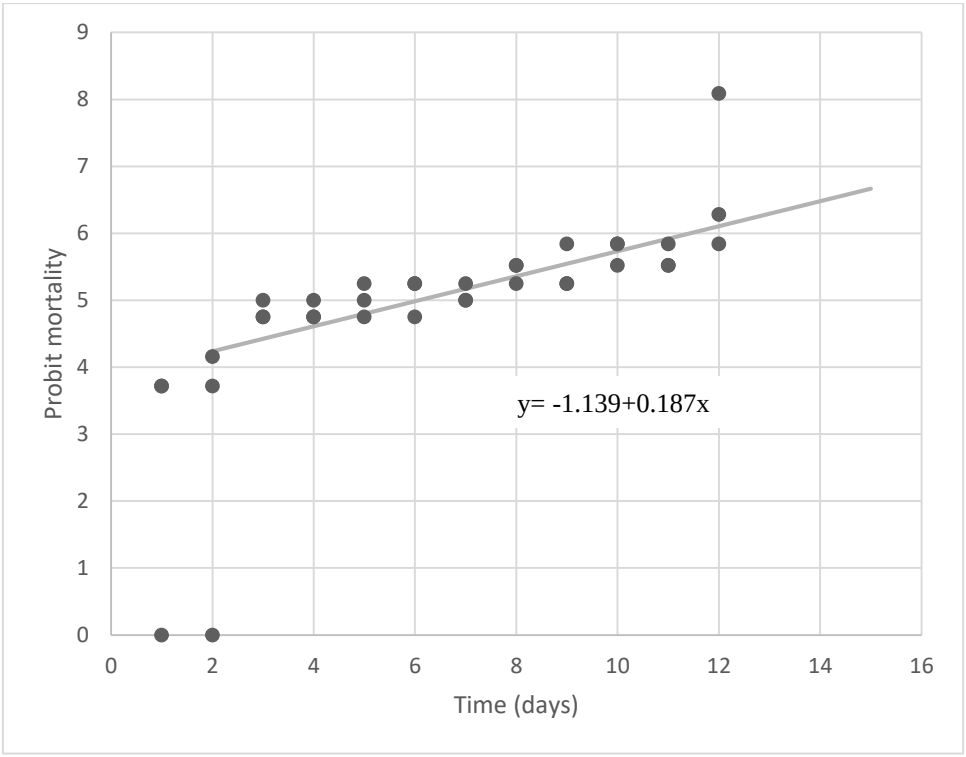

Figure 5. Time-dependent mortality of *Palomena prasina* nymphs due to applications of *Beauveria bassiana* FAI-38 WP formulation at 10X

Şekil 5. Beauveria bassiana FAI-38 WP formülasyonunun 10X uygulaması sonucu Palomena prasina nimflerinin zamana bağlı ölümleri

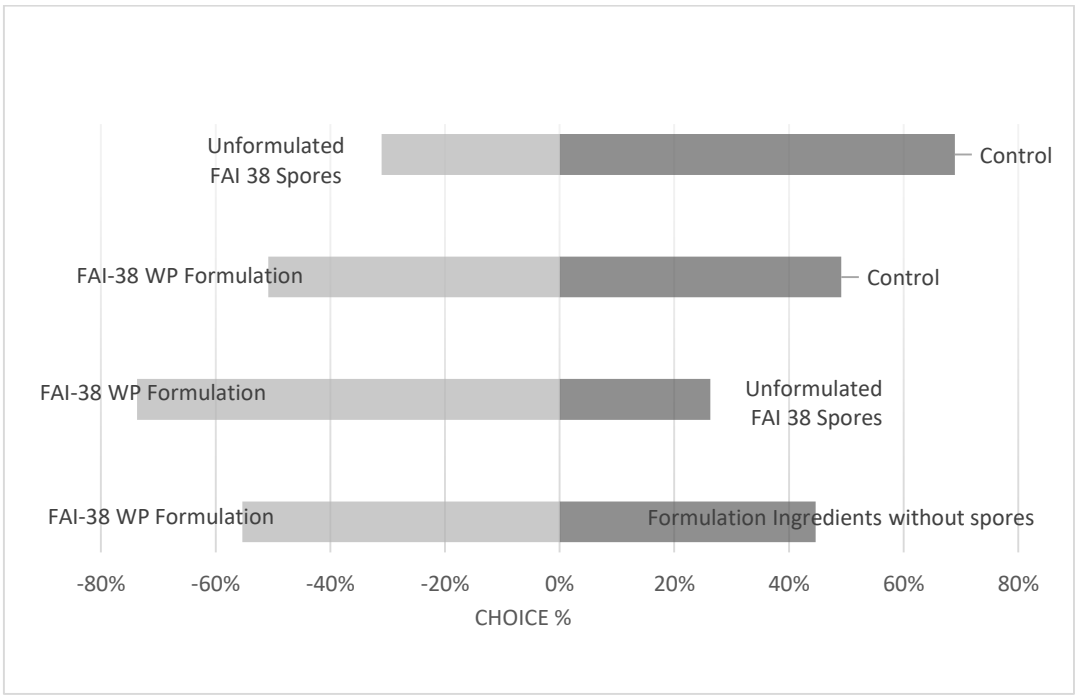

Figure 6. Results of the Y-tube olfactometer bioassays showing the behavioral response of *Palomena prasina* nymphs to the presence of *Beauveria bassiana* conidia (Formulations were used at 10X and unformulated spores at 1×10^7 spores ml^{-1} concentration)

