Türk. entomol. derg., 2024, 48 (3): 327-342
DOI: http://dx.doi.org/10.16970/entoted.1498947

ISSN 1010-6960
E-ISSN 2536-491X

*Original article (Orijinal araştırma)*

# Environmental persistence of the conidia of native entomopathogenic fungi and their efficiency on *Ceratitis capitata* (Wiedemann, 1824) (Diptera: Tephritidae)[1]

Yerel entomopatojen fungus konidialarının çevresel kalıcılığı ve *Ceratitis capitata* (Wiedemann, 1824) (Diptera: Tephritidae) üzerindeki etkinliği

**Mehmet Sedat SEVİNÇ[2*]** **İsmail KARACA[3]**

## Abstract

This study aimed to investigate the effects of local *Beauveria bassiana* (Balsamo) Vuillemin and *Cordyceps fumosorosea* (Wize) (Hypocreales: Cordycipitaceae) isolates on *Ceratitis capitata* (Wiedemann, 1824) (Diptera: Tephritidae) prepupae, during 2020 and 2021 in the Mediterranean Region of Türkiye. In petri dishes, mortality rates varied with doses: for LD.2016 (*B. bassiana*), between 30 and 60%, for M6-4 (*B. bassiana*), 30-46%, of IFR (*C. fumosorosea*), 65-100%. Additionally, differences were detected between the prepupa and pupa periods of the larvae depending on the isolate. In sterile soil studies, the IFR caused mortality rates of 53-93%, while those for the LD.2016 isolate were 20-55%, depending on the doses. In conidial survival studies, the IFR caused mortality rates of 70-93% at 0 months and 44-60% in the 12$^{th}$ month. The LD.2016 mortality was among 37-55% at 0 months, while mortality rates decreased to 2% in the 12$^{th}$ month. The reproductive capacity of adult individuals that emerged alive after infection was negatively affected depending on time and fungal isolate. In semi-controlled field conditions, the IFR isolate had mortality rates varied between 40 and 65%, and LD.2016 mortality rates varied between 34 and 62%, depending on doses. It was observed that the fungi used in the study had potential in biological control of *C. capitata*.

**Keywords:** *Beauveria bassiana*, *Cordyceps fumosorosea*, reproductive capacity, soil surface application, sublethal effects

## Öz

Bu çalışmada, 2020-2021 yıllarında Türkiye Akdeniz Bölgesi'nde yerel *Beauveria bassiana* (Balsamo) Vuillemin ve *Cordyceps fumosorosea* (Wize) (Hypocreales: Cordycipitaceae) izolatlarının *Ceratitis capitata* (Wiedemann, 1824) (Diptera: Tephritidae) prepupaları üzerindeki etkilerinin araştırılması amaçlanmıştır. Petri kaplarında ölüm oranları dozlara göre; LD.2016 (*B. bassiana*) için %30-60 arasında, M6-4 (*B. bassiana*) için %30-46 arasında, IFR (*C. fumosorosea*) için %65-100 arasında değişmiştir. Ayrıca, izolatlara bağlı olarak larvaların prepupa ve pupa dönemleri arasında farklılıklar tespit edilmiştir. Steril toprak çalışmalarında IFR %53-93 arasında mortaliteye neden olurken, LD.2016 izolatı için bu oran doza bağlı olarak %20-55 olmuştur. Konidial hayatta kalma çalışmalarında IFR 0. ayda %70-93, 12. ayda ise %44-60 arasında mortaliteye neden olmuştur. LD.2016 mortalitesi 0. ayda %37-55 arasında iken, 12. ayda mortalite oranları %2'ye düşmüştür. Enfeksiyondan sonra canlı çıkan ergin bireylerin üreme kapasitesi zamana ve fungal izolata bağlı olarak olumsuz etkilenmiştir. Yarı kontrollü arazi koşullarında IFR izolatında mortalite oranları doza bağlı olarak %40-65 arasında, LD.2016 mortalite oranları ise %34-62 arasında değişmiştir. Çalışmada kullanılan fungusların *C. capitata*'nın biyolojik mücadelesinde potansiyel taşıdığı görülmüştür.

**Anahtar sözcükler:** *Beauveria bassiana*, *Cordyceps fumosorosea*, üreme kapasitesi, toprak yüzey uygulaması, ölümcül olmayan etkiler

[1] This study was supported by General Directorate of Agricultural Research and Policies, Türkiye, Grant Project No: TAGEM/BSAD/A/21/A2/P5/2404. A part of this study, were presented as an oral presentation at the 8th International Entomopathogens and Microbial Control Congress held in Antalya-Turkey on 6-8 October 2022, and the summary was included in the congress summary book.
[2] Fruit Research Institute, Plant Health Department, 32500, Eğirdir, Isparta, Türkiye
[3] Isparta University of Applied Sciences, Faculty of Agriculture, Department of Plant Health, 32200, Isparta, Türkiye
* Corresponding author (Sorumlu yazar) e-mail: mehmetsedat.sevinc@tarimorman.gov.tr
Received (Alınış): 12.06.2024 Accepted (Kabul ediliş): 07.10.2024 Published Online (Çevrimiçi Yayın Tarihi): 08.10.2024

## Introduction

Terrestrial ecosystems are characterized by a wide variety of species and the diversity of interactions between these species (Tscharntke & Hawking, 2000). The fungi kingdom, which is referred to as the 'communication networks of the forest' in the book 'Entangled Life' due to their biology (Sheldrake, 2020), contains an important group called 'entomopathogenic fungi (EPF)' within the ecosystem. The majority of EPF are pathogenic to insects in nature (Shah & Pell, 2003; Scholte et al., 2004; Vega et al., 2009; Dash et al., 2018) and they exhibit a high degree of efficiency in infecting their hosts to manage the pest population (Ortiz-Urquiza & Keyhani, 2013; Vidal & Jaber; 2015; Lu & St Leger, 2016). EPF have attracted attention in multitrophic studies in recent years because of their many positive aspects such as contribution to plant nutrition and soil improvement, support to the plant kingdom in the competitive/antagonistic relationships between plant pathogenic fungi, and lack of toxic effects (O'Callaghan et al., 2022; Quesada-Moraga et al., 2022). EPF not only kill arthropods in nature, but also contribute to natural regulation by changing their reproductive abilities (Castrillo et al., 2000; Quesada-Moraga et al., 2006; Ullah & Lim, 2017) and their spores and toxins have been used in biological control for many years (Villaseñor et al., 2019).

*Beauveria bassiana* (Balsamo) Vuillemin (Hypocreales: Cordycipitaceae) has been the most studied entomopathogenic fungus species as a biological control agent for many years after it was first detected on silkworms (Muscardine diseases) (Feng et al., 1994; Zimmermann, 2007). *Beauveria bassiana* is very common in soil in agricultural ecosystems. It has a wide host range and is widespread throughout the world (Roberts & St. Leger, 2004; Rehner & Buckley, 2005). While *Cordyceps fumosorosea* (Wize) (formerly *Isaria fumosorosea* or *Paecilomyces fumosoroseus*) (Hypocreales: Cordycipitaceae), which is in the same family with *B. bassiana*, was not mentioned among the important species in the past (Zimmermann, 2008). It has been included in the list as an important microbial control agent due to its effectiveness in recent years (Zimmermann, 2008; Villaseñor et al., 2019). Commercial preparations of both species are used as biological control agents nowadays.

Entomopathogenic fungi can be found in every layer of the ecosystem (Behie et al., 2015). The fungi are shielded from UV radiation and are able to withstand biotic and abiotic stresses thanks to the protective qualities of the soil environment, which prolongs their viability (Samson et al., 1988; Keller & Zimmermann, 1989). In general terms, soil is the natural reservoir of entomopathogens, where bacteria and fungi can live as saprophytes and can colonize insect cadaver parts and organic materials (Solter et al., 2017). This situation supports the idea that applying fungal propagules to the soil surface may be a good strategy in the biological control of subsoil pests that are difficult to control and harmful insects that spend any biological stage in the soil (Jackson et al., 2000). The persistence of fungal spores in the environment is important for the success of the management. Conidial environmental persistence is essential for the spread of fungi in ecosystems and also serves as a determinant for the biocontrol effectiveness of EPF during integrated pest management (Ding et al., 2023).

While research continues on the possibilities of management of many pests with EPF; researchers, in addition to chemical control studies (Yeşilırmak et al., 2024), have reported that EPF has a lethal effect on different biological stages of an important quarantine pest, *Ceratitis capitata* (Wiedemann, 1824) (Diptera: Tephritidae), which spends its pupation period in the soil. In addition to the studies focused on the pupa and adult stages of *C. capitata* (Castrillo et al., 2000; Dimbi et al., 2003; Quesada-Moraga et al., 2006; Ekesi et al., 2010; Beris et al., 2013; Qazzaz et al., 2015; Chergui et al., 2020; Gava et al., 2020; Soliman et al., 2020; Hallouti et al., 2021), in some other articles it was mentioned that 3$^{rd}$ larval instar was sensitive to *B. bassiana* (Chergui et al., 2020; Gava et al., 2020; Soliman et al., 2020). It was also determined that the fungus changed their flight activities, feeding and sexual behavior, (Dimbi et al., 2009; Bernardo & Singer, 2017), and decreased fertility (Quesada-Moraga et al., 2006) when it was applied to adult individuals. It shows the versatile effects of EPF in the biological control of *C. capitata*. Although melanization and sclerotization in pupae pose major challenges to infection by entomopathogens, larvae and pupae of *C. capitata*

are subject to EPF infection in soil (Dias et al., 2018). The biological period delay ultimately contributes to reducing the population and is an important support for the success of the pest management. For this reason, microbial pathogens are evaluated as an alternative to chemical insecticides due to their high pathogenicity and the fact that they cause delays in biological periods. Therefore, mortality alone should not be used to evaluate effective control (Hussain et al., 2009).

The efficiency rate of each EPF isolate may differ from each other. The sensitivity rates of late-stage larvae may vary compared to pupae and adults at the first moment when *C. capitata* comes into contact with soil, which is an important fungal reserve. However, there is no practical soil applications using EPF in the management of *C. capitata*. The ability of fungi to persist in soil affects the persistence of fungal pathogenicity and this is important for sustainable pest management. For this reason, it is necessary to determine the level of response of pests that visit the application area at different times by the EPF propagules in the environment (Gava et al., 2020).

The present study was initiated at the Department of Plant Health in Fruit Research Institute, Isparta, Türkiye, in order to investigate the influence of native isolates *Beauveria bassiana* and *Cordyceps fumosorosea*, on 3rd instar larvae of *C. capitata* by determining indices of growth times and mortality rates. We investigated whether fungal infection with different densities of conidiospores affect the duration of prepupa and pupation period and survival rate of third instar larvae in the *C. capitata*. Additionally, the objective of this work was not only to select virulent EPF isolates among local strains to control *C. capitata* during the cryptic life cycle in soil, but also to investigate the effects of exposure to EPF at different times, on postemergence survival of adults and oviposition of them under controlled conditions. In addition, lethal effect of EPF under field (semi-controlled) conditions was studied for two years.

## Materials and Methods

### *Ceratitis capitata* culture

*Ceratitis capitata* culture was maintained in climatized rooms with 25±1ºC temperature, 65±10% humidity and 16: 8 hours light and dark conditions in the Fruit Research Institute. Golden delicious apple fruits were used for the mass rearing of *C. capitata*.

### Entomopathogenic fungi culture

*Beauveria bassiana* (Bals.) Vuill (BMAUM M6-4 and BMAUM LD.2016) isolates, which were found to be highly pathogenic, as a result of the preliminary pathogenicity test, were provided from the investigation of Baydar et al. (2016). *Cordyceps fumosorasea* (IFR) isolate were also obtained from stock culture of Center of Biological Management Research and Apply (BMAUM) at University of Applied Science. Entomopathogenic fungus isolates used in the experiment were cultured on Potato Dextrose Agar (PDA) medium in the Phytopathology laboratory using a sterile cabinet and an incubator and the isolates stored-80ºC conditions.

### Preparation of fungal suspensions

Conidiospores were extracted using a scraper from the 10 to 14 days old fungal cultures in Petri dishes with PDA incubated at 25ºC. Conidiospores of each isolate were suspended in 50 mL sterile distilled water amended with 0.01 mL Tween 80. In order to achieve densities of $1x10^7$, $1x10^6$, $1x10^5$, and $1x10^4$ conidia/mL, the initial suspension with a density of $1x10^8$ conidiospore/mL was diluted by one-tenth for each new concentration. Spore counts were checked by using a Thoma hemacytometer.

### Petri dishes bioassay

The experiment was set up in 9 cm diameter petri dishes to assess the percentage of lethality and duration of lethality of *B. bassiana* (LD.2016 & M6-4) and *C. fumosorosea* (IFR) on 3rd instar larvae of *C. capitata*.

A double layer of filter paper was placed on the inner surface of each petri dish, leaving no gap at the bottom. 2 mL of the suspensions with densities of $1x10^8$, $1x10^7$, $1x10^6$, $1x10^5$, $1x10^4$ conidia/mL were applied on filter papers. For control group, the same volume of distilled water with 0.01 mL Tween 80 was used. All treatments were applied with a hand spray from a 20-cm distance. The prepared insect diet (*Cydia pomonella* diet-Southland Products® Arkansas, USA), a piece of roughly 1 $cm^3$, was placed in the center of the filter paper to prevent the larvae from dying of malnutrition. The last instar larvae, which emerged from the fruit to enter the pupa period, were placed on an insect diet. The treatment consisted of 5 replications for each density, with 20 late-stage larvae were used in each replication. The treatment was conducted in climatic chambers at 25ºC, 65% R.H, and 16: 8 (L: D). As a result of determining the mortality rates in bioassay studies larval and pupal development periods of surviving individuals were monitored and recorded daily in the same conditions.

**Conidial persistence studies**

The soil used in the planting of saplings in Eğirdir Fruit Research Institute was sterilized in an oven at 105ºC for 48 hours in case it contained live microorganisms before the first treatment. The lids of 10x12x5 cm -sized plastic boxes were cut and covered with tulle, and 50 g of sterile soil was placed into these plastic containers. As a result of petri bioassay, the most effective two EPF isolates' $1x10^8$, $1x10^7$, $1x10^6$ conidia/mL of the spore suspensions were applied to soil with 5 replicates and 1$cm^3$ insect diet was placed at the center of the plastic container on the soil. The last instar larvae, which were emerging from the fruit for enter the pupa period, were placed on the insect diet in the trial container. 20 third instar larvae were used for each dose and replicates. In all replicates, containers were checked until all individuals reached adult stage and the data were recorded.

This experiment was named '$0^{th}$ month' after application of EPF spores to the soil surface. EPF was not applied to these plastic containers again for subsequent trials for determination the conidial persistence, but 2 mL of sterile water was sprayed to provide moisture before the treatments. The experiment was repeated at $3^{rd}$, $6^{th}$, $9^{th}$ and $12^{th}$ months. This was done by adding 20 late-stage larvae to each replication container at 3-month intervals. The containers were checked daily until all individuals reached the adult stage and data were recorded to determine conidial persistence. The treatment was conducted in climatic chambers at 25ºC, 65% R.H, and 16: 8 (L: D).

**Detecting adult infertility**

Individuals that became adults in the plastic containers used in the sterile soil application experiments were transferred to different insectariums for each application, to examine their reproduction. In the experiment, number of adult emergence and dates were recorded starting from the emergence day of the first individual. Golden delicious fruits were given to the individuals in the population every day so that they could lay eggs, and those given the previous day were taken from the insectarium. Fruits where eggs were thought to have been laid were placed into double layer insect netted insectarium of different sizes which intertwined to prevent contamination from flies outside (if present) to monitor larval development in climate chambers.

**Field trials (semi-controlled conditions)**

During field trials, apertures were cut into the lids of 2-liter plastic sealed containers with a utility knife and closed with tulle. The soil taken from the Antalya region was put in the plastic containers up to a height of 4-5 cm without any application. The suspensions of IFR and LD.2016 isolates with $1x10^8$, $1x10^7$, $1x10^6$ conidia/mL densities prepared in the laboratory the day before and 6 ml of these suspensions were applied to the soil surface from a distance of 20 cm with the help of a hand spray for each container. 1 $cm^3$ insect diet was placed in the center of the boxes and 20 late-stage larvae were added to the containers. After the emergence of adults from the soil, the soil used in the experiments was examined and the numbers of empty pupal capsules and dead *C. capitata* adults were evaluated to obtain the mortality rates. The experiment was conducted with 5 replications for each concentration and control group in field conditions of Antalya province.

### Statistical analyses

The obtained data were used to determine the actual mortality values (% effect) by applying the Abbott formula (Abbott, 1925);

$$\text{Corrected mortality rate (\%)} = \frac{\text{Mortality rate of treated larvae} - \text{Mortality rate of control}}{100\text{x Mortality rate of control}}$$

The mortality rates were first subjected to Levene's homogeneity test, Shapiro-Wilk's normality test was used and then one-way analysis of variance (One-Way ANOVA) was applied. Then, Tukey multiple comparison test (Tukey, 1949) was performed to determine the source of differences. Differences were considered significant at $p<0.05$. Statistical analyses were performed with the help of IBM SPSS® Statistics (Version 23.0, 2015, IBM SPSS, Armonk, New York, USA) (IBM SPSS, 2015).

## Results and Discussion

### Petri dishes bioassay result

Mortality data obtained at the end of the trial are shown in Figure 1. Although doses of $1x10^8$ conidia/mL and $1x10^7$ conidia/mL of IFR caused 100 percent mortality, the mortality rates in subsequent doses were 73%, 65% and 65%, respectively. The mortality of the doses of the LD.2016 isolate was 60%, 53%, 46%, 40% and 30%, respectively. The mortality rates of the M6-4 isolate according to the doses were 46%, 40%, 40%, 30% and 30% from highest to lowest. In the control group, all individuals remained alive at the end of the experiment.