Şekil 6. Palomena prasina nimflerinin Beauveria bassiana sporlarına karşı davranışsal tepkisini gösteren Y tüpü olfaktometre testlerinin sonuçları (Formülasyon 10X ve formüle edilmemiş spor 1×10^7 spor ml^{-1} konsantrasyonunda kullanılmıştır)

Formulation of *B. bassiana* has not been tested before on *P. prasina*; therefore, there are not any records to compare these results. However, there are few studies on formulated entomopathogenic fungi against other pest species in the family Pentatomidae. Parker et al. (2015) tested two formulations (wettable powder and emulsifiable suspension) of *B. bassiana* against the brown marmorated stink bug, *Halyomorpha halys*. They found both formulations effective at 1×10^7 conidia ml^{-1} with 67–80% and 95–100% mortality 9 and 12 days post treatment, respectively. The wettable powder formulation was concluded as slightly more efficacious. Kaolin powder formulations of *B. bassiana* and *M. anisopliae* were used against three species of stink bugs (*Nezara viridula* (L.), *Euschistus heros* (F.) and *Piezodorus guildinii* (Westwood)) by Sosa-Gomez et al (1998). *M. anisopliae* formulation was found better and the LT_{50} value was 4.3±0.2 days for *P. guildinii*, 4.6±0.2 days for *N. viridula* and 7.4 ± 0.5 days for *E. heros*. In field trials, up to 48% mortality was recorded.

Behavioral responses of *P. prasina* nymphs in Y-tube olfactometer tests are presented in Figure 6. When one of the choices was unformulated spores, significantly more nymphs preferred the alternative choice. In one test the preferred alternative was control (39 nymphs vs 19 nymphs; no choice=2 nymphs; X^2= 6.897, P=0.009), indication of avoidance from fungal spores. In another test, the alternative to unformulated spores was FAI-38 formulation, and the nymphs still showed avoidance and preferred the formulation (45 nymphs vs 14 nymphs; no choise=1 nymph X^2=16.288, P=0.000). The nymphs did not show a significant preference between FAI-38 formulation (30 nymphs) and control (29 nymphs) (no choice=1 nymhps; X^2= 0.017, P=0.8960), demonstrating that the nymphs stopped avoiding the spores when formulated. Preferences between FAI-38 formulation (26 nymphs) and formulation ingredients without spores (21 nymphs) were not significantly different either (no choice=13 nymphs; X^2= 0.532, P=0.466). Changing the places of two choices in Y-tube olfactometer in all four tests did not cause significant change in the preferences [unformulated FAI-38 spores vs control (t=0.271, P=0.849), FAI-38 formulation vs control (t=13.024, P=0.646), FAI-38 formulation vs formulation ingredients without spores (t=1.103, P=0.646), unformulated FAI-38 vs FAI-38 formulation (t=0.302, P=0.533)].

It is quite commonly reported that insects can recognize and stay away from entomopathogenic fungi (Baverstock et al., 2010; Wei et al., 2020; Avery et al., 2021; Geedi et al., 2022; Daisy, 2022). A similar reaction was evident from the results when *P. prasina* nymphs were set to choose between unformulated spores and control. However, such reaction was not seen when the choices were FAI-38 formulation and control. Similar phenomenon was reported previously. Wang & Powell (2004) demonstrated that *Reticulitermes flavipes* and *Coptotermes formosanus* (Isoptera: Rhinotermitidae) avoided *M. anisopliae* conidia but its bait formulation did not repel the insects. Fernandez-Grandon et al. (2020) showed that *Aphidius colemani* was deterred by plants treated with *M. anisopliae* but not by plants treated with a combination of *M. anisopliae* and pyrethrum.

CONCLUSION

This study shows that *Beauveria bassiana* FAI-38 provides a high level of *P. prasina* nymphal mortality comparable to those in literature, presenting the potential for further studies for developing as a control agent. Furthermore, formulating *B. bassiana* FAI-38 as wettable powder increases and accelerates the mortality with an additional advantage of removing the avoidance reaction of *P. prasina* nymphs.

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Author's Contributions

The contribution of the authors is equal.

Conflict of Interest

The author declares that there is no conflict of interest in the study.

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