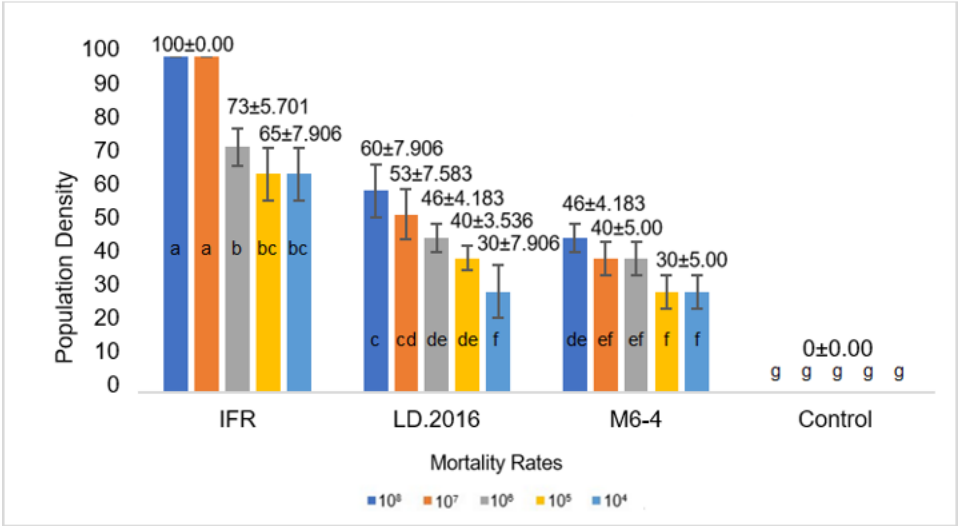


Figure 1. Mortality rates (±std. dev.) of *Cordyceps fumosorosea* (IFR) and *Beauveria bassiana* (LD.2016 & M6-4) isolates on 3rd larva of *Ceratitis capitata*. *According to Tukey's multiple comparison test, different letters are statistically different ($p<0.05$). (F: 110,493; df:15,64; p<000.1).

The petri dishes bioassay was designed to be studied in petri dishes to clearance whether the larval period in which the 3rd instar larvae that spend their pupa period in the soil, are most sensitive to pathogens, is delayed. Because the soil wasn't suitable for visible the development stages, however, it is easier to observe the biological process in petri dishes. Chergui et al. (2020), reported that petri dishes bioassays had higher mortality compared to the sterile soil experiment with their study of *C. capitata* larvae-EPF interaction bioassays both petri dishes and sterile soil. Previous sterile soil studies reported high mortality rates on *C. capitata*. The findings were consistent with the findings of other research (Ekesi et al., 2005; Quesada-Moraga et al., 2006; Chergui et al., 2020) that found that the larval stage of *C. capitata* was the most susceptible to EPF.

The mortality times of larvae in petri dishes and the effects of the isolates on the pupal stages of surviving individuals were determined. In the application of $1x10^8$ conidia/mL dose of IFR isolate, larval deaths due to infection occurred from the 3rd day to the 7th day after the treatment. On the 3rd day, 25 individuals died totally and reducing the population to 75 individuals; on the 5th day of IFR application, the population were 5 individuals and on the 7th day, all individuals died.

After the treatment of $1x10^8$ conidia/mL dose of LD.2016 isolate, larval deaths occurred from the 3rd day to the 13th day after the treatment. However, all of the individuals (40 individuals) that will emerge as adults on the 13th day have entered the pupation period (Figure 2). These individuals completed their pupation period in 2 days, becoming 27 pupae on the 12th day and 40 pupae on the 13th day. The first adult emergence was seen on the 25th day of the experiment, and emergence continued until the 31st day (Figure 3). In the application of $1x10^8$ conidia/mL dose of M6-4 isolate, infection-related deaths started on the 11th day and continued until the 17th day, and 54 individuals entered the pupal stage (Figure 2). The pupation process, which started on the 14th day of the experiment, ended on the 17th day, taking a total of 3 days. The pupal stage lasted until the 29th day, while adult emergence ended on the 35th day of the experiment (Figure 3).

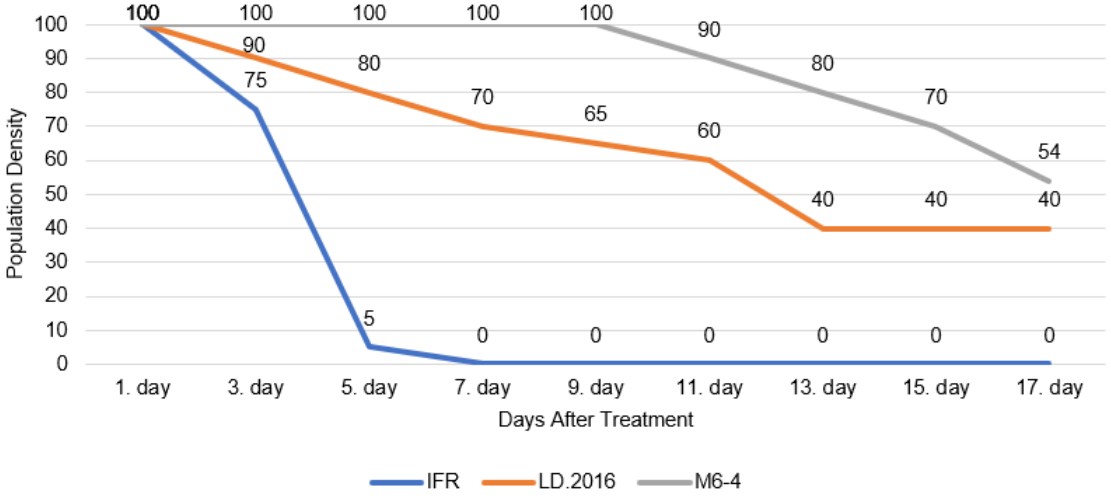


Figure 2. Time-dependent change of the *Ceratitis capitata* population applied with $1x10^8$ conidia/mL.

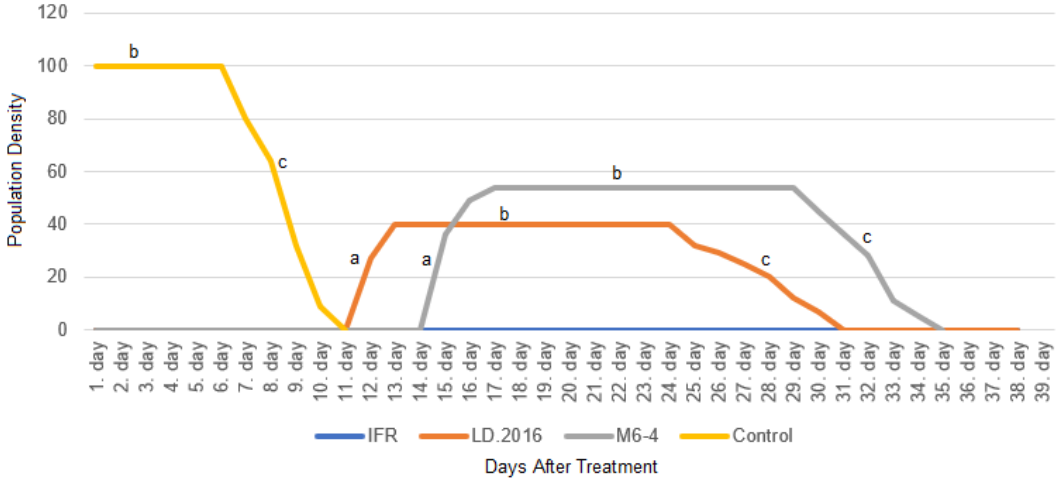


Figure 3. Time-dependent population fluctuation of the pupal stages of individuals that emerged as adults after $1x10^8$ conidia/mL applications of all isolates. (a: entry into the pupal stage, b: the entire population is in the pupal stage, c: adult *Ceratitis capitata* emergence).

Individuals exposed to the IFR dose of $10^7$ conidia/mL died within 7 days, similar to the dose of $1x10^8$ conidia/mL. After the application of $10^7$ conidia/mL dose of LD.2016 isolate, larval deaths due to infection occurred from the 3rd day to the 17th day. However, on the 17th day, all of the individuals (47 individuals) that will emerge as adults entered the pupal period (Figure 4). These individuals completed their pupation period from in 1 day. The first adult emergence was seen on the 23rd day of the treatment, and adult emergence continued until the 29th day (Figure 5). In the M6-4 isolate, infection-related deaths started on the 9th day and continued until the 17th day, and 60 individuals entered the pupal stage (Figure 4). The pupation process, which started on the 16th day of the experiment, ended on the 17th day, taking a total of 1 day. The pupal stage lasted until the 29th day and adult emergence ended on the 35th day, which took 6 days in total (Figure 5).

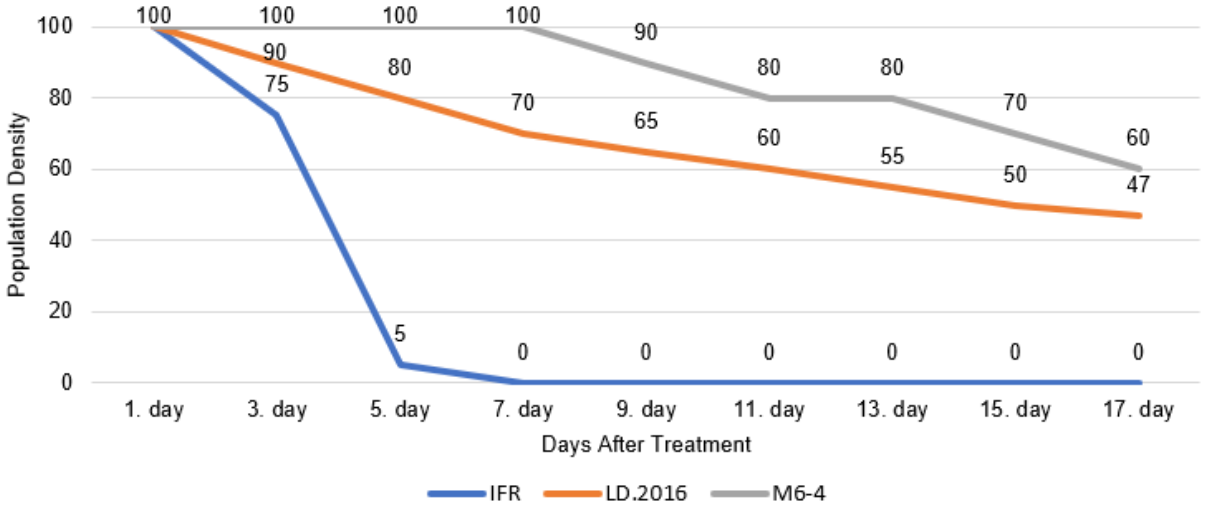


Figure 4. Time-dependent change of *Ceratitis capitata* population applied with $1x10^7$ conidia/mL.

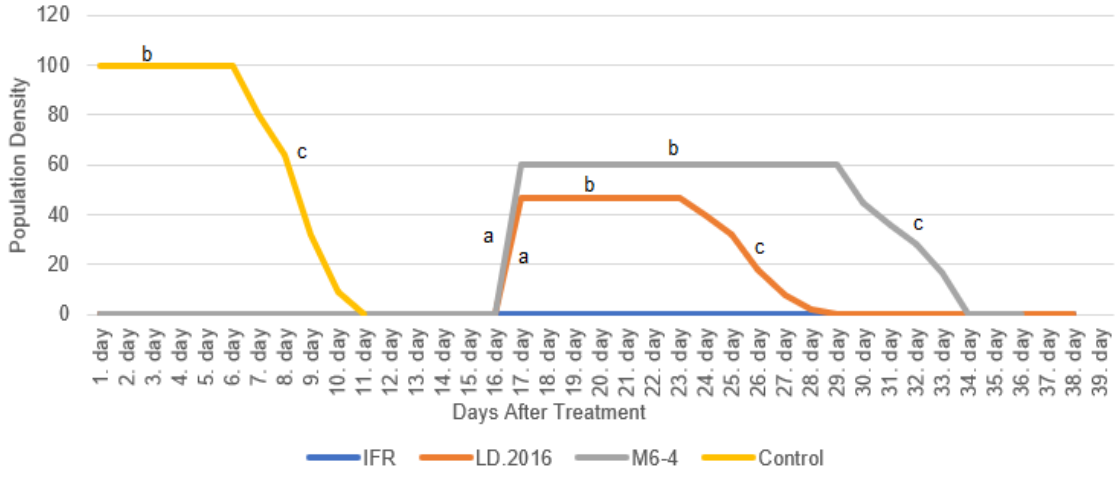


Figure 5. Time-dependent population fluctuation of the pupal stages of individuals that emerged as adults after $1x10^7$ conidia/mL applications of all isolates. (a: entry into the pupal stage, b: the entire population is in the pupal stage, c: adult *Ceratitis capitata* flights).

After applying a dose of $1x10^6$ conidia/mL of the IFR isolate, larval deaths due to infection occurred from the 5th day to the 11th day (Figure 6). Pupation started on the 10th day and was completed on the 13th day with 27 individuals. Adult emergence continued from the 17th to the 20th day (Figure 7). After the application of $1x10^6$ conidia/mL dose of LD.2016 isolate, larval deaths due to infection occurred from the 11th day to the 17th day. However, all of the individuals (54 individuals) that will emerge as adults on the 17th day have

entered the pupal period (Figure 6). These individuals completed their pupation period from day 16 to day 19 in 3 days. The first adult emergence was seen on the 23rd day of the experiment, and flights continued until the 29th day (Figure 7). In the M6-4 isolate, infection-related deaths started on the 11th day and continued until the 15th day, and 60 individuals entered the pupal stage (Figure 6). The pupation process, which started on the 16th day of the experiment, ended on the 19th day, taking a total of 3 days. Adult emergence from the pupal stage, which lasted until the 24th day, ended on the 32nd day, a total of 6 days (Figure 7).

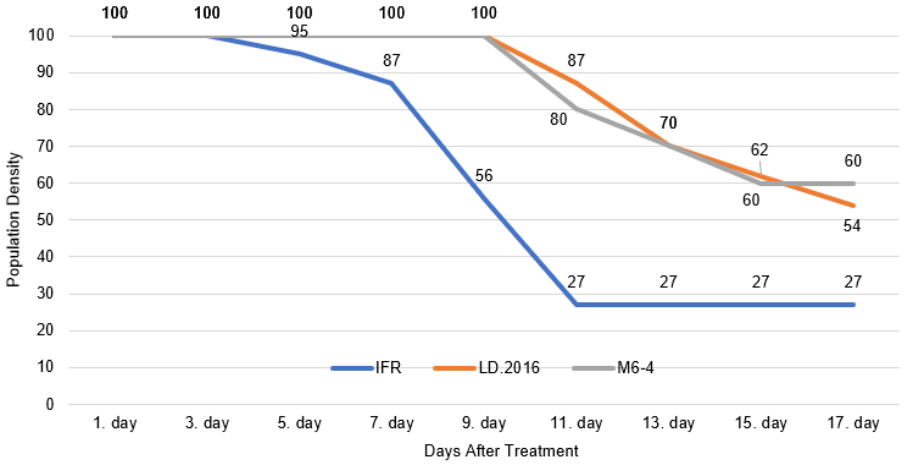


Figure 6. Time-dependent change of the *Ceratitis capitata* population applied with 1x10$^6$ conidia/mL.

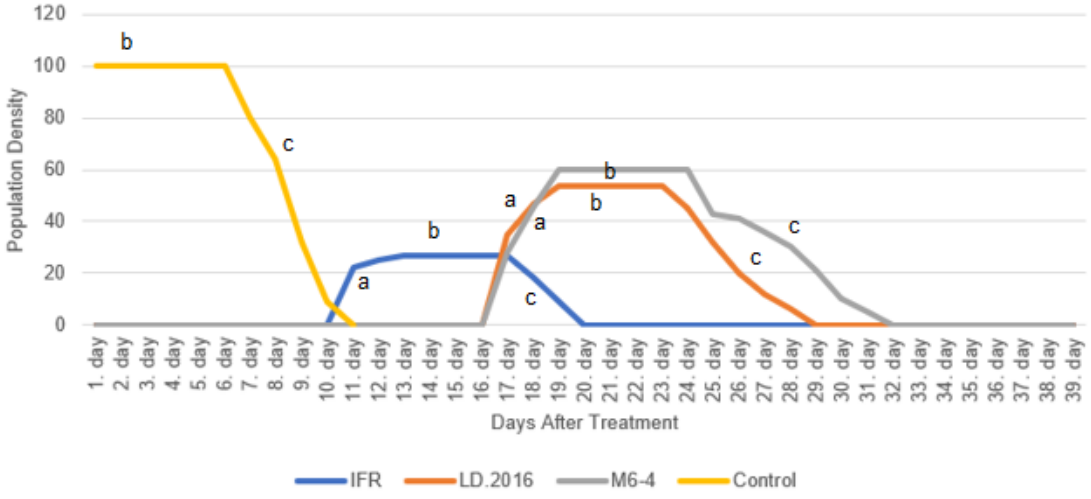


Figure 7. Time-dependent population fluctuation of the pupal stages of individuals that emerged as adults after 1x10$^6$ conidia/mL applications of all isolates. (a: entry into the pupal stage, b: the entire population is in the pupal stage, c: adult *Ceratitis capitata* flights).

In the control group of the experiment, all larvae treated with water became pupae within the first two hours of the experiment. Adult flights were recorded from the 7th to the 11th day. The pupal period was determined as 7 days for 20 individuals, 8 days for 16 individuals, 9 days for 32 individuals, 10 days for 23 individuals and 11 days for 9 individuals. In total, the pupal period of the population ended in 11 days. Sevinç & Karaca (2024) reported the differences between the developmental periods of individuals laid on the same day. According to the study, the period from egg to adult varied between 18 and 55 days. In this study, only the pupal period lasted between 7 and 11 days in the control group and these differences were thought to be possible. In addition, all individuals in the control group survived. As seen in Figures 3, 5 & 7, pupae formed on the first day of the experiment and their pupal periods and adult emergence times were completed earlier than those of individuals exposed to EPF.

**Conidial persistence bioassay results**

The lethal effects of 1x10$^8$, 1x10$^7$, 1x10$^6$ conidia/mL doses of the IFR isolate (*C. fumosorosea*) on 3$^{rd}$ instar *C. capitata* at different time intervals are shown in Figure 8. According to the Figure 8, mortality rates of larvae added to the plastic containers 0, 3, 6, 9 and 12 months after IFR 1x10$^8$ conidia/mL dose applications to the sterile soils were 93%, 65%, 60%, 60% and 60%, respectively. In soils where 1x10$^7$ conidia/mL IFR suspension was applied, larvae mortality rates were 82%, 55%, 52%, 52% and 52% at 0, 3, 6, 9 and 12 months, respectively. Larval mortality rates in soil where 1x10$^6$ conidia/mL dose was applied are as follows; 70%, 52%, 45%, 45% and 44%. When the mortality rates of all conidia densities in same months were compared, it was seen that the highest effect was obtained from 1 X10$^8$ conidi/ mL, conidia densities affected the mortality rates and statistical differences emerged. However, in the first application, different conidia densities were included in different groups.

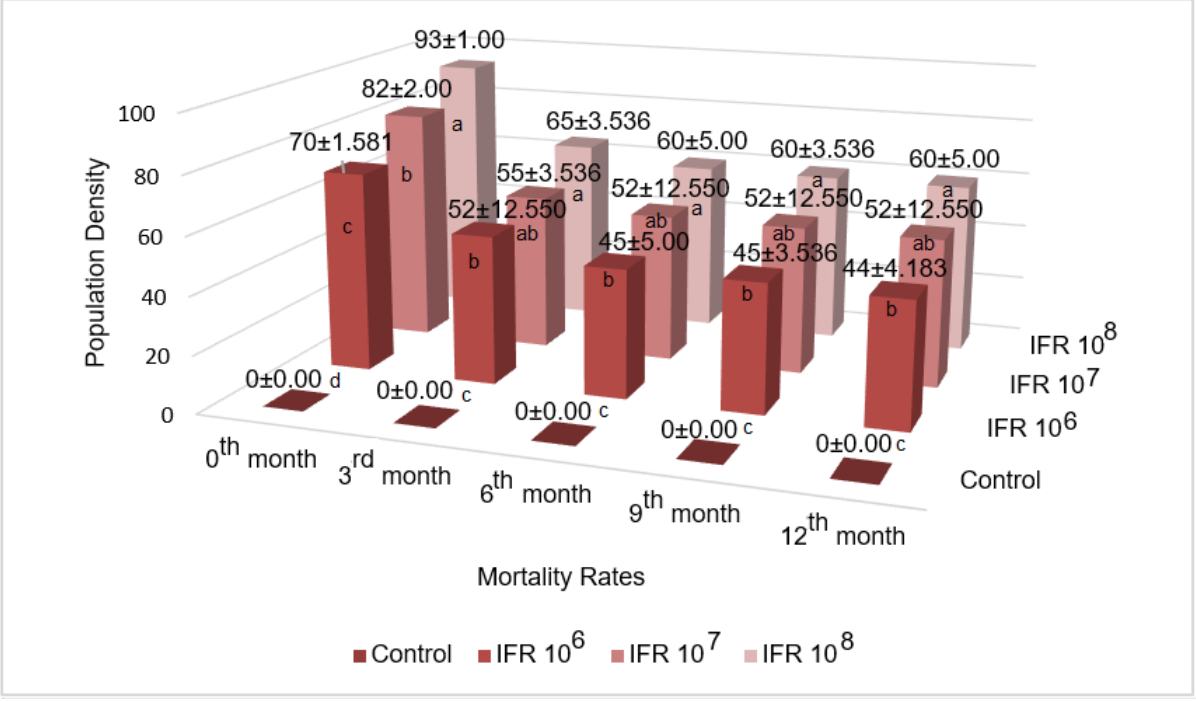


Figure 8. Percentage mortality rates (±std. deviation) of larvae released after 0, 3, 6, 9 and 12 months into sterile soil treated with 1x10$^8$ conidia/mL, 1x10$^7$ conidia/mL, 1x10$^6$ conidia/mL doses of the IFR isolate. *According to Tukey's multiple comparison test, different letters in the same month application are statistically different ($p<0.05$) (0$^{th}$ month's F value: 4681,556; 3$^{rd}$ month's F value: 93,443; 6$^{th}$ month's F value: 69,614; 9$^{th}$ month's F value: 79,151; 12$^{th}$ month's F value: 71,867). (df:3, 16; p<000.1).

The mortality rates of 1x10$^8$ conidia/mL, 1x10$^7$ conidia/mL, 1x10$^6$ conidia/mL doses of LD.2016 isolate (*Beauveria bassiana*) on 3$^{rd}$ instar of *C. capitata* in different 3-month time periods are given in Figure 9. According to Figure 9, the mortality rates of larvae left to soils treated with LD.2016 1x10$^8$ conidia/mL dose after 0, 3, 6, 9 and 12 months were; 55%, 40%, 40%, 12% and 2%, respectively. with IFR 1x10$^7$ conidia/mL dose after 0, 3, 6, 9 and 12 months were; 46%, 12%, 4%, 2% and 2%, respectively. IFR 1x10$^6$ conidia/mL dose after 0, 3, 6, 9 and 12 months were; 37%, 4%, 3%, 2% and 2%, respectively. Similarly, when the mortality data of the studies conducted in different months with different conidia densities were evaluated statistically within each trial month, the highest effect was seen in the first application of 1x10$^8$ conidia/mL of LD.2016, followed by 1x10$^7$ conidia/mL and 1x10$^8$ conidia/mL in the 3$^{rd}$ and 6$^{th}$ months, respectively. When different letters were taken into account in the relevant application month, it was determined that the 10$^8$ conidia/mL dose was statistically different compared to the other doses and the control in the 0$^{th}$, 3$^{rd}$, 6$^{th}$ and 9$^{th}$ months, but no statistical difference was observed between the 10$^8$ conidia/mL dose and the other doses and the control group in the 12$^{th}$ month.

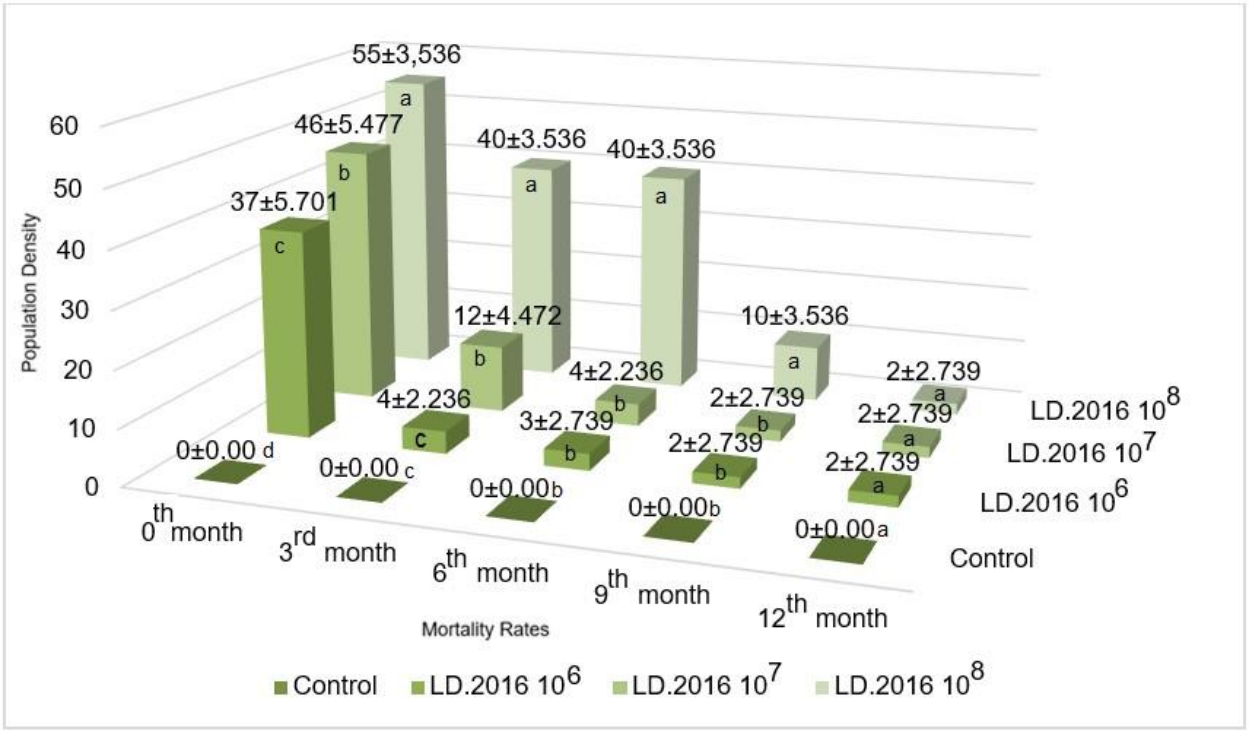


Figure 9. Percentage mortality rates (±std. deviation) of larvae released after 0, 3, 6, 9 and 12 months into sterile soil treated with $1x10^8$ conidia/mL, $1x10^7$ conidia/mL, $1x10^6$ conidia/mL doses of the LD.2016 isolate. *According to Tukey's multiple comparison test, different letters in the same month application are statistically different ($p<0.05$) ($0^{th}$ month's F value: 155,467; $3^{rd}$ month's F value: 173,511; $6^{th}$ month's F value: 286,067; $9^{th}$ month's F value: 14,303; $12^{th}$ month's F value: 0,889). (df:3, 16; p<000.1).

**Detecting adult infertility**

The time-dependent population fluctuation of individuals reaching adulthood from the first EPF application ($0^{th}$ month) is shown in Figures 10 & 11. Adult flight was detected in individuals in the control group earlier than in the EPF-contaminated groups, and a difference was observed between the isolates. There was variability in the time it took for individuals in each replication to reach adulthood. On the first day, 38 individuals from the control group were transferred to the rearing insectarium. After 13 days, the population reached 98 individuals. It remained at its peak until the $32^{nd}$ day, and while the $38^{th}$ day continued with 95 individuals, the experiment was terminated when there were no individuals left in the other experimental characters.

At the IFR dose of $1x10^8$ conidia/mL, the first adult flight started with 5 individuals on the $7^{th}$ day, reached its peak with 7 individuals on the $8^{th}$ day, remained at the peak until the 10th day, and then the population was completely exhausted on the $19^{th}$ day. At IFR $1x10^7$ conidia/mL, the first adult flight started on the $4^{th}$ day with 10 individuals, reached its peak on the $6^{th}$ day with 18 individuals, and the population was exhausted by 20 days At IFR $1x10^6$ conidia/mL, the first adult flight started on the $3^{rd}$ day with 6 individuals, reached its peak on the $7^{th}$ day with 30 individuals, and the population became extinct on the $20^{th}$ day. At the IFR dose of $1x10^5$ conidia/mL, the first adult flight started with 12 individuals on the $2^{nd}$ day, reached its peak with 38 individuals on the $5^{th}$ day, remained at the peak until the $7^{th}$ day, and then the population was completely exhausted on the $26^{th}$ day. At the IFR dose of $1x10^4$ conidia/mL, the first adult flight started with 18 individuals on the $2^{nd}$ day, reached its peak with 47 individuals on the $8^{th}$ day, and the population was completely exhausted on the $34^{th}$ day (Figure 10).

According to Figure 11, the first adult flight of the LD.2016 isolate at a dose of $1x10^8$ conidia/mL started with 5 individuals on the $11^{th}$ day, reached its peak with 45 individuals on the $21^{st}$ day, and the

population was completely exhausted on the 26th day. At the dose of LD.2016 1x10$^7$ conidia/mL, the first adult flight started on the 10th day with 22 individuals, reached its peak on the 16th day with 54 individuals, and the population was exhausted by 23 days. At a dose of 1x10$^6$ conidia/mL, the first adult flight started on the 5th day with 26 individuals, reached its peak on the 12th day with 63 individuals, and the population became extinct on the 31st day. At a dose of 1x10$^5$ conidia/mL, the first adult flight started with 27 individuals on the 4th day, reached its peak with 35 individuals on the 10th day, and the population was completely exhausted on the 30th day. At a dose of 1x10$^4$ conidia/mL, the first adult flight started with 25 individuals on the 1st day, reached its peak with 80 individuals on the 14th day, and the population was completely exhausted on the 38th day (Figure 11).

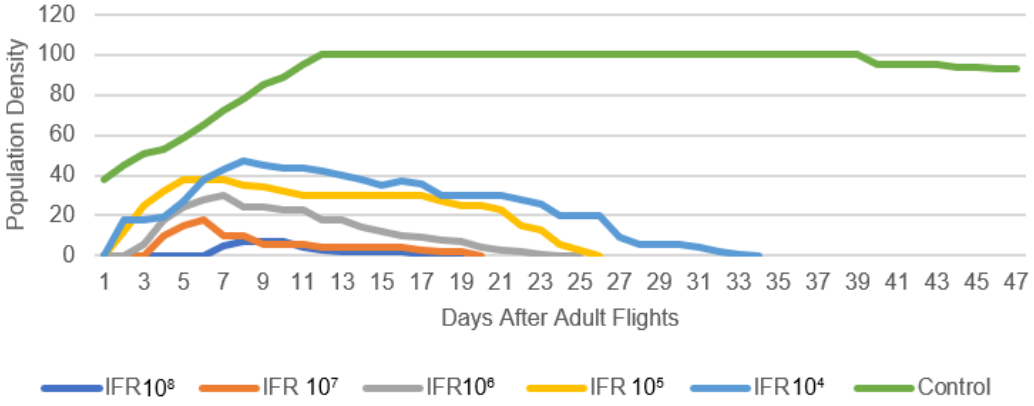


Figure 10. Population fluctuation of adult individuals emerging alive as a result of application of different IFR doses.

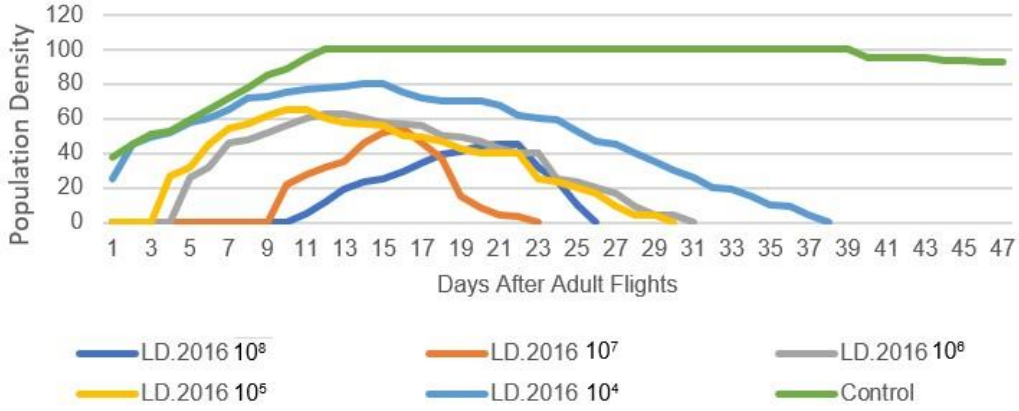


Figure 11. Population fluctuation of adult individuals emerging alive as a result of application of different LD.2016 doses.

The reproductive abilities of individuals were determined by checking the larval development in the Golden delicious apple fruit. The results are shown as present or absent in Table 1. As seen in Table 1, no larval development was observed at the IFR dose of 1x10$^8$ conidia/mL. In the remaining doses, larval development was observed, and adults were obtained. No larvae or adult development was observed at the doses of 1x10$^8$ conidia/mL, 1x10$^7$ conidia/mL and 1x10$^6$ conidia/mL of LD.2016. At other doses, there was larval development and adults were obtained. Additionally, individuals in groups with no larval development were observed to exhibit passive behavior and die before mating after a while in the 0th month. In the 12th month, in the adult individuals developed from the larvae left to the 1x10$^8$ conidia/mL dose application trial of the IFR isolate, no fertility was observed, while larval development was observed in all other trial characters.

Table 1. Larval development in apples given to individuals that survived after EPF application

| EPF isolates | $1x10^8$ | $1x10^7$ | $1x10^6$ | $1x10^5$ | $1x10^4$ |
|---|---|---|---|---|---|
| IFR (0th Month) | Absent | Present | Present | Present | Present |
| IFR (12th Month) | Absent | Present | Present | Present | Present |
| LD.2016 (0th Month) | Absent | Absent | Absent | Present | Present |
| LD. 2016 (12th Month) | Present | Present | Present | Present | Present |

**Field trials result**

The mortality rates of IFR and LD.2016 isolate doses on *C. capitata* in the field studies conducted in Antalya province in July-August 2020 and 2021 are shown in Figure 5. *C. capitata* larvae that died in soil experiments disintegrated and disappeared in a very short time. For this reason, counts were made by comparing emerging adult individuals and empty pupal capsules. In 2020, the mortality rates of IFR and LD.2016 isolates at a dose of $1x10^8$ conidia/mL were 65% and 62%, and the mortality rates at a dose of $1x10^7$ conidia/mL were 60%, 55% and 42% and 35% at a dose of $1x10^6$ conidia/mL, respectively. In 2021, the mortality rates of IFR and LD.2016 isolates at a dose of $1x10^8$ conidia/mL were 60% and 60%, and the mortality rates at a dose of $1x10^7$ conidia/mL were 56%, 55%, and the mortality rates at a dose of $1x10^7$ conidia/mL were 40% and 34%, respectively. In the control, the number of empty pupal capsules and dead adults was same in all replicates. The mortality rate in the control group was 0%. While IFR $1x10^8$ conidia/mL had the highest effect in 2020, in 2021 the mortality rates of the $1x10^8$ and $1x10^7$ conidia/mL doses of both isolates were statistically in the same group. In both years, all conidia concentrations were found to be different compared to the control group (Figure 12).

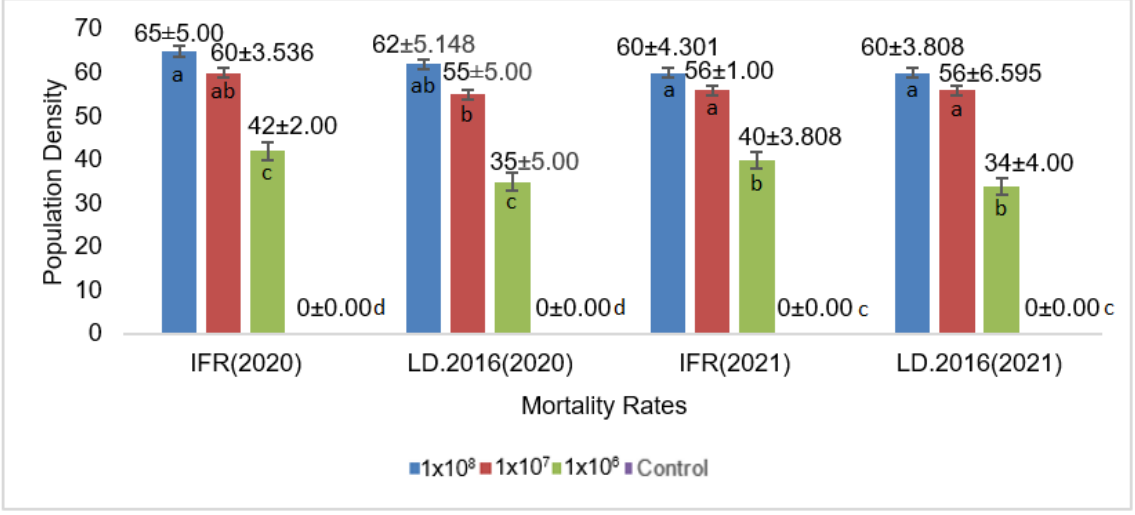


Figure 12. Mortality percentages (±std. deviation) on *Ceratitis capitata* of IFR and LD.2016 isolates in Antalya Province in 2020 and 2021.* Statistical analyses were made separately for 2020 and 2021. *According to Tukey's multiple comparison test, different letters are statistically different ($p<0.05$). (2020; F: 155,508; Between groups df:7; Within groups df: 32; p<000.1), (2021; F: 154,012; Between groups df:7; Within groups df: 32; p<000.1).

In this study conducted with *Beauveria bassiana* and *Cordyceps fumosorosea*, the change in dose-related mortality rates seems to be compatible with previous studies targeting different biological stages of *C. capitata* (Castrillo et al., 2000; Dimbi et al., 2003; Quesada-Moraga et al., 2006; Ekesi et al., 2010; Beris et al., 2013; Qazzaz et al., 2015; Chergui et al., 2020; Soliman et al., 2020; Hallouti et al., 2021). Studies targeting late-stage larvae of *C. capitata*, as in our study; In 2020, Cherqui et al. (2020) studied the *Beauveria bassiana* isolate with a dose of $1x10^7$ conidia/mL and reported the mortality rate as 44%. Gava et al. (2020) and Soliman et al. (2020) obtained similar results as our study in their study on late-stage larvae, which reported death at varying rates depending on the dose. The success of infection and the development of

fungal structures are affected by the pathogenicity of the disease, spore density, and the immune response of the host (Islam et al., 2021). For this reason, it is acceptable to obtain different results from other studies. Continuity of fungal populations is essential for sustainable management of pests. The formation of the fungal population is based on the transformation of resources within the host cadaver because infective spores spread from the cadaver (Meyling and Eilenberg, 2007). One reason why mortality rates are decreasing over time in our conidial persistence studies (Figures 8 & 9) can be attributed to the small size of *C. capitata* larvae and their inability to provide sufficient nutrients for a larger amount of spore production.

However, it is important to note that EPFs help minimize damage to crops (below the economic threshold) by triggering infection of pests, which ultimately leads to a reduction in feeding, oviposition, development, mating and other physiological characteristics of insects (Thomas et al., 1997). As seen in our study results, the negative effects on larval development and the fertility of individuals who emerged as adults after infection, as well as the shortening of the lifespan of adults, were parallel to some previous studies. Although not on the same biological period comparison to present study (Table 1), Quesada-Moraga et al. (2006) stated that the fertility of *C. capitata* adults decreased 6, 8, and 10 days after the applications. Castrillo et al. (2000) reported that fertility was reduced over 53% compared to control group. Another study was by Gava et al. (2020), who reported that EPF applications shortened adult lifespan. Although there was a difference in application, similar results were seen in our study (Figures 10 & 11). Although the delay or prolongation of a biological period of the target organism is a negative feature for the target organism, it is an advantage in terms of management.

In studies conducted with non-sterile field soil under semi-controlled conditions, it is seen that the mortality rates caused by IFR and LD.2016 isolate were close to each other in both years, unlike climate chamber studies (Figure 12). This may be caused by climatic conditions or the influence of other organisms in the soil environment. Additionally, the soil environment has a diverse microbiota with antagonistic fungi and bacteria that may inhibit EPF survival or reduce biocontrol activity (Jaronski, 2007).

In the light of all these results and evaluations, it is thought that the *C. fumosorosea* and *B. bassiana* isolates used in the experiment have a significant potential to be used at the soil level to apply to late-stage larvae in the management against *C. capitata*. For this reason, the application of fungal soil inoculations to the tree crown projection, including rhizosphere, in fruit cultivation may be a good strategy for both pest control and benefiting from its other contributions. In addition, it is necessary to continue detailed research to better understand the interactions of fungal species in the soil with the entomopathogenic fungi to be used and to define their effects on the reproductive capacity of the target organism for sustainable pest management strategies.

## Acknowledgement

This study was funded by the Ministry of Agriculture and Forestry of the Republic of Turkey, General Directorate of Agricultural Research and Policies, (with the project number TAGEM/BSAD/A/21/A2/P5/2404), and the manuscript was produced from a part of data from first author's PhD. dissertation. Petri dishes bioassay results and the first application results of conidial persistence studies ($0^{th}$ month on sterile soil bioassays), which form a part of this study, were presented as an oral presentation at the $8^{th}$ International Entomopathogens and Microbial Control Congress held in Antalya-Turkey on 6-8 October 2022, and the summary was included in the congress summary book.

## References

Abbott, W. S., 1925. A method of computing the effectiveness of an Insecticide. Journal of Economic Entomology, 18 (2): 265-267.

Baydar, R., Ö. Güven & İ. Karaca, 2016. Occurrence of entomopathogenic fungi in agricultural soils from Isparta Province in Turkey and their pathogenicity to *Galleria mellonella* (L.) (Lepidoptera: Pyralidae) larvae. Egyptian Journal of Biological Pest Control, 26 (2): 323-327.

Behie, S. W., S. J. Jones & M. J. Bidochka, 2015. Plant tissue localization of the endophytic insect pathogenic fungi *Metarhizium* and *Beauveria*, Fungal Ecology, 13 (1): 112-119.

Beris, I. E., D. P. Papachristos, A. Fytrou, S. A. Antonatos & D. C. Kontodimas, 2013. Pathogenicity of three entomopathogenic fungi on pupae and adults of the Mediterranean fruit fly, *Ceratitis capitata* (Diptera: Tephritidae). Journal of Pest Science, 86 (1): 275-284.

Bernardo, M. A. & M. S. Singer, 2017. Parasite-altered feeding behavior in insects: Integrating functional and mechanistic research frontiers. The Journal of Experimental Biology, 220 (16): 2848-2857.

Castrillo, M. A., P. Moya, E. Hernández & E. Primo-Yufera, 2000. Susceptibility of *Ceratitis capitata* Wiedemann (Diptera: Tephritidae) to entomopathogenic fungi and their extracts. Biological Control, 19 (3): 274-282.

Chergui, S., K. Boudjemaa, A. Benzehra & İ. Karaca, 2020. Pathogenicity of indigenous *Beauveria bassiana* (Balsamo) against *Ceratitis capitata* Wiedemann (Diptera: Tephritidae) under laboratory conditions. Egyptian Journal of Biological Pest Control, 30 (1): 1-7.

Dash, C. K., B. S. Bamisile, R. Keppanan, M. Qasim, Y. Lin, S. U. Islam, M. Hussain & L. Wang, 2018. Endophytic entomopathogenic fungi enhance the growth of *Phaseolus vulgaris* L. (Fabaceae) and negatively affect the development and reproduction of *Tetranychus urticae* Koch (Acari: tetranychidae), Microbial Pathogenesis, 125 (1): 385-392.

Dias, N. P., M. J. Zotti, P. Montoya, I. R. Carvalho & D. E. Nava, 2018. Fruit fly management research: A systematic review of monitoring and control tactics in the world. Crop Protection, 112 (1): 187-200.

Dimbi, S., N. K. Maniania & S. Ekesi, 2009. Effect of *Metarhizium anisopliae* inoculation on the mating behavior of three species of African Tephritid fruit flies, *Ceratitis capitata*, *Ceratitis cosyra* and *Ceratitis fasciventris*. Biological Control, 50 (2): 111-116.

Dimbi, S., N. K., Maniania, S. A. Lux, S. Ekesi & J. K. Mueke, 2003. Pathogenicity of *Metarhizium anisopliae* (Metsch.) Sorokin and *Beauveria bassiana* (Balsamo) Vuillemin, to three adult fruit fly species: *Ceratitis capitata* (Weidemann), *C. rosa* var. fasciventris Karsch and *C. cosyra* (Walker) (Diptera: Tephritidae). Mycopathologia, 156 (4): 375-382.

Ding, J. L., H. Y. Lin, J. Hou, M. G. Feng & S. H. Ying, 2023. The entomopathogenic fungus *Beauveria bassiana* employs autophagy as a persistence and recovery mechanism during conidial dormancy. Mbio, 14 (2): e03049-22.

Ekesi, S., K. Maniania & S. A. Lux, 2010. Mortality in three African Tephritid fruit fly puparia and adults caused by the entomopathogenic fungi, *Metarhizium anisopliae* and *Beauveria bassiana*. Biocontrol Science and Technology, 12 (1): 7-17.

Feng, M. G., T. J. Poprawski & G. G. Khachatourians, 1994. Production, formulation and application of the entomopathogenic fungus *Beauveria bassiana* for insect control: current status, Biocontrol Science and Technology, 4 (1): 3-34.

Gava, C. A. T., P. F. D. S. Tavares, J. S. Gonçalves & B. A. J. Paranhos, 2020. Applying local entomopathogenic fungi strains to the soil can control *Ceratitis capitata* (Diptera: Tephritidae) Wiedemann adults. Biocontrol Science and Technology, 30 (2): 103-115.

Hallouti, A., H. Benjlil, A. El Hamdaoui, R. A. Hammou, M. A. Hamza, A. Zahidi, A. A. B. Aoumar & H. Boubaker, 2021. Evaluation of native entomopathogenic fungi isolates for microbial control of the Mediterranean fruit fly (*Ceratitis capitata* (Diptera: Tephritidae)) pupae and adults. Sumerianz Journal of Biotechnology, 4 (4): 153-164.

Hussain, A., M. Y. Tian, Y. R. He & S. Ahmed, 2009. Entomopathogenic fungi disturbed the larval growth and feeding performance of *Ocinara varians* (Lepidoptera: Bombycidae) larvae. Insect Science, 16 (6): 511-517.

IBM SPSS, 2015. Statistics for Windows, 2015 Version 23.0. Armonk, NY: IBM Corporation.

Islam, W., M. Adnan, A. Shabbir, H. Naveed, Y. S. Abubakar, M. Qasim, M. Tayyab, A. Noman, M. S. Nisar, K. A. Khan & H. Ali, 2021. Insect-fungal-interactions: A detailed review on entomopathogenic fungi pathogenicity to combat insect pests. Microbial Pathogenesis, 159 (1): 105-122.

Jackson, T. A., S. B. Alves & R. M. Pereira, 2000. "Success in Biological Control of Soil-Dwelling İnsects by Pathogens and Nematodes, 271-296". In: Biological Control: Measures of Success (Eds. G. Gurr & S. Wratten). Kluver Academics Publishers: New York, USA, 429 pp.

Jaronski, S. T., 2007. "Soil Ecology of The Entomopathogenic Ascomycetes: A Critical Examination of What We (think) We Know, 1-53". In: Use of Entomopathogenic Fungi in Biological Pest Management (Eds. S. Ekesi & N. K. Maniania). Research Signpost: Kerala, 321 pp.

Keller, S. & G. Zimmerman, 1989. "Mycopathogens of Soil Insects, 240-270". In: Insect-Fungus Interactions (Eds. N. Wilding, N. M. Collins, P. M. Hammond & J. F. Webber). Academic Press, London, 343 pp.

Lu, H. L. & R. J. St Leger, 2016. Insect immunity to entomopathogenic fungi. Advances in Genetics, 94 (1): 251-285.

Meyling, N. V. & J. Eilenberg, 2007. Ecology of the entomopathogenic fungi *Beauveria bassiana* and *Metarhizium anisopliae* in temperate agroecosystems: Potential for conservation biological control. Biological Control, 43 (2): 145-155.

O'Callaghan, M., R. A. Ballard & D. Wright, 2022. Soil microbial inoculants for sustainable agriculture: Limitations and opportunities. Soil Use and Management, 38 (3): 1340-1369.

Ortiz-Urquiza, A. & N. O. Keyhani, 2013. Action on the surface: entomopathogenic fungi versus the insect cuticle. Insects, 4 (3): 357-374.

Qazzaz, F. O., M. I. Al-Masri & R. M. Barakat, 2015. Effectiveness of *Beauveria bassiana* Native Isolates in the Biological Control of the Mediterranean Fruit Fly (*Ceratitis capitata*). Advances in Entomology, 3 (2): 44-55.

Quesada-Moraga, E., A. Ruiz-García & C. Santiago-Álvarez, 2006. Laboratory evaluation of entomopathogenic fungi *Beauveria bassiana* and *Metarhizium anisopliae* against puparia and adults of *Ceratitis capitata* (Diptera: Tephritidae). Journal of Economic Entomology, 99 (6): 1955-1966.

Quesada-Moraga, E., I. Garrido-Jurado, M. Yousef-Yousef & N. González-Mas, 2022. Multitrophic interactions of entomopathogenic fungi in BioControl. BioControl, 67 (5): 457-472.

Rehner, S. A. & E. Buckley, 2005. A *Beauveria* phylogeny inferred from nuclear ITS and EF1-α sequences: evidence for cryptic diversification and links to *Cordyceps* teleomorphs. Mycologia, 97 (1): 84-98.

Roberts, D. W. & R. J. St. Leger, 2004. *Metarhizium* spp., cosmopolitan insect-pathogenic fungi: Mycological aspects. Advances in Applied Microbiology, 54 (1): 1-70.

Samson, R. A., H. C. Evans & J. P. Latge, 1988. Atlas of Entomopathogenic Fungi. Springer-Verlag: New York, USA, 187 pp.

Scholte, E. J., B. G. J. Knols, R. A. Samson & W. Takken, 2004. Entomopathogenic fungi for mosquito control: a review, Journal of Insect Science, 4 (1): 1-24.

Sevinç, M. S. & İ. Karaca, 2024. *Ceratitis capitata* (Wiedemann) (Diptera: Tephritidae) popülasyonunda ana yaşının biyolojik parametrelere etkisi. Meyve Bilimi, 11 (1): 35-42 (in Turkish with abstract in English).

Shah, P. A. & J. K. Pell, 2003. Entomopathogenic fungi as biological control agents, Applied Microbiology and Biotechnology, 61 (5): 413-423.

Sheldrake, M., 2020. Saklı Dünya (Turkish version of "Entangled Life: How Fungi Make Our Worlds, Change Our Minds & Shape Our Futuresbook"), First Edition, August 2022, Domingo: İstanbul, 368 pp (in Turkish).

Soliman, N. A., S. M. Al-amin, A. E. Mesbah, A. Ibrahim & A. Mahmoud, 2020. Pathogenicity of three entomopathogenic fungi against the Mediterranean fruit fly, *Ceratitis capitata* (Wiedemann) (Diptera: Tephritidae). Egyptian Journal of Biological Pest Control, 30 (1): 1-8.

Solter, L. F., A. E. Hajek & L. A. Lacey, 2017. "Exploration for Entomopathogens, 13-23". In: Microbial Control of Insect and Mite Pests (Eds. L. A. Lacey). Academic Press: London, 482 pp.

Thomas, M. B., S. Blanford & C. Lomer, 1997. Reduction of feeding by the variegated grasshopper, *Zonocerus variegatus*, following infection by the fungal pathogen, *Metarhizium flavoviride*. Biocontrol Science and Technology, 7 (3): 327-334.

Tscharntke, T. & B. A. Hawking, 2000. “Multitrophic Level Interactions: An Introduction, 1-7”. In: Multitrophic Level Interactions (Eds. T. Tscharntke & B. A. Hawking). Cambridge University Press: Cambridge. 284 pp.

Tukey, J. W., 1949. Comparing individual means in the analysis of variance. Biometrics, 5 (2): 99-114.

Ullah, M. S. & U. T. Lim, 2017. Laboratory evaluation of the effect of *Beauveria bassiana* on the predatory mite *Phytoseiulus persimilis* (Acari: Phytoseiidae). Journal of Invertebrate Pathology, 148 (1):102-109.

Vega, F. E., M. S. Goettel, M. Blackwell, D. Chandler, M. A. Jackson, S. Keller, M. Koike, N. K. Maniania, A. Monz´on, B. H. Ownley, J. K. Pell, D. E. N. Rangel & H. E. Roy, 2009. Fungal entomopathogens: new insights on their ecology. Fungal Ecology, 2 (4): 149-159.

Vidal, S & L. R. Jaber, 2015. Entomopathogenic fungi as endophytes: plant-endophyteherbivore interactions and prospects for use in biological control. Current Science, 109 (1): 46-54.

Villaseñor, A., S. Flores, S. E. Campos, J. Toledo, P. Montoya, P. Liedo & W. Enkerlin, 2019. Use of entomopathogenic fungi for fruit fly control in area-wide SIT programmes. (Website: https://www.iaea.org/sites/default/files/21/05/2019_ipc_use_of_entomopathogenic_fungi_eng.pdf), (Access date: 25.09.2024).

Yeşilırmak, M., M. S. Sevinç, & B. Yaman, 2024. Elma bahçelerinde elma içkurdu ve kırmızı örümceklere karşı kullanılan bazı insektisit ve akarisitlerin Akdeniz meyve sineği, *Ceratitis capitata* (Diptera: Tephritidae) üzerinde etkileri. ANADOLU Ege Tarımsal Araştırma Enstitüsü Dergisi, 34 (1): 33-37 (in Turkish with abstract in English).

Zimmermann, G., 2007. Review on safety of the entomopathogenic fungi *Beauveria bassiana* and *Beauveria brongniartii*. Biocontrol Science and Technology, 17 (6): 553-596.

Zimmermann, G., 2008. The entomopathogenic fungi *Isaria farinosa* (formerly *Paecilomyces farinosus*) and the *Isaria fumosorosea* species complex (formerly *Paecilomyces fumosoroseus*): Biology, ecology and use in biological control. Biocontrol Science and Technology, 18 (9): 865-901.